Interventions for patients with primary biliary cirrhosis: systematic reviews and meta-analyses of randomised clinical trials

Ph.D. Thesis

Yan Gong

Faculty of Health Sciences
University of Copenhagen
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Yan Gong

Copenhagen Trial Unit

Centre for Clinical Intervention Research

Rigshospitalet

Copenhagen University Hospital

Supervisors: Christian Gluud, Erik Christensen, Jiyao Wang

Examiners: Rolf Olsson, Troels Haveland, Lene Theil Skovgaard

". . . (A) long habit of not thinking a thing wrong, gives it a superficial appearance of being right, and raises at first a formidable outcry in defence of custom. But the tumult soon subsides. Time makes more converts than reason."

— Thomas Paine, Common Sense, 1776

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Preface/Acknowledgement

The present Ph.D. study has been conducted during my employment at the Copenhagen Trial Unit, Centre for Clinical Intervention Research, Rigshospitalet, Copenhagen University Hospital during the period September 2002 – June 2006. I should like to express my great gratitude to Christian Gluud, my main supervisor, who provided me with time, facilities, encouragement, and input during the whole period. I should also like to thank my second supervisor Erik Christensen for valuable advice on the reviews. My third supervisor Jiyao Wang is thanked for stimulating talks and assistance to access to the Chinese Biomedical Database and talk with patients with primary biliary cirrhosis. My co-authors Sarah Louise Klingenberg and Zhibi Huang as well as my other co-authors are thanked for their hard work on data extraction, performing electronic literature searches, and valuable contribution to the reviews.

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Finally, I owe the greatest thanks to my parents and my sister. Thank you all – and in particular thank you to my friend Huaifen Liao for her devotion and encouragement.

Original papers

This Ph.D. thesis is based on the following papers:

1. A) Gong Y, Huang ZB, Christensen E, Gluud C. Ursodeoxycholic acid for patients with primary biliary cirrhosis. *Cochrane Database of Systematic Reviews* 2007 (submitted).
   B) Gong Y, Huang ZB, Christensen E, Gluud C. Ursodeoxycholic acid for primary biliary cirrhosis: an updated systematic review and meta-analysis of randomised clinical trials using Bayesian approach as sensitivity analyses. American Journal of Gastroenterology 2007;102(8):1799-1807.

2. A) Gong Y, Frederiksen SL, Gluud C. D-penicillamine for primary biliary cirrhosis. *Cochrane Database of Systematic Reviews* 2004, Issue 4. Art. No.: CD004789. DOI: 10.1002/14651858.CD004789.pub2.
   B) Gong Y, Klingenberg SL, Gluud C. D-penicillamine vs. placebo/no intervention in patients with primary biliary cirrhosis: Cochrane Hepato-Biliary Group systematic review with meta-analyses of randomised clinical trials. Alimentary Pharmacology and Therapeutics 2006;24:1535-1544.

3. A) Gong Y, Gluud C. Colchicine for primary biliary cirrhosis. *Cochrane Database of Systematic Reviews* 2004, Issue 2. Art. No.: CD004481. DOI: 10.1002/14651858.CD004481.pub2.
   B) Gong Y, Gluud C. Colchicine for primary biliary cirrhosis: a systematic review of randomised clinical trials. American Journal of Gastroenterology 2005;100:1876-1885.

4. Gong Y, Gluud C. Methotrexate for primary biliary cirrhosis. *Cochrane Database of Systematic Reviews* 2005, Issue 3. Art. No.: CD004385. DOI: 10.1002/14651858.CD004385.pub2.

5. Gong Y, Christensen E, Gluud C. Azathioprine for primary biliary cirrhosis. *Cochrane Database of Systematic Reviews* 2007, Issue 3. Art. No.: CD006000. DOI: 10.1002/14651858.CD006000.pub2.

6. Gong Y, Christensen E, Gluud C. Cyclosporin A for primary biliary cirrhosis. *Cochrane Database of Systematic Reviews* 2007, Issue 3. Art. No.: CD005526. DOI: 10.1002/14651858.CD005526.pub2.

Abstract

Objectives: To assess the effects of ursodeoxycholic acid (UDCA), d-penicillamine, colchicine, methotrexate, azathioprine, and cyclosporin A in patients with primary biliary cirrhosis (PBC).

Methods: We performed six systematic reviews of relevant randomised clinical trials. Trials were identified mainly through The Cochrane Hepato-Biliary Group Controlled Trials Register, The Cochrane Library, MEDLINE, and EMBASE. We applied meta-analyses, where appropriate, to determine intervention effects on mortality, mortality or liver transplantation, clinical symptoms, liver biochemistry, liver histology, and adverse events.

Results: Six systematic reviews include a total of 42 trials with 4009 patients with PBC. Two thirds of the trials had low methodological quality regarding generation of the allocation sequence, allocation concealment, blinding, and follow-up. The meta-analyses did not show significant benefits of UDCA, d-penicillamine, colchicine, methotrexate, azathioprine, and cyclosporin A on survival of patients with PBC. UDCA improved biochemical variables and clinical symptoms such as ascites and jaundice, but it was associated with adverse events, mainly weight gain. D-penicillamine had no significant beneficial clinical effects, but significantly increased adverse events. Colchicine may improve pruritus, but it tended to lead to more adverse events (mostly transient diarrhoea), although it is not statistically significant. Methotrexate may improve pruritus and decrease the levels of serum alkaline phosphatases and plasma immunoglobulin M, but the hepatotoxicity could not be ruled out. Patients given azathioprine experienced more adverse events than patients given no intervention or placebo, such as rash, severe diarrhoea and bone marrow depression. Cyclosporin A might improve pruritus, reduce alanine aminotransferase, and increase serum albumin level. But cyclosporin A caused more adverse events, including renal dysfunction and hypertension.

Conclusions: We did not find reliable evidence to support the clinical use of the assessed interventions in patients with PBC. A large proportion of the trials is flawed by low methodological quality, small number of patients, and short trial duration. None of the interventions can be recommended for general use in clinical practice.

Dansk resumé

Formål Formålet var at vurdere effekten af ursodeoxykolsyre, d-penicillamin, kolkicin, methotrexat, azathioprin, og ciclosporin A hos patienter med primær biliær cirrose (PBC).

Materialer og metoder Vi foretog seks systematiske bedømmelser af relevante randomiserede kliniske forsøg. Forsøgene blev hovedsageligt identificeret i The Cochrane Hepato-Biliary Group Controlled Trials Register, The Cochrane Library, MEDLINE, og EMBASE. Vi anvendte meta-analyser hvor det var hensigtsmæssigt for at vurdere interventionseffekten på dødelighed, dødelighed eller lever transplantation, kliniske symptomer, lever biokemi, lever histologi, og utilsigtede hændelser.

Resultater De seks systematiske bedømmelser inkluderede i alt 42 forsøg med 4.009 PBC patienter. To tredjedele af forsøgene havde lav metodisk kvalitet hvad angår generering af allokerings sekvens, skjult allokering, blinding, og patient opfølgning. Meta-analyserne viste ingen signifikant gavnlig effekt af ursodeoxykolsyre, d-penicillamin, kolkicin, methotrexat, azathioprin, og ciclosporin A på dødeligheden af PBC patienter. Ursodeoxykolsyre forbedrede biokemiske variable og kliniske symptomer som bugvattersot og gulsot, men var også forbundet med utilsigtede hændelser, hovedsageligt vægtøgning. D-penicillamin havde ingen gavnlige kliniske effekter, og øgede signifikant utilsigtede hændelser. Kolkicin kan muligvis forbedre hudkløe, men kunne øge utilsigtede virkninger (hovedsageligt forbigående diare), selvom det ikke var statistisk signifikant. Methotrexat kan muligvis forbedre hudkløe og sænke aktiviteten af serum basiske fosfataser og plasma immunglobulin M, men lever toksicitet kan ikke udelukkes. Patients som fik azathioprin udviklede flere utilsigtede hændelser end patenter som ikke fik nogen intervention eller som fik placebo, fx udslet, svær diare, og knoglemarvsdepression. Ciclosporin A kan muligvis forbedre hudkløe, sænke alanin aminotransferase aktiviteten, og øge serum albumin koncentrationen. Men ciclosporin A forårsage de flere utilsigtede hændelser, omfattende nyresvigt og forhøjet blodtryk.

Diskussion

Vi fandt ikke troværdig evidens der understøtter den kliniske brug af de bedømte interventioner til PBC patienter. En stor del af forsøgene er belastede af lav metodisk kvalitet, få inkluderede patienter, og kort varighed. Ingen af interventionerne kan anbefales til generel brug i klinisk praksis.

Introduction

Primary biliary cirrhosis (PBC) is a slowly progressive autoimmune liver disease of unknown aetiology that is characterised by destruction of the intra-hepatic bile ducts.[1] The loss of bile ducts leads to decreased bile secretion and the retention of toxic substances within the liver. This leads to necrosis and inflammation and eventually to liver cirrhosis and liver failure over a period of time that can vary widely among patients.[2] Evidence to date suggests that immunological and genetic factors might play a role in disease progression. PBC primarily affects middle-aged women with asymptomatic rises of serum hepatic biochemical variables. Fatigue, pruritus, or unexplained hyperlipidaemia at initial presentation may suggest a diagnosis of PBC. Serum antimitochondrial antibody positivity is almost diagnostic of the disease.[3]

A number of drugs have been evaluated for PBC patients, especially ursodeoxycholic acid (UDCA),[4] d-penicillamine,[5-8] colchicine,[9-11] methotrexate,[12;13] azathioprine,[14;15] cyclosporin A,[16] chlorambucil,[17] glucocorticosteroids,[18] malotilate,[19] and thalidomide.[20]

Despite of numerous treatment options, PBC is now a frequent cause of liver morbidity, and the patients are significant users of health resources, including liver transplantation.[21]

Epidemiology

PBC was first comprehensively described around 1950.[22;23] During the last 10-20 years, substantial increases in the prevalence of PBC have been observed.[24] Estimates of annual incidence range from 2 to 24 patients per million population and estimates of prevalence ranges from 19 to 240 patients per million population.[21] The disease affects all races, yet seems to cluster within specific geographical areas. It is most prevalent in northern Europe.[21]

PBC is considerably more common in first-degree relatives of patients than in unrelated persons. First-degree relatives of people with PBC are

also known to have at least a two-fold increased risk of autoimmune disease.[25]

Pathogenesis

Present evidence supports to the notion of PBC as an immune-mediated disease. Cellular and humoral abnormalities have both been noted. The major finding associated with humoral immunity in PBC resides with recognition of the antimitochondrial antibody. Formation of this antibody is presented in more than 95% of patients. Although the mechanism of biliary destruction remains enigmatic, the specificity of pathological changes in the bile ducts, the presence of lymphoid infiltration in the portal tracts, and the presence of major-histocompatibility-complex class II antigen on the biliary epithelium suggest that an intense autoimmune response is directed against the biliary epithelial cells.[26]

The world-wide variation in disease prevalence suggests that environmental factors likely play a role in causes of the disease, including bacteria, viruses, and chemicals. Bacteria have attracted the most attention because of the reported elevated incidence of urinary tract infections in patients with PBC. Other potential causes include exposure to environmental chemicals. However, it is unclear whether the chemical immunisation is serendipitous and capable of eliciting antimitochondrial antibodies or whether these antibodies are capable of inducing PBC.[26]

Clinical findings

Individuals with asymptomatic disease consist of 20-60% of all first-time diagnoses. However, most asymptomatic patients, over time, will develop symptoms and hepatic disease will progress.

Fatigue and pruritus are the most common presenting symptoms. Other findings include hyperlipidaemia, hypothyroidism, osteopenia, and

coexisting autoimmune diseases, including Sjögren's syndrome and scleroderma.[3]

Natural history and prognosis

From time of diagnosis, asymptomatic patients have a greater overall median survival than do symptomatic individuals. Those remaining asymptomatic patients have about equivalent survival rates compared with an age-matched and sex-matched healthy population.[27] Estimates of overall median survival for symptomatic patients range between 10 and 15 years from time of diagnosis, whereas patients with advanced histological disease (stage 3 or 4) have a median survival approaching eight years.[1] The Mayo Clinic model is most frequently used for predicting long-term survival. The patient’s age, serum total bilirubin, albumin, prothrombin time, and presence or absence of oedema and ascites are the model’s independent predictor variables.[28]

Diagnosis

The diagnosis of PBC is currently based on the triad: the presence of detectable antimitochondrial antibodies in serum; elevation of liver enzymes (most commonly alkaline phosphatases) for more than six months; and characteristic liver histological changes in the absence of extrahepatic biliary obstruction.[3]

Interventions

A number of drugs have been evaluated for PBC as indicated above in order to affect the liver disease per se. Therapies have aimed to reduce tissue damage by toxic bile salts following bile duct destruction and immuno-inflammatory reactions.

Ursodeoxycholic acid

Bile duct destruction leads to the retention of hydrophobic bile acids within the liver. This likely contributes to the gradual deterioration in liver function observed in patients with PBC. Ursodeoxycholic acid (UDCA), the epimer of chenodeoxycholic acid, increases the rate of

transport of intracellular bile acids across the liver cell and into the canaliculus in patients with PBC.[29] UDCA treatment reduces intracellular hydrophobic bile acid levels and thereby may have a cytoprotective effect on cell membranes. UDCA is the only drug approved for PBC by the Food and Drug Administration. Dosage of 13 to 15 mg UDCA/kg/day is recommended for obtaining significant improvements in liver biochemistry and immunoglobulin levels and reduces titres of antimitochondrial antibodies.[30;31] However, the effect of UDCA on mortality and histological progression remains controversial.[4;32] Since 2001, several randomised clinical trials have been published with the results of longer-term follow-up on patients' survival.[33;34] We therefore re-evaluated the effects of UDCA in patients with PBC using updated data and new statistical analyses.[35]

Immunosuppressants

As PBC is considered an autoimmune disorder, another logical approach to therapy could employ immunosuppressants, e.g., d-penicillamine, colchicine, methotrexate, azathioprine, cyclosporin A, or glucocorticosteriods. The immunosuppressants have shown some benefit in clinical trials, but the results of trials are conflicting.

D-penicillamine

D-penicillamine is a cupruretic drug known for its efficacy in treating Wilson's disease.[36;37] D-penicillamine has antifibrogenic effects, ability to decrease circulating immune complexes, and inhibitory effect on lymphocyte function.[4;38;39] Furthermore, d-penicillamine is able to lead to cupruresis. PBC patients have increased hepatic copper levels. Early reports showed that d-penicillamine was a promising drug, improving survival in patients with PBC and having relatively few side-effects.[5;40;41] Several later studies showed that d-penicillamine did decrease hepatic levels of copper, but it did not have a beneficial effect on symptoms related to PBC, hepatic biochemistry, histological progression, or survival. In addition, d-penicillamine was associated with up to a 46% incidence of major toxic events, most commonly

proteinuria, allergic drug reaction, and more rarely bone marrow depression.[7;8;42;43]

Colchicine

Colchicine is a plant alkaloid. It is effective against gouty arthritis and other forms of rheumatic diseases (rheumatoid arthritis, familial Mediterranean fever, Bechet's disease, etc.).[44] The basis for effect of colchicine is inhibition of the migration of granulocytes into inflamed areas and decreased metabolic and phagocytic activity of granulocytes. Furthermore, colchicine is an anti-mitotic[45] and anti-fibrotic agent. Colchicine retards the microtubule mediated transport of procollagen[46] and enhances collagenase activity.[47] Colchicine has been used for PBC patients because of its immunomodulatory and anti-fibrotic potential. Colchicine has been reported to slow the rate of progression of PBC[48] and to produce improvements in liver function tests and immunoglobulin levels.[9;10] However, colchicine does not affect clinical symptoms or liver histology.[48] The effect of combination therapy with colchicine and UDCA in patients with PBC has been reported, but the results have been conflicting.[49-52]

Methotrexate

Methotrexate is a folic acid antagonist that blocks nucleic acid synthesis. Additionally, folic acid antagonists are potent inhibitors of cell-mediated (T and B cells) immune reactions and have been employed as immunosuppressive agents, for example, in allogeneic bone marrow and organ transplantation, and for the treatment of dermatomyositis, rheumatoid arthritis, Wegener's granulomatosis, and Crohn's disease.[53] Low-dose methotrexate has immunosuppressive properties that may be mediated through inhibition of human interleukin-1 beta-induced leukocyte proliferation.[54] Based on small pilot studies,[55;56] methotrexate was initially suggested as monotherapy for PBC since the degree of hepatic inflammation and bile duct injury improved in some patients. The degree of liver fibrosis and histological stage, however, were not improved.[55;56] The first placebo-controlled trial of methotrexate for PBC did not support the clinical use of low-dose methotrexate.[57] The addition

of methotrexate did not seem to confer additional benefit in patients receiving UDCA.[58-60]

Azathioprine

Azathioprine is an immunosuppressant, suppressing delayed hypersensitivity and cellular cytotoxicity more than antibody responses. The immunosuppressive action of azathioprine depends on its conversion to active 6-mercaptopurine by thiopurine S-methyl-transferase.[61] Azathioprine is used for Crohn's disease,[62] renal homotransplantation,[63] and severe, active rheumatoid arthritis[64] in PBC showed no efficacy and suggested the possibility of significant toxicity of azathioprine therapy.[65] In contrast, a large multicentre trial showed evidence of efficacy with very little toxicity.[15]

Cyclosporin A

Cyclosporin A has proved effective in preventing immune-mediated rejection of a variety of transplanted human allografts[66] and has been shown to produce clinical improvement in a number of autoimmune conditions.[67] Cyclosporin A is a cyclic endecapeptide of fungal origin. It alters lymphokine production so that the T-helper-inducer subpopulations are attenuated, T-cell help required for B-cell activation is blocked, cytotoxic T-cell generation is attenuated, and T-suppressor cell subpopulations are expanded.[47] Thus cyclosporin A would appear a potential ideal agent to modify the immunologic irregularities in PBC.[68] Since 1980 when Routhier showed beneficial effects of cyclosporin A on serum aspartate transaminase and alkaline phosphatases in six patients with PBC,[69] several randomised clinical trials have been carried out with different results.[70;71]

Other interventions

Chlorambucil

The alkylating agent chlorambucil (0.5-4 mg/day) was shown to have rather marked beneficial effects on biochemistry and histology in a small randomised clinical trial including 24 patients, but 4 of 13 (31%) on chlorambucil were withdrawn because of adverse effects.[72]

Malotilate

Malotilate (1.5 g/day) has been evaluated versus placebo in a double-blind multicentre randomised clinical trial including 101 patients.[19] After a mean follow-up of 28 months significant beneficial effects were found on liver enzymes, immunoglobulin G and M, liver necrosis and inflammatory cell infiltration, but not on fibrosis, pruritus, disease progression, or survival. The observed benefits appeared too slight to recommend the drug as therapy.

Thalidomide

Thalidomide 100 mg/day has been tested against placebo in a small double-blind trial involving 18 patients. Except for a possible effect on pruritus no significant effects of the drug were found, and adverse effects occurred in 40%.[20]

Glucocorticosteroids

Only two small randomised clinical trials on this topic were identified.[18;73] Glucocorticosteroids were associated with improvement in serum markers of inflammation and liver histology, both of which were of uncertain clinical significance. Glucocorticosteroids were also associated with adverse events, including reduced bone mineral density.

The above mentioned ’other interventions’ have not been planned into the scope of the thesis, either because too few trials have been performed on the interventions or other authors have already done the work of systematically reviewing the literature, i.e., glucocorticosteriods.[74] Therefore, they will not be mentioned further in this thesis.

Objectives

The objectives were to assess the beneficial and harmful effects of the following interventions for patients with PBC by performing systematic reviews and meta-analyses, if appropriate, on:

1. UDCA
2. D-penicillamine
3. Colchicine
4. Methotrexate
5. Azathioprine
6. Cyclosporin A.

Methods

All reviews were performed according to published protocols following the recommendations given by the Cochrane Handbook for Systematic Reviews of Interventions[75] and the QUOROM Statement (www.consort-statement.org/QUOROM.pdf).

Searching

We searched for randomised trials in *The Cochrane Hepato-Biliary Group Controlled Trials Register, The Cochrane Central Register of Controlled Trials* in *The Cochrane Library, MEDLINE, EMBASE, Science Citation Index-Expanded, The Chinese Biomedical CD Database, LILACS*, and in references of identified studies. We screened bibliographies of relevant articles and conference proceedings and wrote to trialists and pharmaceutical companies producing the drugs in question.

Trial selection

We only included randomised clinical trials comparing the interventions as mentioned below.

1. UDCA versus placebo or no intervention.
2. D-penicillamine versus placebo or no intervention.
3. Colchicine versus placebo or no intervention.

4. Methotrexate versus placebo or no intervention; methotrexate versus colchicine.
5. Azathioprine versus placebo or no intervention.
6. Cyclosporin A versus placebo or no intervention.

Inclusion was regardless of publication status, language, or blinding status.

At least two authors independently evaluated whether identified trials fulfilled the inclusion criteria. Disagreements were resolved by discussion among all the authors involved.

Trial quality assessment

We assessed the methodological quality of the randomised clinical trials using four components[76-78] as follows. Trials with low risk of bias were the ones meeting the adequacy criteria of the first three components.

Generation of the allocation sequence

- Adequate, if the allocation sequence was generated by a computer or random number table. Drawing of lots, tossing of a coin, shuffling of cards, or throwing dice are considered as adequate if a person who was not otherwise involved in the recruitment of participants performed the procedure;
- Unclear, if the trial was described as randomised, but the method used for the allocation sequence generation was not described.
- Inadequate, if a system involving dates, names, or admittance numbers were used for the allocation of patients. These trials are known as quasi-randomised and were excluded from the present reviews.

Allocation concealment

- Adequate, if the allocation of patients involved a central independent unit, on-site locked computer, numbered drug bottles or containers

with identical appearance prepared by an independent pharmacist or investigator, or sealed envelopes;

- Unclear, if the trial was described as randomised, but the method used to conceal the allocation was not described;
- Inadequate, if the allocation sequence was known to the investigators who assigned participants.

Blinding (or masking)

- Adequate, if the trial was described as double blind and the method of blinding involved identical placebo or active drug;
- Unclear, if the trial was described as double blind, but the method of blinding was not described;
- Not performed, if the trial was not double blind.

Follow-up

- Adequate, if the numbers and reasons for dropouts and withdrawals in all intervention groups were described or if it was specified that there were no dropouts or withdrawals;
- Unclear, if the report gave the impression that there had been no dropouts or withdrawals, but this was not specifically stated;
- Inadequate, if the number or reasons for dropouts and withdrawals were not described.

Data extraction

The primary outcome measures were mortality and mortality or liver transplantation. Secondary outcome measures included: pruritus; fatigue; liver complications, liver biochemistry; liver biopsy; quality of life; adverse events (excluding mortality and liver transplantation); and cost-effectiveness indicators.

Baseline data were recorded at trial level: mean (or median) age, sex ratio, histological stage, serum (s)-bilirubin concentration, intervention doses, and any co-interventions.

Statistical methods

We performed meta-analyses with Review Manager 4.2 (www.cochrane.dk). We analysed data by random-effects[79] and fixed-effect models.[80] We presented binary outcome measure as relative risk (RR) with 95% confidence interval (CI), and continuous outcome measure as weighted mean difference (WMD) with 95% CI. Heterogeneity was explored by chi-squared test with significance set at $P < 0.10$. The degree of heterogeneity was measured by I^2 [81] and, in UDCA review, between-trial variance was also estimated by the method of moments.[79] The larger the I^2 and moment-based between-trial variance, the larger degree of heterogeneity is present.

In the UDCA review, we performed a meta-regression analysis with STATA (Intercooled STATA 8.0, Texas, USA), which examined the effect size of UDCA in relation to the risk of bias, UDCA dosage, trial duration (treatment and follow-up), and severity of PBC at entry. Due to paucity of trials such analyses were not conducted in the other reviews.

In the UDCA review, we conducted the following sensitivity analyses to investigate the robustness of our main analyses on primary outcomes: (a) The influence of missing data: the missing data could be due to patient dropouts or lost to follow-up. We used an uncertainty method to allow for missing data.[82] The uncertainty method was developed for incorporating uncertainty, with weights assigned to trials based on uncertainty interval widths. The uncertainty interval for a trial incorporates both sampling error and the potential impact of missing data. (b) Bayesian meta-analytic approaches with WINBUGS (version 1.4.1), in which Markov chain Monte Carlo with Gibbs sampling was applied. This approach is able to account for uncertainty of all relevant sources of variability in the random-effects model. The analogue of a classical estimate is the marginal posterior median and the analogue of a classical confidence interval is the credibility interval (CrI).[83] We used odds ratio (OR) as summary statistic. For the ease of comparison, we reported the Bayesian results together with results from the classical meta-analysis presented as OR. (c) Bayesian meta-regression to estimate the UDCA effects adjusted for underlying

risk. The underlying risk is a convenient and clinically relevant trial-level measure, which can be interpreted as a summary of a number of unmeasured patient characteristics.[84] We use this approach to investigate the relationship between one specific covariate (e.g., UDCA dosage, trial duration, or disease severity of patients at entry) and the effects of UDCA adjusted for the underlying risk.

In the d-penicillamine and colchicine reviews, we also used the uncertainty method to pool the data on primary outcomes in order to allow for missing data due to dropouts as sensitivity analyses.[82] We performed subgroup analyses,[85] in which trials were grouped according to the risk of bias, dosage of experimental intervention, and duration of treatment and follow-up.

We explored publication bias and other biases according to Begg's and Egger's methods[86] with STATA[87] in the UDCA review. We did not perform the tests in the other reviews due to low number of trials included, as the power of those analyses would have been low.

Results

UDCA versus placebo or no intervention

Description of included trials

Figure 1 summarises the literature search. Sixteen trials met the selection criteria and were included. One of the trials provided no extractable data. In the follow-up period, seven trials continued UDCA treated patients on open-label UDCA (UDCA→UDCA) and offered open-label UDCA to all or some patients originally given placebo (placebo→UDCA).[30;33;34;88-91] Compared to the first publication of this systematic review in 2001,[3] we updated the data on mortality and liver transplantation from three trials.[33;34;89] and on adverse events from one trial.[89]

UDCA dose varied from 7.7 to 15.5 mg/kg/day with a median of 10 mg/kg/day. The duration of the trials varied from 3 to 92 months with a median of 24 months. The percentage of patients with advanced PBC or presenting symptoms at entry varied from 15% to 83% with a median of 51%.

Mortality

Mortality data from 14 trials were combined. UDCA had no significant effects on mortality (RR 0.97, 95% CI 0.67 to 1.42, I^2 = 0%, Figure 2). In the UDCA group 45/699 (6.4%) patients died versus 46/692 (6.6%) patients in the control group. The moment-based estimate of between-trial variance is 0.042, which is relatively small.

To take the missing data into account, we used uncertainty method to estimate the UDCA effect on mortality.[82] The result was consistent with the main finding above (RR 1.08, 95% CI 0.68 to 1.70). The Bayesian meta-analysis results (median OR 0.89, 95% CrI 0.50 to 1.49) also supported the main analysis presented as OR (OR 0.97, 95% CI 0.62 to 1.51). When adjusted for underlying risks the median OR is 0.82 and 95% CrI from 0.43 to 1.51 (Table 3 in Appendix 1B).

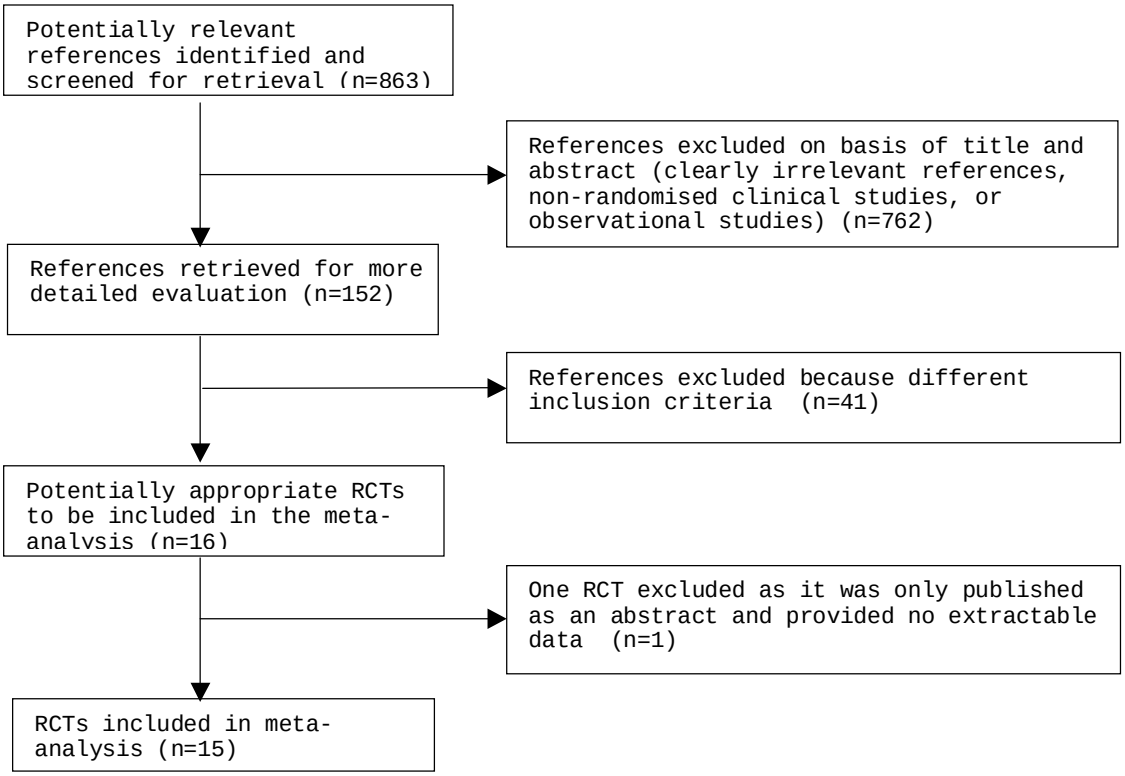
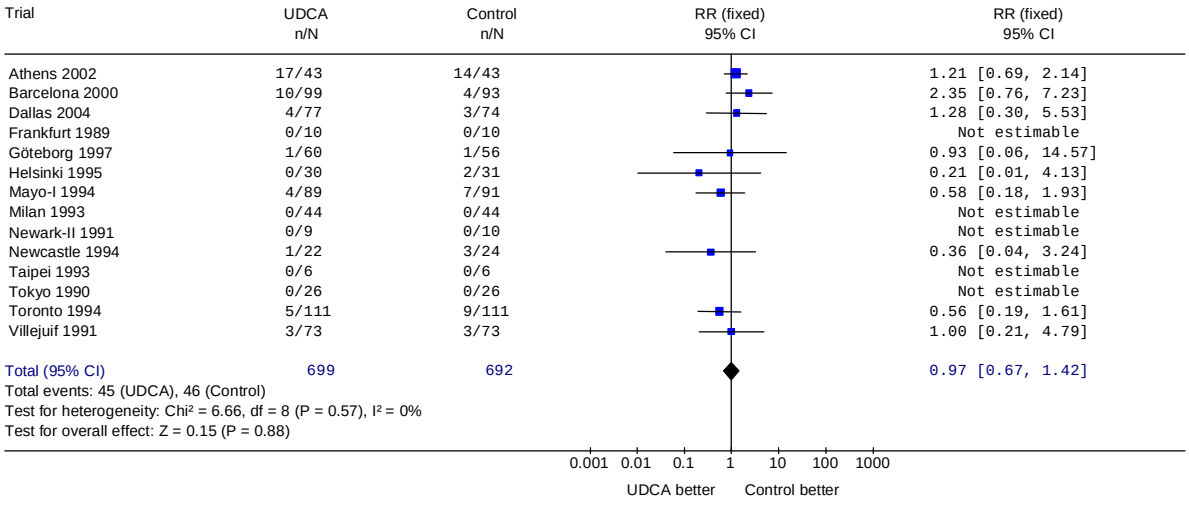


Figure 1. Flow diagram of trial.



Abbreviations: CI - confidence interval; n - number of patients with outcome; N - number of participants at risk; df - degrees of freedom; Chi^2 - chi-squared statistic; I^2 - the percentage of total variation across studies that is due to heterogeneity rather than chance. The result and its 95% CI are represented by a diamond, with the relative risk

(95% CI) and its statistical significance given alongside. Squares or diamonds to the left of the solid vertical line indicate benefit with UDCA, but this is conventionally significant (p < 0·05) only if the horizontal line or diamond does not overlap the solid vertical line.

Figure 2. Forest plot of effect of UDCA on mortality.

In meta-regression model we included risk of bias of the trials, UDCA dose, trial duration, and severity of PBC at entry as covariates and the effects of UDCA on mortality as a dependent variable. The model identified trial duration and severity of PBC as two covariates, which might have associations with the effects of UDCA (Table 2 in Appendix 1B). The moment-based estimate of between-trial variance changed from 0.042 to 0. As a sensitivity analysis, Bayesian meta-regression was also used to estimate the influence of the trial duration and disease severity on UDCA effect (Table 3 in Appendix 1B).

Including data from the extended follow-up during UDCA→UDCA versus placebo→UDCA into the analyses demonstrated a RR of 0.97 with 95% CI 0.73 to 1.30. It comprised 76 deaths in 699 patients (10.9%) originally randomised to UDCA versus 78 deaths in 692 patients (11.3%) originally randomised to placebo.

Other outcomes

Combining the results of 15 trials demonstrated no significant effects on mortality or liver transplantation either favouring UDCA or placebo (RR 0.92, 95% CI 0.71 to 1.21). In the UDCA group 83/713 (11.6%) patients died or were transplanted versus 89/706 (12.6%) patients in the control group.

UDCA effect on the composite outcome allowing for missing data was estimated as RR 1.05 with 95% CI 0.75 to 1.48. The Bayesian analysis (median OR 0.84, 95% CrI 0.53 to 1.30) supported the main analysis presented as OR (OR 0.90, 95% CI 0.65 to 1.26). When adjusted for baseline risk, the median OR is 0.77 (95% CrI from 0.43 to 1.37).

Combining the results of the 14 trials that were able to provide data demonstrated no significant effects on liver transplantation favouring UDCA (RR 0.82, 95% CI 0.53 to 1.26). UDCA did not improve patients' pruritus, fatigue, autoimmune conditions, or liver histology. UDCA improved biochemical variables, such as serum bilirubin, and might ameliorate ascites and jaundice. The use of UDCA is significantly associated with adverse events, mainly weight gain (See Appendix 1A for details)

Publication bias and other biases

Neither the Egger's nor the Begg's graphs and their tests on mortality data provided evidence for asymmetry (Egger's test, P = 0.47; Begg's test, P = 0.83).

Conclusion

We found no significant benefit of UDCA on mortality and mortality or liver transplantation. It confirms and extends the main findings of the Goulis et al meta-analysis[32] and the previous Cochrane review.[4] The effects of UDCA on mortality seem to associate with trial duration and disease severity: the longer the trial, the less effects of UDCA, if any, and the more severe the PBC, the more effects of UDCA, if any. These findings are in direct contrasts to the common claims that UDCA ought to be started early in less diseased patients in order to show its 'full effect'.[3;92] There has been no updated data on liver biochemistry since 2001, and we confirm an improvement in liver biochemistry, jaundice, and ascites following UDCA intervention. However, these results are based on few trials with sparse data. Trial selection bias and outcome reporting bias should, therefore, be considered. UDCA is generally well tolerated in patients with primary cirrhosis.

D-penicillamine versus placebo or no intervention

Description of included trials

Figure 3 summarises the literature search. Six trials compared d-penicillamine versus placebo or no intervention.[5;6;8;41;93;94] Bodenheimer et al. compared two different d-penicillamine dosages: 750 mg/day versus 250 mg/day.[43]

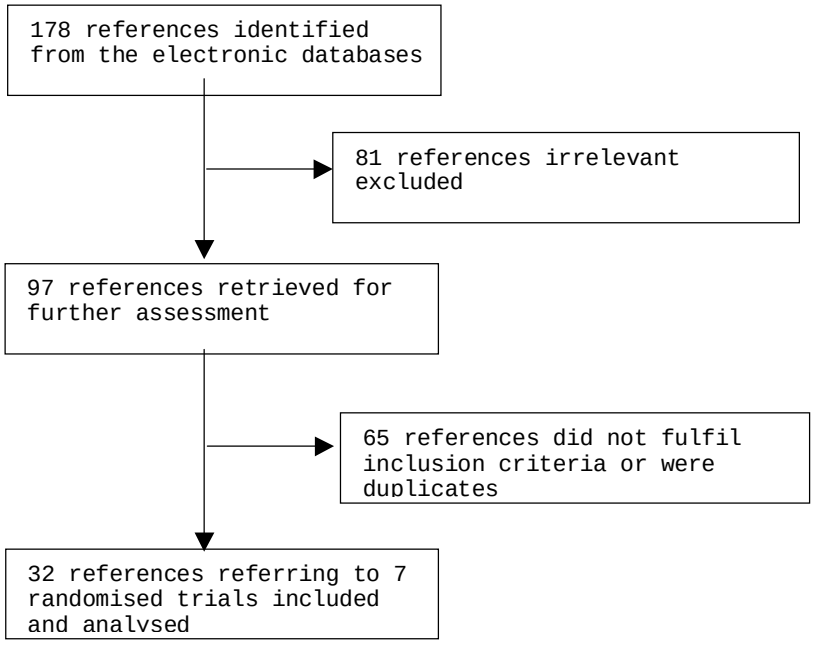


Figure 3. Flow diagram of trial.

The mean age of the patients was 51 years. Most of the patients were women (90.3%, 495/548) in the four trials reporting gender. Most of patients had advanced histological stages at entry (stage III or IV/stage I or II: 443/168).[5;6;8;94] The trial duration, including treatment and follow-up, varied from 1.5 to 10 years. Only Taal et al. reported the length of treatment and follow-up separately.[41]

In terms of methodological quality Dickson et al. and Matloff et al. were regarded as low-bias risk trials. [6;94] No trials reported sample size estimation. Five trials reported the number of dropouts in d-penicillamine (74 patients) and in control group (16 patients), respectively.[5;6;8;41;93;94] Bodenheimer et al. only reported the total number

of dropouts in both groups (26 patients).[43] Epstein et al. did not report the extractable data on dropouts.[5] No trials reported that they have used intention-to-treat analyses.

Mortality

D-penicillamine has no significant effects on mortality (RR 1.08, 95% CI 0.82 to 1.43, P = 0.56, six trials, 525 patients)(Figure 4). The RR of mortality allowing for the missing data was 0.92 with an uncertainty interval from 0.61 to 1.38. The degree of heterogeneity was moderate (I^2 = 42.9% and 0%, respectively).

We performed subgroup analyses according to methodological quality, dosage of d-penicillamine, duration of treatment and follow-up (shorter or longer than three years), and histological stage (Table 3 of Appendix 2B).

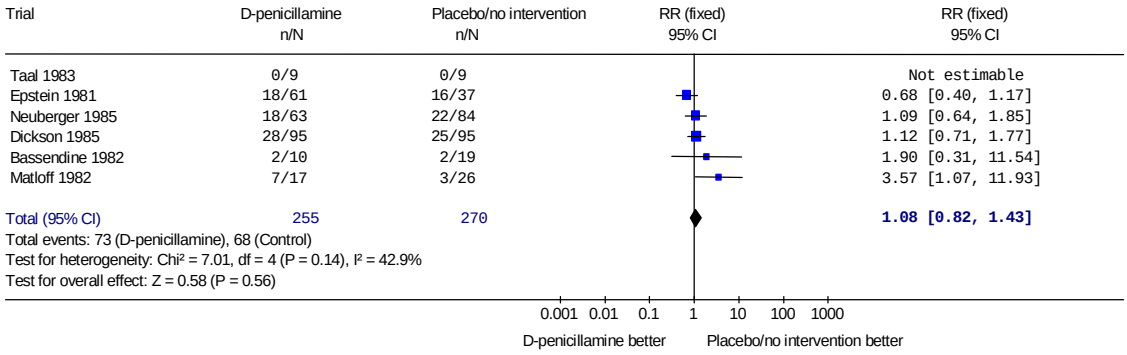


Figure 4. Relative risk of mortality in PBC patients randomised to d-penicillamine versus placebo or no intervention (complete case analysis).

Other outcomes

D-penicillamine did not significantly affect the composite outcome of mortality or liver transplantation (RR 1.09, 95% CI 0.83 to 1.43, P = 0.54, six trials, 525 patients), pruritus, liver complications, progression of liver histological stage, or liver biochemical variables. D-penicillamine decreased serum alanine aminotransferase activity but

led to significantly more adverse events (RR 4.18, 95% CI 1.38 to 12.69, P = 0.01). (See Appendix 2A for details)

Conclusion

We found that d-penicillamine had no significant effect on mortality. The pooled estimate from high-quality trials supports this finding. The estimate also holds after increasing uncertainty to allow for informative missing data due to dropouts. D-penicillamine has no significant effects on pruritus, liver complications, progression of liver histological stage, and liver biochemical variables. D-penicillamine significantly decreased serum alanine aminotransferase activity, but at the cost of significantly more adverse events.

Colchicine versus placebo or no intervention

Description of included trials

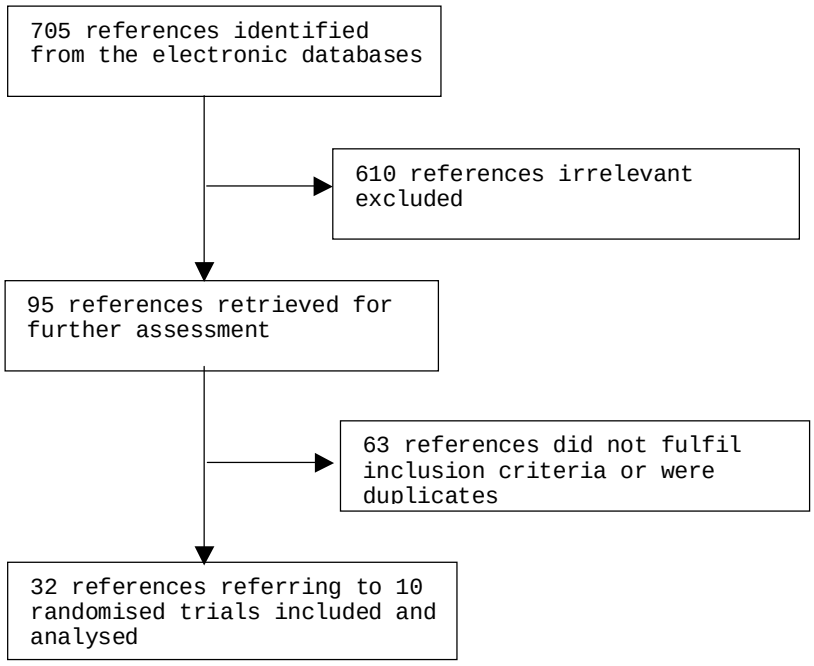


Figure 5. Flow diagram of trial selection.

Figure 5 summarises the literature search. The baseline characteristics of the 10 trials and patients are summarised in Table 1 in Appendix 3B. In terms of methodological quality, four trials were considered as low-bias risk trials. An intention-to-treat analysis was claimed in four trials. Sample size estimation was mentioned in one trial, but no estimation was based on mortality.

Mortality

Data from seven trials with 398 patients were available to estimate the risk of mortality. The available patients' course analysis (RR 1.12, 95% CI 0.51 to 2.46), the scenario assuming poor outcome (RR 1.21, 95% CI, 0.71 to 2.06), the extreme case favouring colchicine analysis (RR 0.59, 95% CI 0.30 to 1.15), and the scenario assuming good outcome (RR 1.13, 95% CI 0.52 to 2.47) showed no significant differences between colchicine and placebo or no intervention (Figure 6). The analysis favouring placebo or no intervention detected a significant detrimental effect of colchicine (RR 2.28, 95% CI 1.17 to 4.44). There was no significant heterogeneity (I^2 = 0%).

There are no significant differences across all the subgroup analyses regarding methodological quality of the trials, dosage of colchicine, trial duration, and combination of colchicine with UDCA (Table 3 of Appendix 3B).

Other outcomes

Data from eight trials with 455 patients were available to estimate the risk of mortality or liver transplantation. Neither the analysis based completed patients and the scenario assuming poor outcome, nor the scenario assuming good outcome showed any significant difference between colchicine and placebo or no intervention. The extreme case favouring colchicine or placebo or no intervention showed the significant effect favouring colchicine or placebo or no intervention. There are no significant differences across the subgroups methodological quality of the trials, dosage of colchicine, trial duration, and combination of

colchicine with UDCA (data not shown).

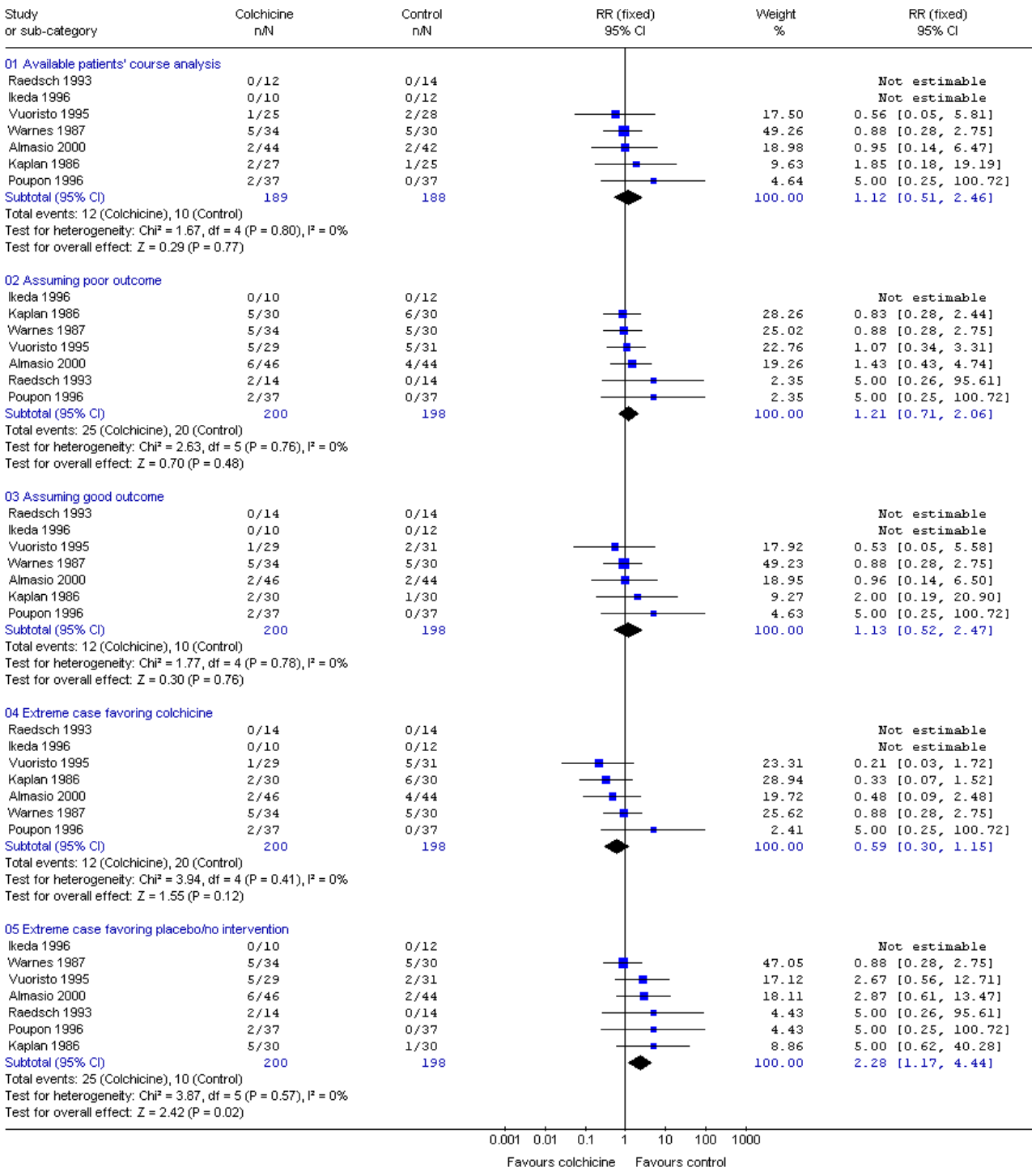


Figure 6. Relative risk of mortality in PBC patients randomised to colchicine versus placebo or no intervention – sensitivity analysis.

No significant differences were detected between colchicine and placebo or no intervention on liver biochemical variables, liver histology, or adverse events. The number of patients without improvement of pruritus significantly decreased in the colchicine group (RR 0.75, 95% CI 0.65 to 0.87). However, this estimate was based on only 156 patients from three trials. (See Appendix 3A for details).

Conclusions

We found not significant effects of colchicine on mortality and mortality or liver transplantation, compared to placebo or no intervention in patients with PBC. These observations were robust to different scenarios when missing data were considered. The scenario of extreme case favouring placebo or no intervention detected a significant detrimental effect of colchicine on mortality.

Colchicine has not improved liver biochemical and histological outcomes. Evidence showed that colchicine may have beneficial effect on pruritus. This finding was however based upon only three trials with only 156 patients. Therefore it needs to be interpreted with caution.

Methotrexate

Description of included trials

Figure 7 summarises the literature search. Five trials were included, among which four trials compared methotrexate versus placebo or no intervention and one trial compared methotrexate versus colchicine. The mean age of patients in the included trials was 53 years and 96% of the patients were female. About half the patients had liver histological stage I/II and half had stage III/IV in the three trials which reported histological stage at entry. The dosage of methotrexate differed from 7.5 mg/week, 10 mg/week, 15 mg/week, and 15 mg/m^2 body surface (maximal dose 20 mg/week). The duration of methotrexate treatment varied from 48

weeks to 10 years, and the median duration was six years. In terms of methodological quality of trials, two trials were considered as low-bias risk trials. [95;96]

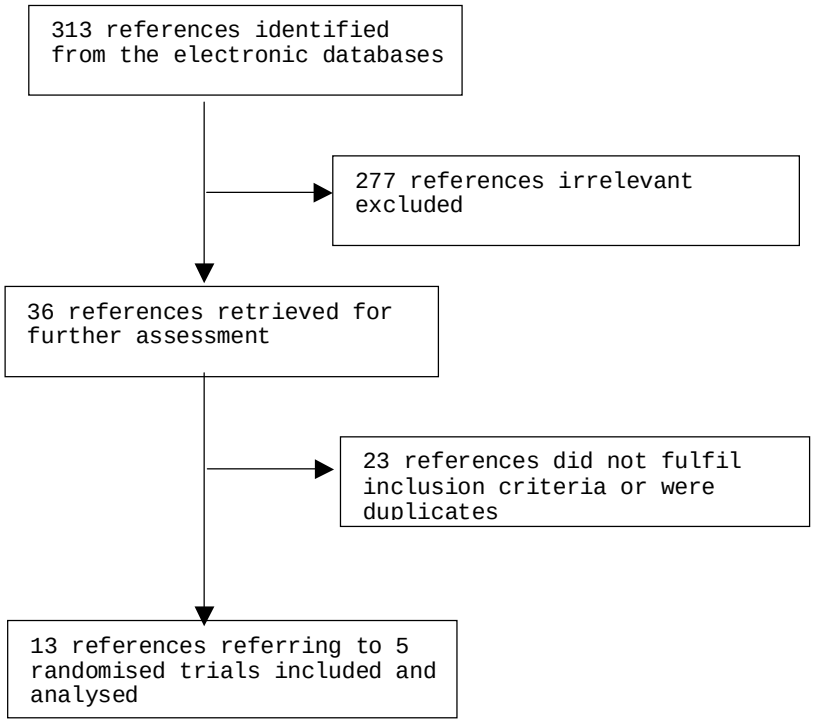


Figure 7. Flow diagram of trial selection.

Methotrexate versus placebo

Mortality

Two trials[60;95]showed that methotrexate had a significantly detrimental effect on mortality(RR 5.00, 95% CI 1.19 to 20.92, Figure 8). The sensitivity analyses did not significantly change the estimate.

Other outcomes

We pooled the estimate of hazard ratio from Hendrickse et al and Combes et al to achieve the overall effect on survival plus liver

transplantation (HR 1.44, 95% 0.46 to 4.54, random effects; HR 1.18, 95% 0.64 to 2.16, fixed effect, I^2 = 63.0%), pruritus, fatigue, liver complications, liver biochemistry, or liver histology (Appendix 4 for details).

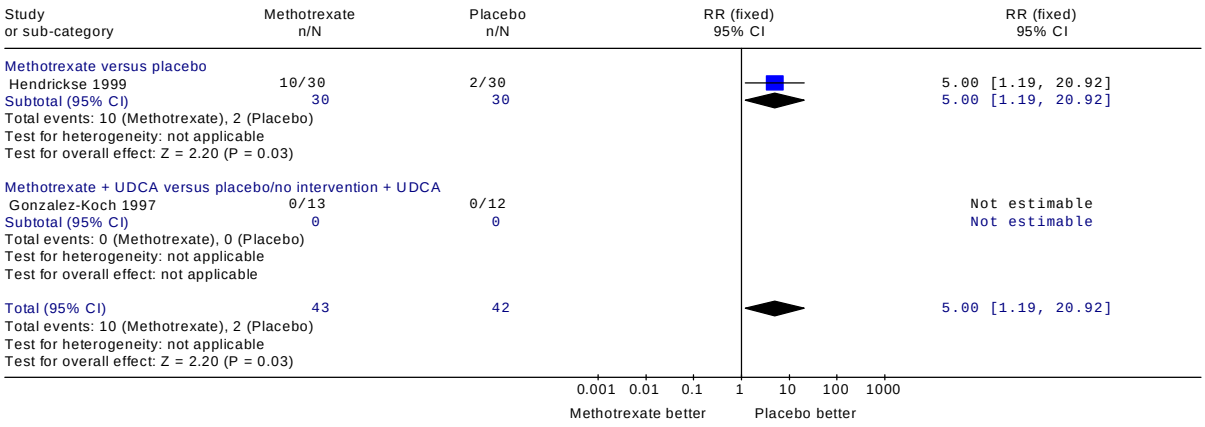


Figure 8. Relative risk of mortality in PBC patients randomised to methotrexate versus placebo or no intervention.

Conclusions

Evidence showed that methotrexate increased mortality in patients with PBC from two long-period randomised clinical trials. We do not advocate the use of methotrexate for patients with PBC. Although the majority of the evidence did not point to a beneficial effect of methotrexate for patients with PBC, we are not able to exclude the possibility for a beneficial effect in certain patient groups. We advise that any new placebo-controlled trials with methotrexate for patients with PBC should monitor harmful effects closely.

Azathioprine versus placebo or no intervention

Description of included trials

Figure 9 summarises the literature search. Two randomised clinical trials were included and they were parallel group trials published as full articles. Both trials reported random allocation of 293 patients with PBC to: azathioprine versus no intervention;[65] azathioprine versus placebo.[15] The mean age of patients in the included trials was 53 years and 90% of the patients were women. Half patients had histological stage III or IV in Christensen et al.[15] The dosage of azathioprine used in the Heathcote et al trial (2 mg/kg/day) was higher than used in Christensen et al trial (300-700 mg/week).[15;65] The trial duration (treatment plus follow-up) was 5 years in Heathcote et al and 11 years in Christensen et al trial. [15;65] Christensen et al was considered as a low-bias risk trial. [8]

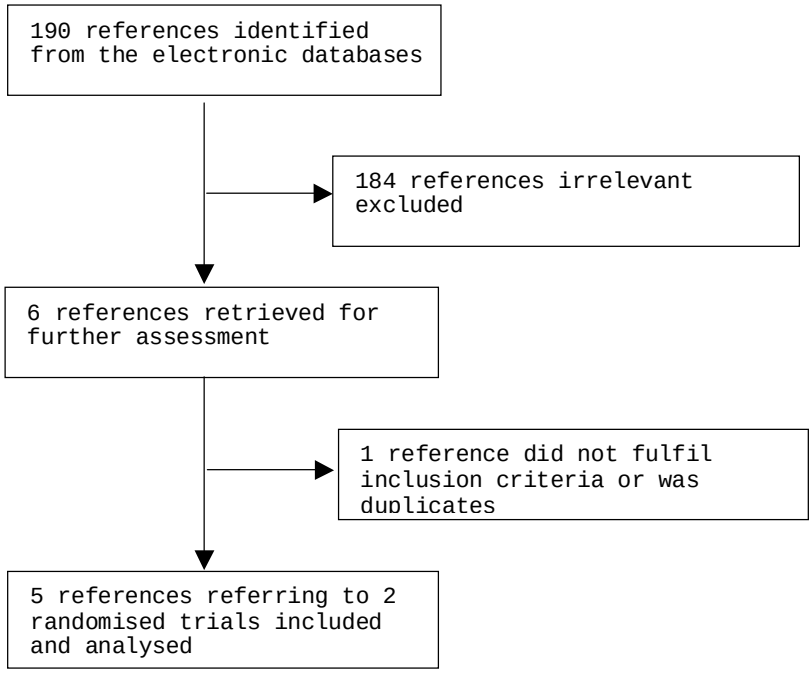


Figure 9. Flow diagram of trial selection.

Mortality

Seventeen patients died in Heathcote et al trial, whereas 119 patients died in Christensen et al trial. Considering the impact of missing data, azathioprine did not significantly reduce the risk of death (RR 0.80, 95% CI 0.49 to 1.31, pooled uncertainty intervals)(Figure 10). The finding still holds when only data on available patients were included (RR 0.88, 95% CI 0.74 to 1.06)(Figure not shown). It is noteworthy to mention the standard survival analysis in the Christensen et al trial revealed no significant difference between the two groups. When adjustment for imbalances between the two groups (primarily serum bilirubin) was made, however, there was a slight but statistically significant difference in survival favouring azathioprine ($P < 0.01$).

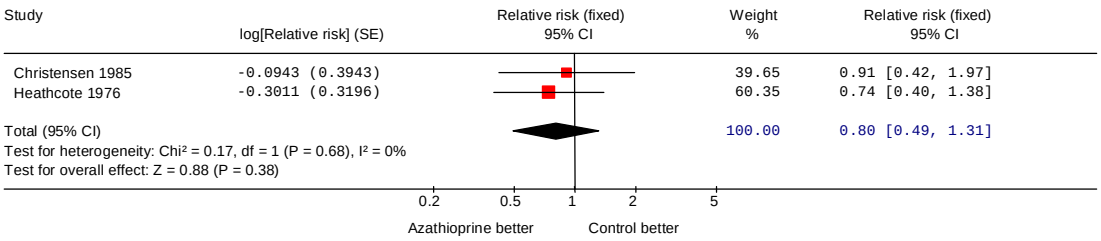


Figure 10. Relative risk of mortality in PBC patients randomised to azathioprine versus placebo or no intervention using the uncertainty method.

Other outcomes

No patients had liver transplanted so this evaluation could not be done. Azathioprine did not improve pruritus at one-year intervention (RR 0.71, 95% CI 0.28 to 1.84, 1 trial), cirrhosis development, or quality of life. Patients given azathioprine experienced significantly more adverse events than patients given placebo or no intervention (RR 2.44, 95% CI 1.14 to 5.20, 2 trials). The common adverse events were rash, severe diarrhoea, and bone marrow depression (Appendix 5 for details).

Conclusions

The results of our systematic review do not support azathioprine for

patients with PBC. Patients given azathioprine suffered from more adverse events than patients given no intervention or placebo, though not all adverse events were severe.

Cyclosporin A versus placebo

Description of included trials

Figure 11 summarises the literature search. Three randomised clinical trials with 390 patients were included. The mean age of the patients was about 52 years. The majority of the patients were women (women/men: 338/52). Slightly more patients had stage III or IV than stage I or II (178/154). The dose of cyclosporin A was 2.5, 3, or 4 mg/kg/day. The duration of treatment and follow-up varied from one to three years. Overall, two trials were regarded as low-bias risk trials.[97]
[70]

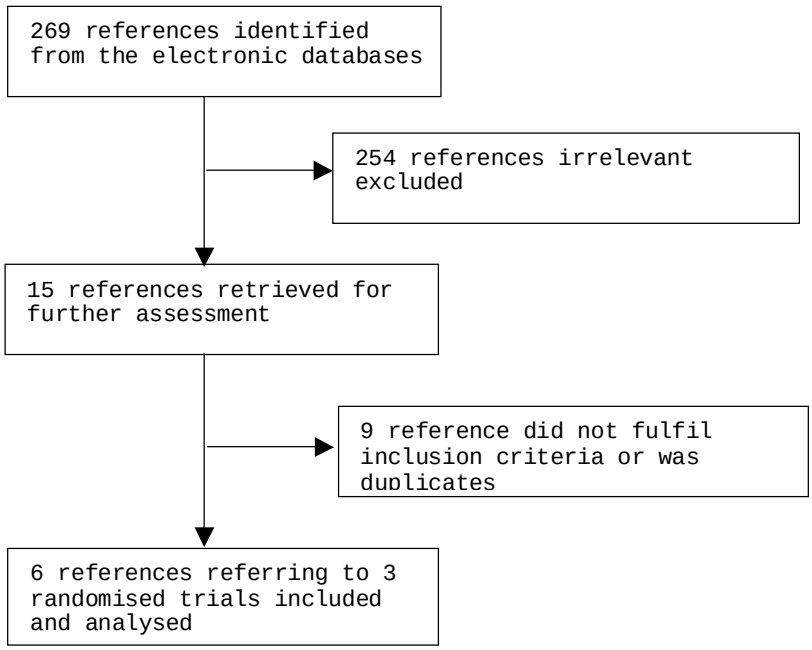


Figure 11. Flow diagram of trial selection.

Mortality

The three trials provided data to estimate the risk of mortality of cyclosporin A versus placebo. Compared with placebo, cyclosporin A did not significantly affect mortality (15% vs. 17%). The relative risk was 0.92 (95% CI 0.59 to 1.45) (Figure 12).

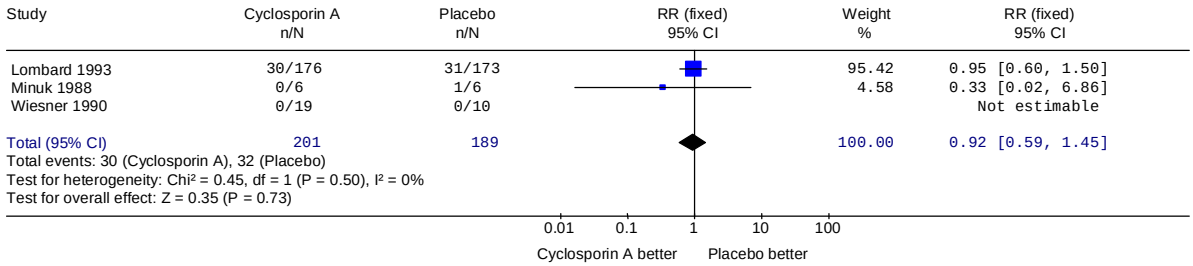


Figure 12. Relative risk of mortality in PBC patients randomised to cyclosporin A versus placebo or no intervention.

Other outcomes

Compared with placebo, cyclosporin A did not significantly affect mortality or liver transplantation (RR 0.85, 95% CI 0.60 to 1.20). It significantly improved pruritus (Standardised Mean Difference (SMD) -0.38, 95% CI -0.63 to -0.14) and significantly reduced alanine aminotransferase (WMD -41U/L, 95% CI -63 to -18) and increased serum albumin level (WMD 1.66 g/L, 95% CI 0.26 to 3.05). More patients experienced adverse events in cyclosporin A group, especially renal dysfunction (Peto odds ratio 5.56, 95% CI 2.52 to 12.27) and hypertension (SMD 0.88, 95% CI 0.27 to 1.48)(Appendix 6 for details).

Conclusions

We found no evidence supporting or refuting that cyclosporin A may delay the progression to death or liver transplantation, and advanced stage of PBC. Cyclosporin A may beneficially affect pruritus and liver biochemistry of patients with PBC, but at the cost of adverse events, especially renal dysfunction and hypertension. We do not recommend use of cyclosporin A outside randomised clinical trials.

Discussion

The six systematic reviews include 42 trials with 4009 patients with primary cirrhosis and the duration of the trials range from three months to eleven years. The currently available reliable evidence has not shown beneficial effects of UDCA, d-penicillamine, colchicine, methotrexate, azathioprine, or cyclosporin A on survival of patients with PBC. However, the trials and reviews on these interventions are under-powered to draw firm conclusions; the confidence intervals include both possible beneficial and possible detrimental effects. UDCA might improve biochemical variables and clinical symptoms such as ascites and jaundice. Colchicine might improve pruritus. Methotrexate might improve pruritus and levels of serum alkaline phosphatases and plasma immunoglobulin M. Cyclosporin A significantly improved pruritus, reduced alanine aminotransferase, and increased serum albumin level. However, these results are based on few trials with sparse data. Trial selection bias and outcome reporting bias should therefore be considered.

UDCA was associated with adverse events, mainly weight gain. D-penicillamine did not appear to reduce the risk of mortality or morbidity, and led to significantly more adverse events. Colchicine tended to lead to more adverse events (mostly transient diarrhoea, usually resolved by lowering the dose), although it is not statistically significant. Patients given azathioprine experienced more adverse events than patients given no intervention or placebo, such as rash, severe diarrhoea and bone marrow depression. Cyclosporin A caused more adverse events, especially renal dysfunction and hypertension.

The reader is referred to the attached papers (Appendix 1A-6)for detailed discussion of each intervention.

Limitations of the trials in PBC patients

In general, the methodological quality was low in most trials in PBC. Among the 42 trials, 26 (62%) had high risk of bias, i.e., low methodological quality. Such trials tend to significantly overestimate intervention effects.[98-100] If the overestimation is valid also in the PBC trials, the prospects for the six interventions investigated may be worse than observed.

In addition, most trials (95%, 40/42) have shorter follow-up than the estimated median survival of PBC, i.e., 10 to 15 years.[101] Therefore, it is difficult to detect a significant difference on mortality based on the trials, most of which have low statistical power.

In our studies, we could not demonstrate beneficial effects of UDCA, d-penicillamine, colchicine, methotrexate, azathioprine, or cyclosporin A on survival of patients with PBC. However, UDCA, d-penicillamine, methotrexate, and cyclosporin A seemed to improve some liver biochemical outcome measures. This may place clinicians and researchers in a dilemma. If therapeutic decisions are based on clinical outcomes (e.g., mortality), there is insufficient evidence to support their use in PBC. But if based on non-validated 'surrogate' outcomes (e.g., serum bilirubin level), there is evidence favouring the interventions for the disease. This dilemma was reflected in a survey among Danish doctors why they prescribed UDCA to patients with PBC.[102] It turns out that they have very different reasons for choosing an intervention. Sixteen percent of the doctors thought UDCA reduced mortality, 27 percent thought UDCA reduced morbidity, and 23 percent thought it benefited 'surrogate' outcomes.[102] However, as along as we have no significant effect on chemical relevant outcomes we have no validated surrogate.[103]

The Mayo Risk Score Model has identified several prognostic biomarkers for PBC, e.g., serum bilirubin. These biomarkers may respond to intervention and may be predictive of survival. But they do not necessarily predict clinical benefit of the interventions in question because "a perfect correlation does not a surrogate make".[104] In the absence of validated surrogate outcomes in these interventions for PBC,

confirmatory trials assessing their effects should only be based on clinical outcomes, e.g., survival. We believe that assessment of the effect of interventions on clinical outcomes will benefit patients in the long run.[103]

We realise that the challenges of performing new trials on interventions for PBC. The estimated median survival of PBC is 10 to 15 years. To spend 15 years planning and carrying out a trial for each new potential treatment for PBC would consume many patients' lifetimes, not to mention the expense and difficulty of retaining patients in such long trials.[105] Nevertheless, there are at least an estimated one million patients with PBC in the world. Therefore, it is possible to conduct large trials with appropriate statistical power, if international groups of investigators active in intervention for PBC collaborate. Such large trials do not need to be conducted for more than two to four years. The main objective is to collect enough outcomes, i.e., patients surviving or patients without jaundice, ascites, pruritus, fatigue, etc.

Strengths and limitations of the systematic review

Despite the limitations within the clinical intervention research in PBC, it is important to perform a systematic, critical appraisal of the available data. Systematic review has several strengths. It allows a more objective appraisal of the evidence than traditional narrative reviews; provides a more precise estimate of a treatment effect; may explain heterogeneity between the results of individual studies; and may highlight weaknesses within the research field and generate important research questions to be addressed in future studies.[106] The strengths of Cochrane Reviews are, in addition, that they are made available electronically (both on CD-ROM and the Internet) and regularly updated.

Systematic review may, however, also have its limitations. The nature of systematic reviews is that of a retrospective observational study with all the bias risks this entails. Third, systematic reviews are threatened by publication bias and selective reporting of outcomes in

the individual identified trials.

As other observational studies, systematic reviews have a considerable risk of bias and confounding.[107] In order to minimise this and to enhance transparency, a good systematic review should be based on a pre-specified, peer-reviewed, published protocol. This contains a clearly formulated question and descriptions of explicit methods in the identification, selection, and evaluation of included trials.

In many cases a systematic review will include a meta-analysis, which offers a quantitative summary of the results from individual study. Meta-analysis is often performed retrospectively on studies, which have not been planned with this in mind. Such meta-analysis can include only studies for which relevant data are retrievable. If only published studies are included, this raises concerns about publication bias, whereby probability of a study being published depends on the statistical significance of the results.[108]

Even if a study is published, there may be selective reporting of results, so that only the outcomes showing a statistically significant treatment difference are chosen from amongst the many analyses. Recent research has shown that the reporting of outcomes in randomised trials is frequently inadequate and biased to favour statistical significant outcomes.[109] Meta-analysts may use different methods to reduce the risks of bias by selective reporting.[110;111]

Meta-analyses are most often based on aggregate patient data (APD) from completed studies that have been published in the medical literature, like these six reviews. Few would argue that properly conducted meta-analyses based on individual patient data (IPD) have several advantages. Clearly, IPD is advantageous when different outcomes or cut-points are reported in the APD. However, when based on the same studies, summary

effect measures based on IPD and APD meta-analyses are virtually identical.[112] While both approaches permit exploration of study and summary patient sources of heterogeneity, only IPD permits full exploration of and adjustment for patient characteristics.[113] It is important to remember, that such analyses, e.g., identifying a subgroup of patients particularly benefit from the interventions in investigation, are only exploratory and hypothesis generating.

Overall, the present work comprises systematic reviews, which were all based on pre-specified, peer-reviewed, and published protocols. In all reviews, we performed comprehensive searches of major databases and contacted authors and pharmaceutical companies. We appraised the quality of all included trials and emphasised the results of trials with low bias risk in our conclusions. Nevertheless, our systematic reviews may still be prone to both publication and reporting bias. Therefore, the results may well tend to overestimate the possible benefits of the interventions evaluated in the present systematic reviews.

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Appendix 1A

Ursodeoxycholic acid for primary biliary cirrhosis

Review information

Authors

Yan Gong[1], Zhi Bi Huang[2], Erik Christensen[3], Christian Gluud[1]

[1]Cochrane Hepato-Biliary Group, Copenhagen Trial Unit, Centre for Clinical Intervention Research, Copenhagen, Denmark
2
Department of Epidemiology, Institute of Public Health, The University of Copenhagen, Copenhagen, Denmark
[3]Clinic of Internal Medicine I, Bispebjerg Hospital, Copenhagen, Denmark

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Contact person

Yan Gong

Cochrane Hepato-Biliary Group
Copenhagen Trial Unit, Centre for Clinical Intervention Research
Rigshospitalet, Dept. 3344
Blegdamsvej 9
DK-2100 Copenhagen
Denmark

E-mail: ygong@ctu.rh.dk

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Abstract

Background

Primary biliary cirrhosis is an uncommon autoimmune liver disease with unknown etiology. Ursodeoxycholic acid (UDCA) has been used for primary biliary cirrhosis, but the effects on survival remain controversial.

Objectives

Evaluate the effects of UDCA on patients with primary biliary cirrhosis against placebo or no intervention.

Search strategy

We searched through *The Cochrane Hepato-Biliary Group Controlled Trials Register, The Cochrane Central Register of Controlled Trials on The Cochrane Library, MEDLINE, EMBASE, SCI-EXPANDED, The Chinese Biomedical CD Database, LILACS*, and in references of identified studies. The last search was done in December 2005.

Selection criteria

Randomised clinical trials evaluating UDCA versus placebo or no intervention in patients with primary biliary cirrhosis.

Data collection and analysis

The primary outcomes are mortality and mortality or liver transplantation. Binary outcomes were reported as odds ratio (OR) and continuous outcomes as weighted mean difference, both with 95 confidence intervals (CI). Meta-regression was used to investigate the associations between UDCA effects with quality of trial, UDCA dose, trial duration, and patients' severity of primary biliary cirrhosis. We also used Bayesian meta-analytic approach to estimate the UDCA effect as sensitivity analysis.

Results

Sixteen randomised clinical trials evaluating UDCA against placebo or no intervention were identified. Data from three trials has been updated. Nearly half of the trials has high risk of bias. The combined results demonstrated no significant effects favouring UDCA on mortality (OR = 0.97; 95% CI 0.62 to 1.51) and mortality or liver transplantation (OR = 0.90; 95% CI 0.65 to 1.26). The findings were supported by the Bayesian meta-analyses. Meta-regression analyses identified trial duration and disease severity having associations with UDCA effect on mortality. UDCA did not improve patients' pruritus, fatigue, autoimmune conditions, quality of life, liver histology, or portal pressure. However, UDCA significantly reduced ascites, jaundice, and biochemical variables such as serum bilirubin. The use of UDCA is significantly associated with adverse events, mainly weight gain. Including data after patients had been switched onto open label UDCA found no significant effect of ursodeoxycholic acid on mortality and mortality or liver transplantation. However, a significant effect was observed on liver transplantation (OR = 0.70; 95% CI 0.50 to 0.98).

Authors' conclusions

This systematic review did not demonstrate any benefit of UDCA on mortality and mortality or liver transplantation of patients with primary biliary cirrhosis, although it showed a reduction in liver biochemistry, jaundice, and ascites following UDCA intervention. UDCA intervention is associated with weight gain.

Plain language summary

Ursodeoxycholic acid is not likely to yield a benefit on survivals of patients with

primary biliary cirrhosis

Primary biliary cirrhosis is an uncommon cholestatic liver disease, occurring mainly in middle-aged women. Ursodeoxycholic acid is the only drug proved to treat primary biliary cirrhosis. Although ursodeoxycholic showed a reduction in liver biochemistry, jaundice, and ascites, this review did not demonstrate any benefit of ursodeoxycholic acid on mortality and mortality or liver transplantation. Its use is associated with weight gain.

Background

Primary biliary cirrhosis is an uncommon and slowly progressive autoimmune disease of the liver that primarily attacks middle-aged women. It was first comprehensively described around 1950 (MacMahon 1949; Ahrens 1950). Over the last 30 years, substantial increases in the prevalence of primary biliary cirrhosis have been observed (Kim 2000). Primary biliary cirrhosis is now a frequent cause of liver morbidity, and the patients are significant users of health resources, including liver transplantation (Prince 2003).

Histopathologically, a progressive granulomatous hepatitis destroys small septal and interlobular bile ducts. The loss of bile ducts leads to decreased bile secretion and the retention of toxic substances within the liver, resulting in further hepatic damage, fibrosis, cirrhosis, and eventually, liver failure (Kaplan 2005). Fatigue and pruritus are the most common presenting symptoms. Other findings include hyperlipidaemia, hypothyroidism, osteopenia, and coexisting autoimmune diseases, including Sjögren's syndrome and scleroderma. The diagnosis of primary biliary cirrhosis is currently based on the triad: the presence of detectable antimitochondrial antibodies in serum; elevation of liver enzymes (most commonly alkaline phosphatases) for more than six months; and characteristic liver histological changes in the absence of extrahepatic biliary obstruction (Kaplan 1996).

Bile duct destruction leads to the retention of hydrophobic bile acids within the liver cell, and this most likely contributes to the gradual deterioration in liver function observed in patients with primary biliary cirrhosis. Ursodeoxycholic acid (UDCA), the epimer of chenodeoxycholic acid, increases the rate of transport of intracellular bile acids across the liver cell and into the canaliculus in patients with primary biliary cirrhosis (Jazrawi 1994). UDCA treatment reduces intracellular hydrophobic bile acid levels and thereby may have a cytoprotective effect on cell membranes. UDCA may also act as an immunomodulatory agent (Calmus 1992).

UDCA is the only drug approved for primary biliary cirrhosis by the Food and Drug Administration in 1997. Doses of 13 to 15 mg/kg/ day cause significant improvements in liver tests and immunoglobulin levels and reduce titers of antimitochondrial antibodies (TORONTO; BARCELONA). However, the effect of UDCA on mortality and histological progression remains controversial (Goulis 1999; Gluud 2001 b). Since 2001, several randomised clinical trials have been published with the results of longer-term follow-up on patients' survivals (ATHENS; DALLAS; MAYO-I). We therefore intend to re-evaluate the effects of UDCA in patients with primary biliary cirrhosis using the updated results.

Objectives

The objectives are to evaluate the effects of UDCA on patients with primary biliary cirrhosis.

Methods

Criteria for considering studies for this review

Types of studies

We included randomised clinical trials irrespective of blinding, language, year of publication, and publication status. We excluded studies using quasi-randomisation (for example, allocation by date of birth).

Types of participants

Patients with primary biliary cirrhosis, ie, a positive result for serum mitochondrial antibody, and/or elevated serum activity of alkaline phosphatases (or other markers of intrahepatic cholestasis), and/or liver biopsy findings diagnostic for or compatible with primary biliary cirrhosis.

Types of interventions

Peroral administration of UDCA at any dose versus placebo or no intervention. Co-interventions were allowed as long as the intervention arms of the randomised clinical trial receive similar co-interventions.

Types of outcome measures

Primary outcome measures were:
Mortality. Mortality or liver transplantation.
Secondary outcome measures were:

- Liver transplantation.
- Pruritus: number of patients with pruritus or pruritus score.
- Fatigue: number of patients with fatigue.
- Other clinical symptoms: number of patients developing jaundice, portal pressure, (bleeding) oesophageal varices, (bleeding) gastric varices, ascites, hepatic encephalopathy, hepato-renal syndrome, sicca complex, scleroderma-like lesions.
- Liver biochemistry: serum (s-)bilirubin; s-alkaline phosphatases; s-gamma-glutamyltransferase; s-aspartate aminotransferase; s-alanine aminotransferase; s-albumin; s-cholesterol (total); plasma immunoglobulins.
- Liver biopsy: worsening of liver histological stage or score.
- Quality of life: physical functioning (ability to carry out activities of daily living such as self-care and walking around), psychological functioning (emotional and mental well-being), social functioning (social relationships and participation in social activities), and perception of health, pain, and overall satisfaction with life.
- Adverse events (excluding mortality and liver transplantation): The adverse event is defined as any untoward medical occurrence in a patient in either of the two arms of the included randomised clinical trials, which did not necessarily have a causal relationship with the treatment, but did, however, result in a dose reduction, discontinuation of treatment, or registration of the advent as an adverse event/side effect (ICH-GCP 1997).
- Cost-effectiveness: the estimated costs connected with the interventions were weighed against any possible health gains.

Search methods for identification of studies

We searched for trials in *The Cochrane Hepato-Biliary Group Controlled Trials Register* (Gluud 2005), *The Cochrane Central Register of Controlled Trials* on *The Cochrane Library*, *MEDLINE*, *EMBASE, SCI-EXPANDED, The Chinese Biomedical CD Database, LILACS*, and in references of identified studies. The detailed searching strategy is listed in Table 1. The last research was performed in December 2005.

Data collection and analysis

We performed the meta-analysis following the protocol (Gluud 1999 a) and the recommendations given by the Cochrane Handbook for Systematic Reviews of Interventions (Higgins 2005).

Data Extraction
Two authors (YG and EC) independently evaluated whether newly identified trials fulfilled the inclusion criteria. We listed the excluded trials in 'Characteristics of excluded studies' with the reasons for exclusion. YG extracted data and EC validated the data extraction. Disagreements were resolved by discussion with YG, EC, and CG.

We assessed the methodological quality of the randomised clinical trials using four components (Schulz 1995; Moher 1998; Kjaergard 2001) as follows. Trials with low risk of bias were the ones meeting the adequacy criteria of the first three components.

Generation of the allocation sequence
Adequate, if the allocation sequence was generated by a computer or random number table. Drawing of lots, tossing of a coin, shuffling of cards, or throwing dice are considered as adequate if a person who was not otherwise involved in the recruitment of participants performed the procedure;Unclear, if the trial was described as randomised, but the method used for the allocation sequence generation was not described.
Allocation concealment

- Adequate, if the allocation of patients involved a central independent unit, on-site locked computer, numbered drug bottles or containers with identical appearance prepared by an independent pharmacist or investigator, or sealed envelopes;
- Unclear, if the trial was described as randomised, but the method used to conceal the allocation was not described;
- Inadequate, if the allocation sequence was known to the investigators who assigned participants.

Blinding (or masking)
Adequate, if the trial was described as double blind and the method of blinding involved identical placebo or active drug;Unclear, if the trial was described as double blind, but the method of blinding was not described;Not performed, if the trial was not double blind.
Follow-up

- Adequate, if the numbers and reasons for dropouts and withdrawals in all intervention groups were described or if it was specified that there were no dropouts or withdrawals;
- Unclear, if the report gave the impression that there had been no dropouts or withdrawals, but this was not specifically stated;
- Inadequate, if the number or reasons for dropouts and withdrawals were not described.

The following items were recorded from the individual trial: mean (or median) age, sex ratio, histological stage, other baseline characteristics including serum (s)-bilirubin concentration, dose of UDCA, and type of intervention in the control group.

In the protocol for this systematic review (Gluud 1999 a) we only intended to extract data from the time when patients were on UDCA versus placebo/ no intervention in order to secure data from the most unbiased comparisons. However, due to comments raised by some of the peer-reviewers we also extracted data on mortality and/or liver transplantation at the maximal follow-up of each trial, including data from patients switched from blinded UDCA onto open label UDCA (UDCA→UDCA) versus patients switched from placebo onto open label UDCA (placebo→UDCA). The interpretation of these data, however, should be performed with caution (see Discussion).

Statistical Methods
We performed meta-analyses with Review Manager 4.2. We analysed data by a random-effects model (DerSimonian 1986) and a fixed-effect model (DeMets 1987). If the results of both analyses gave the same overall results regarding significance, only the results of the fixed-effect model analysis were reported. We presented binary outcome measure as odds ratio (OR) with 95% confidence interval (CI), and continuous outcome measure as weighted mean difference (WMD) with 95% CI. Heterogeneity was explored by chi-squared test with significance set at $P < 0.10$ and the quantity of heterogeneity was measured by *I2* (Higgins 2002) and the moment-based estimate (DerSimonian 1986).

We performed a meta-regression analysis with STATA® on primary outcomes, ie, mortality and mortality or

liver transplantation. Meta-regression analysis examined the effect size of UDCA in relation to methodological quality of trials, UDCA dosage, trial duration (treatment and follow-up), and disease severity of patients at entry.

We used funnel plot to provide a visual assessment of whether treatment estimates are associated with study size. We explored publication bias and other bias according to Begg's and Egger's methods (Begg 1994; Egger 1997) with STATA®.

Sensitivity analyses

We also did sensitivity analyses to investigate the robustness of our main analyses. These sensitivity analyses were only performed on the primary outcomes, ie, mortality and mortality or liver transplantation. Bayesian approach with WINBUGS (version 1.4.1): this approach is able to account for uncertainty of all relevant sources of variability in the random-effects model. The analogue of a classical estimate is the marginal posterior median and the analogue of a classical confidence interval is the credibility interval (CrI) (). We also predicted UDCA intervention effect in a new trial;UDCA effects adjusted for underlying risk: the underlying risk is a convenient and clinically relevant trial-level measure which can be interpreted as a summary of a number of unmeasured patient characteristics. We are interested in estimating the effects of UDCA adjusted for underlying risk (). andThe influence of missing data: the missing data could be due to patient dropouts or lost to follow-up. We used an uncertainty method to pool the data on the primary outcomes allowing for missing data () .

Results

Description of studies

We identified 863 references through electronic and hand searches. We excluded 762 duplicates and clearly irrelevant references, non-randomised clinical studies, or observational studies. The remaining 101 references referred to 16 randomised clinical trials including 1447 patients. A summary of the 16 trials was listed in 'Characteristics in the included trials'. Two of the 16 randomised clinical trials were published as abstracts only (MANCHESTER; MEXICO CITY) and the MEXICO CITY trial provided no extractable data on trial's characteristics and outcomes. The excluded studies are listed under 'Characteristics of excluded studies', and the reasons for exclusion are given there. Comparing with the first version of this systematic review (Gluud 2001 b), we updated with new mortality and liver transplantation data from three trials (ATHENS; DALLAS; MAYO-I) and adverse events data from the MAYO-I trial.

UDCA dose varied from 7.7 to 15.5 mg/kg/day with a median of 10. The duration of the trials varied from 3 to 92 months with a median of 24. The percentage of patients with advanced primary biliary cirrhosis or presenting symptoms at entry varied from 15% to 83% with a median of 51%. The details are displayed in Table 2.

Following the stipulated follow-up in the UDCA-group and the placebo-group, six trials (GÖTEBORG; DALLAS; MAYO-I; MILAN; TORONTO; VILLEJUIF) continued UDCA treated patients on open label UDCA (UDCA→UDCA) and offered open label UDCA to the patients originally given placebo (placebo→UDCA). The ATHENS-trial gave continued UDCA intervention to all patients randomised to the UDCA arm and switched 14/43 'no intervention' patients to UDCA after they had been followed for a mean duration of 3.5 years. It is not possible in this trial to separate clearly data from the UDCA versus no intervention period and from the UDCA→UDCA versus no intervention-UDCA period, and only data from the latter is given.

Risk of bias in included studies

The methods to generate the allocation schedule were considered to be adequate in nine trials (ATHENS; BARCELONA; FRANKFURT; GÖTEBORG; HELSINKI; MAYO-I; MILAN; NEWCASTLE; TAIPEI). The remainder of the trials did not describe the method to generate the randomisation schedule.

The methods to conceal allocation were considered to be adequate in ten trials (ATHENS; BARCELONA; FRANKFURT; GÖTEBORG; HELSINKI; MAYO-I; NEWCASTLE; TOKYO; TORONTO; VILLEJUIF). The other

six trials had inadequate or unclear allocation of concealment.

All the trials employing placebo were described as double blind. However, the description of the placebo was only sufficient in five trials (BARCELONA; FRANKFURT; HELSINKI; MAYO-I; TORONTO), ie, the placebo was identical in appearance and smell (and to some extent taste) to UDCA. All of the remaining 'placebo'-controlled trials seemed to have problems with double blinding in that a number of the 'placebo'-controlled trials only stated that the 'placebo' tablets were identical in appearance, but did not mention smell and taste (DALLAS; GÖTEBORG; MILAN; NEWCASTLE; TAIPEI; TOKYO; VILLEJUIF). Therefore, these trials may easily have lost the essences of double blinding.

In all, six trials have met the criteria of being trials with low risk of bias (BARCELONA; FRANKFURT; GÖTEBORG; HELSINKI; MAYO-I; NEWCASTLE) and the other nine trials with high risk of bias (ATHENS; DALLAS; MANCHESTER; MILAN; NEWARK-II; TAIPEI; TOKYO; TORONTO; VILLEJUIF).

There was generally a fair description of follow-up and withdrawals/dropouts. Details could be seen in the 'Characteristics of included studies'. However, only eight trials stated that they used the intention-to-treat method in the evaluation of their data (ATHENS; BARCELONA; DALLAS; HELSINKI; NEWCASTLE; TAIPEI; TORONTO; VILLEJUIF).

Effects of interventions

Mortality (Comparison 01-01; 02-03; 04-01)
Combining the results of 14 trials demonstrated no significant effects favouring UDCA on mortality (OR = 0.97; 95% CI 0.62 to 1.51). In the UDCA group 45/699 (6.4%) patients died versus 46/692 (6.6%) patients in the control group. The moment-based estimate of between trials variance is 0.042.

The finding is consistent with the one using Bayesian approach. The marginal posterior median OR is 0.89 with 95% CrI from 0.50 to 1.49. After adjusting baseline, the median OR is 0.82 and 95% CrI from 0.43 to 1.51. We predicted that UDCA effect in a new trial may increase or decease the risk of mortality (OR 0.89; 95% CrI 0.27 to 2.69). We used an uncertainty method to estimate the UDCA effect allowing for the missing data (OR = 1.03; 95% CI 0.64 to 1.25).

In meta-regression model we included quality of trials, UDCA dose, trial duration, and patients' severity of primary biliary cirrhosis at entry as covariate and the effects of UDCA on mortality as a dependent variable. The model identifies trial duration and severity of primary biliary cirrhosis being two covariates, which might have associations with the effects of UDCA. The moment-based estimate of between trials variance changed from 0.042 to 0 (see Table 3). As a sensitivity analysis, Bayesian meta-regression was also used to estimate the influence of the trial duration and disease severity on UDCA effect (see Table 5 and Table 6).

Including data from the extended follow-up during UDCA→UDCA versus placebo→UDCA into the analyses (now comprising 76 deaths in 699 patients (10.9%) originally randomised to UDCA versus 78 deaths in 692 patients (11.3%) originally randomised to placebo) demonstrated an OR of 0.97 with 95% CI 0.68 to 1.37 (Comparison 04-01).

Mortality or liver transplantation (Comparison 01-02; 02-04; 04-02)
Combining the results of 15 trials demonstrated no significant effects favouring UDCA on mortality (OR = 0.90; 95% CI 0.65 to 1.26). In the UDCA group 83/713 (11.6%) patients died versus 89/706 (12.6%) patients in the control group.

The finding is consistent with the one using Bayesian approach. The marginal posterior median OR is 0.84 with 95% CrI from 0.53 to 1.30. After adjusting baseline risk, the median OR is 0.77 and 95% CrI from 0.43 to 1.37. We predicted that UDCA effect in a new trial may increase or decease the risk of mortality or liver transplantation (OR 0.84; 95% CrI 0.29 to 2.42). We used an uncertainty method to estimate the UDCA effect allowing for the missing data (OR = 0.89; 95% CI 0.64 to 1.25).

In meta-regression model we included quality of trials, UDCA dose, trial duration, and patients' severity of

primary biliary cirrhosis at entry as covariate and the effects of UDCA on mortality or liver transplantation as a dependent variable. No covariates seem to be significantly associated with the effect of UDCA (see Table 4).

Including data from the extended follow-up during UDCA→UDCA versus placebo/no intervention→UDCA (now comprising 146 deaths or liver transplantations in 713 patients (20.5%) originally randomised to UDCA versus 169 deaths or liver transplantations in 706 patients (23.9%) originally randomised to placebo/no intervention) demonstrated an OR of 0.81 with 95% CI from 0.62 to 1.05.

Liver transplantation (Comparison 01-03 & 04-03)
Combining the results of 14 RCTs demonstrated no significant effects on liver transplantation favouring UDCA (OR = 0.80; 95% CI 0.50 to 1.29). In the UDCA group 34/699 (5.0%) patients had liver transplantion as versus 41/692 (5.9%) patients in the control group.

Including data from the extended follow-up during UDCA→UDCA versus placebo/no intervention→UDCA (now comprising 66 liver transplantations in 699 patients (9.4%) originally randomised to UDCA versus 89 deaths or liver transplantations in 692 patients (12.9%) originally randomised to placebo/no intervention) demonstrated an OR of 0.70 with 95% CI from 0.50 to 0.98 (Comparison 04-03).

Pruritus, fatigue, and jaundice (Comparison 01-04 to 01-07)
UDCA did not significantly influence either the number of patients with pruritus (OR = 0.93; 95% CI 0.63 to 1.39, 5 trials) or the pruritus score (WMD = -0.20, 95% CI -0.44 to 0.05, 3 trials). Fatigue was not significantly influenced by UDCA (OR = 0.76; 95% CI 0.49 to 1.17, 3 trials). The two trials (TOKYO; VILLEJUIF) reporting the number of patients with jaundice observed a significant (P = 0.02) effect of UDCA (OR = 0.32; 95% CI 0.12 to 0.87).

Other clinical symptoms (Comparison 01-08 to 01-13)
In most of the trials information on autoimmune conditions was sparse. However, the MAYO-I trial (Zukowski 1998) evaluated the autoimmune conditions during UDCA and placebo period and did not find any significant effect of UDCA on associated sicca syndrome, Raynaud's phenomenon, arthritis, or Hashimoto's thyroiditis - neither on disappearance of conditions present at entry nor acquisition of new conditions.

Neither portal pressure (weighted mean difference (WMD) = 0.8 mmHg; 95% CI -2.2 to 3.8 mmHg, 1 trial), number of patients with development of varices (OR = 0.54; 95% CI 0.25 to 1.19, 3 trials), number of patients with bleeding varices (OR = 0.53; 95% CI 0.20 to 1.43, 4 trials) nor patients developing hepatic encephalopathy (OR = 0.38; 95% CI 0.06 to 2.60, 2 trials) were significantly affected by UDCA intervention. However, the number of patients developing ascites was significantly (P = 0.02) lower in the UDCA group compared to the control group (OR = 0.41; 95% CI 0.18 to 0.92).

Liver biochemistry (Comparison 01-14 to 01-22)
UDCA intervention led to a significant improvement in:
s-bilirubin WMD (95%CI) = -10.3 µmol/l (-15.5 to -5.1); P < 0.001, 6 trials - corresponding to a decrease compared to the control group of about 25%;
s-alkaline phosphatases WMD (95% CI Random) = 359.1 international units (IU)/l (-525.1 to -193.1); P < 0.001, 6 trials - corresponding to a decrease of about 40%;
s-gamma-glutamyl transpeptidase WMD (95% CI) = -257.8 IU/l (-318.3 to -197.4); P < 0.001, 4 trials - corresponding to a decrease of about 50%;
s-aspartate aminotransferase WMD (95% CI Random) = -35.5 IU/L (-53.1 to -17.8); P < 0.001, 5 trials - corresponding to a decrease of about 33%;
s-alanine aminotransferase (WMD (95% CI Random) = -47.7 IU/l (-76.9 to -18.4); P < 0.001, 5 trials - corresponding to a decrease of about 35%,
s-total cholesterol WMD (95% CI) = -0.5 mmol/l (-0.8 to -0.2); P < 0.001, 5 trials - corresponding to a decrease of about 8%; and
plasma immunoglobulin M WMD (95% CI) = -1.3 g/l (-1.9 to -0.6); P < 0.001, 4 trials - corresponding to a decrease of about 24%.

Only one trial reported s-albumin concentrations (MILAN) and one on prothrombin index (VILLEJUIF). These variables were not significantly affected by UDCA intervention.

Liver histology (Comparison 01-23 to 01-25)
There were no significant effects of UDCA on either worsening of histological stage (OR = 0.65, 95% CI 0.28 to 1.50, random, 5 trials) or worsening of fibrosis (OR = 0.82; 95% CI 0.41 to 1.65, 1 trial), or florid duct lesions (OR = 0.80, 95% CI 0.32 to 2.02, 1 trial). About half of the patients entered into the BARCELONA trial observed significant improvements in the UDCA group versus the placebo group in histological stage, portal inflammation, piecemeal necroses, but no significant effects on ductular proliferation or cholestasis (Comparison 01-26). Further, the placebo group had significantly fewer bile ducts per portal tract .

Quality of life
None of the trials examined specific quality-of-life scales. Two trials (NEWCASTLE; GÖTEBORG) evaluated symptoms using visual analogue scales. None of these showed any significant difference between the UDCA group and placebo group. However, significantly (P<0.01) more patients felt better or much better following UDCA intervention than after placebo in the GÖTEBORG-trial.

Adverse events (Comparison 02-01 & 02-02)
Only the MILAN trial reported one serious adverse event. Other trials reported non-serious adverse events. It seems that using UDCA led to a higher incidence of adverse events (OR = 1.74; 95% CI 1.10 to 2.75, 11 trials) comparing to placebo or no intervention, mainly weight gain.

Publication bias and other biases
Neither the Egger's nor the Begg's graphs and their corresponding tests on mortality provided evidence for asymmetry (Egger's test, P = 0.47; Begg's test, P = 0.83)

Discussion

This review included 16 randomised clinical trials assessing the effects of UDCA against placebo or no intervention for patients with primary biliary cirrhosis. Integrating with updated data since 2001to December 2005, this systematic review did not demonstrate any benefit of UDCA on mortality and mortality or liver transplantation. It confirms and extends the main findings of Goulis et al. and Gluud et al. (Goulis 1999; Gluud 2001 b). Furthermore, the effects of UDCA on mortality seem to associate with trial duration and disease severity: the longer the trial, the less effects of UDCA, if any; the more severe the patients, the more effects of UDCA, if any. There has been no updated data on liver biochemistry since 2001, and this review repeatedly showed a reduction in liver biochemistry, jaundice, and ascites following UDCA intervention. The use of UDCA is associated with weight gain in patients with primary biliary cirrhosis.

There was no significant funnel plot asymmetry, and no statistical signs of publication bias or other biases. However, this review analysed 15 trials involving 1447 patients. This is a low number of patients (Ioannidis 2001). The median length of trial duration was 24 months. This is not sufficiently long considering that the estimated median survival of a patient with primary biliary cirrhosis is 10 to 15 years (Prince 2002). Therefore, it is difficult to detect a significant difference on mortality based on the trials, most of which are under-powered. Further, over half of the trials had high risk of bias in terms of methodological quality. Generally, high-risk trials overestimate intervention effects (Schulz 1995; Moher 1998; Kjaergard 2001). If the same overestimation is valid for the present sample of trials, the prospects for UDCA in primary biliary cirrhosis look even worse.

This systematic review did not demonstrate a benefit favouring UDCA on our pre-defined primary outcomes: mortality and mortality or liver transplantation, neither in the period in which patients were treated with UDCA or placebo/no intervention nor in the later period in which all the patients were treated with open label UDCA. This observation is in contrast to some previous attempts to aggregate data from studies assessing UDCA interventions for primary biliary cirrhosis (Simko 1994; Poupon 1997; Poupon 2000). However, Simko et al. (Simko 1994) included non-randomised studies in their meta-analysis, which are more liable to bias. Poupon et

al. (Poupon 1997) only included three randomised clinical trials in their analysis and Poupon (Poupon 2000) only included five randomised clinical trials in their meta-analysis. Such meta-analyses largely run the risk of trial selection bias (Gluud 2001 a).

Our findings using classical meta-analytic approach are consistent with the results using Bayesian approach as sensitivity analyses. Bayesian approach can make probability statements regarding quantities of interest, eg, the probability that patients receiving drug A have better outcome than those receiving drug B. In our review, the 95% Bayesian credibility intervals, representing the effects of UDCA on mortality and mortality or liver transplantation, both cover one, ie, the null intervention effects. Therefore, it is not possible to conclude the benefits of UDCA on these two major clinical outcomes.

We used Bayesian approach to make predictive statements, conditional on the evidence from the 14 trials included. UDCA effects on mortality in a new trial has been predicted as OR 0.89 with 95% CrI from 0.27 to 2.69, meaning that UDCA may decrease or increase the risk of mortality in a new trial with 'average' size of the 14 trials. Given the evidence from the 15 trials, UDCA effects on mortality or liver transplantation in a new trial has also been predicted: OR 0.84 with 95% CrI from 0.29 to 2.42, meaning that UDCA may decrease or increase the risk of mortality or liver transplantation in a new trial with 'average' size of the 15 trials.

A common criticism about meta-analyses is that they combine information from trials with very different patient characteristics and designs, regarded as sources of heterogeneity. In our review, the percentage of patients with advanced histological stages or symptoms at trial entry varies from 15% to 83% with a median of 51%. Therefore, it is justified to estimate 'true' UDCA effect after adjusting for trial-level covariates. One of the important trial-level covariates is 'underlying risk'. The underlying risk here reflects the risks of the two primary outcomes (ie, mortality and mortality or liver transplantation) for a patient given placebo or no intervention or before given intervention. It indicates the average risk of a patient in that trial if she or he was not treated by UDCA. The 'true' UDCA effect on mortality after adjusting the underlying risk, by using Bayesian approach, is estimated as OR 0.82 with 95% CrI 0.43 to 1.51; the 'true' UDCA effect on mortality or liver transplantation after adjusting underlying risk is estimated as OR 0.77 with 95% CrI 0.43 to 1.37. These results could be interpreted that suppose the 15 trials have the same underlying risks (for example the same percentage of patients with advanced histological stages or symptoms at trial entry), it is impossible to conclude the benefits of UDCA on mortality and mortality or liver transplantation.

We also considered other important and pre-defined trial-level covariates, ie, quality of trial, UDCA dose, trial duration, and patients' severity of primary biliary cirrhosis. We used classical meta-regression model to examine whether and how UDCA effect is associated with these characteristics. It showed that UDCA effect may associate with trial duration and patients' disease severity: the longer the trial, the less effects of UDCA, if any; the more severe the patients, the more effects of UDCA, if any. Heterogeneity across the included trials can be largely explained by these two covariates. The relationship between UDCA effect and trial duration is also supported by Bayesian meta-regression, which included 'trial duration' as covariate. The posterior marginal median coefficient for 'trial duration' was 0.03 with 95% CrI from 0.01 to 0.05 - therefore, the longer the trial, the less chance to detect effect favouring UDCA.

The two previous meta-analyses (Goulis 1999; Gluud 2001 b) were mainly criticised for including many trials of only two-year duration and the studies included were very heterogeneous regarding length of follow-up (Talwalker 2003; Kaplan 2005;). Survival analyses in a disease with a very long natural history over decades ideally are based on longer follow-up periods. So, based on observational studies, benefits of long-term UDCA intervention has been suggested (Rust 2005; Pares 2006). However, given the updated evidence and analyses on data from longer follow-up, our review seems not supportive of long-term UDCA intervention because it seems less possible to detect survival benefit in longer-term UDCA intervention. Further, the Bayesian meta-analyses estimated that UDCA effect, allowing for different length of trial duration and the above mentioned underlying risk, has been consistent with unadjusted pooled results (OR 0.71, 95% CrI 0.39 to 1.29 vs. OR 0.89, 95% CrI 0.50 to 1.49) - impossible to confirm a benefit of UDCA on mortality even if the trials have the same duration and underlying risk.

The relationship between UDCA effect and patients' severity of primary biliary cirrhosis was indicated in the classical meta-regression, meaning that UDCA effect, if any, is more likely to be observed in more severe patients. This indication is supported by an analysis combining the raw data of three large clinical trials, in which the survival benefit of UDCA was observed in patients with moderate-to-severe disease but not in those with mild disease (Poupon 1997). The relationship between UDCA effect and severity of disease is, however, not repeated in our Bayesian meta-regression, which included 'severity' as covariate. The posterior marginal median was -0.67 with 95% CrI from -4.26 to 2.75. Therefore, whether UDCA intervention is related to severity of primary biliary cirrhosis should be further investigated. Despite of the uncertainty, it is estimated that UDCA effect, allowing for different levels of disease severity and the above mentioned underlying risk, is consistent with the unadjusted pooled results (OR 0.80, 95% CrI 0.43 to 1.46 vs. OR 0.89, 95% CrI 0.50 to 1.49) - impossible to confirm a benefit of UDCA on mortality even if the trials have same level of disease severity and underlying risk.

We observed a marginally significant effect of UDCA on liver transplantation only in the later period in which all the patients were treated with open label UDCA, but not in the original period in which patients were treated with UDCA or placebo/no intervention. The decision of whether and when to perform liver transplantation is influenced by many factors: the attitude of the patient, the attitude of the physician, the time of referral, the length of the waiting list, etc. Therefore, liver transplantation is an imprecise measure of the stage of progression of the disease and thus most likely a biased outcome. The fact that liver biochemical outcomes improved in the UDCA group compared to the placebo treated may lead to the observation of fewer liver transplants in the UDCA group. For example, s-bilirubin is one of prognostic indices used for patients with primary biliary cirrhosis (Pasha 1997). A lower s-bilirubin will provide the clinicians with less impetus to transplant. Second, the referrals for liver transplantation occurred mainly after the blinding of the randomised clinical trials had been removed. Unblinded comparisons may exaggerate intervention efficacy significantly (Schulz 1995; Kjaergard 2001). Therefore, whether UDCA decreases the risk of liver transplantation should be confirmed in further research.

We noticed that the number of patients with ascites was significantly less in the UDCA group than in the placebo group. But this is only observed in four trials. Whether this observation is due to a play of chance cannot be excluded, considering that the many comparisons have been made without correction of the significance level. Further, the diagnosis of ascites was clinically based, which is more susceptible to bias. Moreover, in our review, UDCA has not been found to decrease portal pressure and s-albumin, which are important in the pathogenesis of ascites. So whether this observation could be generalised externally should be further investigated.

It is interesting to know if UDCA could slow the histological progression towards more advanced stages. In this review, we were not able to identify any convincible benefits of UDCA on histology. Only one trial found significant effects on liver histology (BARCELONA). It observed positive effects on a number of histological variables, including the histological stage. This finding may be a spurious one, however. Only about half of the randomised patients had a follow-up liver biopsy. Further, as the trial showed a trend towards a higher mortality and liver transplantation rate in the UDCA group, this could have removed some of the more seriously affected livers from the UDCA group, making those having a biopsy look relatively less affected. Such subgroup results should be interpreted cautiously (Yusuf 1991; Oxman 1992; Assmann 2000). On the other hand, the findings of the BARCELONA-trial are interesting and should stimulate more research into the effect of UDCA on progression of fibrosis in primary biliary cirrhosis and eventually cirrhosis development.

UDCA intervention is found to be associated with non-serious adverse events, mostly weight gain. The finding is mainly due to new data from MAYO-I trial. The authors suggest that discussions with patients beginning UDCA should mention weight gain as a possible side effect. Other non-serious adverse events include mild gastrointestinal disorders like diarrhoea, nausea, vomiting, etc. Generally, UDCA is well tolerated to patients with primary biliary cirrhosis.

It has been claimed that UDCA is a cost-effective therapy for primary biliary cirrhosis (Pasha 1999). However, this cost-effectiveness rests on extrapolation from the results of two selected randomised clinical trials (MAYO-I; TORONTO). It is evident that cost-effectiveness analyses ought to be performed on the basis of all available evidence and not just on selected evidence. Considering the annual cost of UDCA of about $2500 (Pasha 1999) and the findings of the present review, we challenge the conclusion drawn by Pasha et al. (Pasha 1999) that UDCA is cost-effective for primary biliary cirrhosis.

UDCA improved most biochemical outcomes and patients' ascites. Also, UDCA appears well tolerated, although it might be associated with weight gain. However, consistent with the previous two meta-analyses (Goulis 1999; Gluud 2001 b), this updated systematic review did not demonstrate any benefit of UDCA on mortality and mortality or liver transplantation in patients with primary biliary cirrhosis. It extends the main findings of Goulis et al. and Gluud et al. (Goulis 1999; Gluud 2001 b) in that the effect of UDCA intervention seems to associate with trial duration and disease severity. This seems to place clinicians and researchers in a dilemma: if therapeutic decisions are based on clinical outcomes (eg, mortality), they have insufficient evidence to support the use of UDCA in primary biliary cirrhosis; but if based on 'surrogate outcomes' (eg, s-bilirubin level), they have evidence favouring the UDCA interventions for the disease. It is true that the use of surrogate outcomes is particularly attractive for studies of complex chronic disease, like primary biliary cirrhosis, since occurrence of the clinical outcome, eg, survival may take 10 to 15 years from the onset of disease. However, it is precisely because of the complexity of the disease that assessment of potential surrogate outcomes is so difficult. Mayo Risk Score Model has identified several prognostic biomarkers for primary biliary cirrhosis, for example, serum bilirubin. Those biomarkers may respond to intervention and are predictive of survival. But they do not necessarily predict clinical benefit of intervention because 'a perfect correlation does not a surrogate make'. In the absence of validated surrogate outcomes in UDCA intervention for primary biliary cirrhosis, confirmatory trials assessing the UDCA effect should only be based on clinical outcomes, eg, survival.

We also realized, when we doing the review, that the challenge of performing a new trial on interventions for primary biliary cirrhosis is high. As mentioned before, the disease's estimated median survival is about 10 to 15 years. To spend 15 years planning and carrying out a trial for each new potential treatment for primary biliary cirrhosis would consume many patients' lifetimes, not to mention the expense and difficulty of retaining patients in such a long study (Mayo 2005). This is an unacceptably low rate of scientific progress for patients who continue to die or need liver transplantation because of primary biliary cirrhosis. We agree with Mayo that 1) integration of international groups of investigators for primary biliary cirrhosis will make large study sizes feasible; 2) 'Development of sensitive and specific markers of disease severity' (e.g., using newer methodologies that use computerised pattern recognition; and non-invasive assessment of disease progression: radiology or by serum test), which facilitates; 3) full validation of surrogate outcome(s) for a given intervention of primary biliary cirrhosis before it can substitute the clinical outcomes.

Authors' conclusions

Implications for practice

UDCA improves liver biochemical variables, including s-bilirubin concentration, jaundice, and ascites in patients with primary biliary cirrhosis. However, this updated review confirms and extends previous observations showing no benefit of UDCA on patients' mortality and mortality or liver transplantation. This review does not support long-term use of UDCA. UDCA has few serious adverse events but is associated with weight gain.

Implications for research

It is less likely to find any benefit of UDCA on patients' survivals in a new trial with the average size of the included trials. Integration of international groups of investigation for primary biliary cirrhosis will make large study sizes feasible. Full validation of surrogate outcome(s) is justified. In the present absence of validated

surrogate outcome(s), trial assessing UDCA or any new potential treatment for primary biliary cirrhosis should only be based on clinical outcomes, eg, survival.

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Contribution of authors

YG made searches, identified trials with updated information, performed statistical analyses, drafted the review; ZBH performed a part of statitical analyses; EC and CG validated selection of studies as well as reviewed the article.

Declarations of interest

None known.

Differences between protocol and review

Published notes

This is an updated systematic review to the Gluud et al (Gluud 2001 b).

Characteristics of studies

Characteristics of included studies

ATHENS

Methods	Generation of allocation schedule: adequate, random table numbers. Allocation concealment: adequate, serially numbered sealed envelops. Blinding: no blinding. Follow-up: no patients lost to follow-up.
Participants	Patients with symptomatic PBC (n = 86) from one centre in Greece. PBC defined as: cholestatic liver disease, positive AMA, liver biopsy compatible with PBC. Exclusion criteria were: asymtomatic PBC, hepatic encephalopathy, sepsis, renal failure, or life-threatening disease.
Interventions	Control: no intervention. Experimental: UDCA 12-15 mg/kg/day.
Outcomes	Liver decompensation. Mortality or liver transplantation. Symptoms. Liver biochemistry. Liver histology.
	14/43 control patients were crossed-over to UDCA at their own request at a

Notes	median of 3.5 years (range 2-8 years) after entry in the study. The authors did both intention-to-treat analysis and treatment-as-received analysis.

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Yes	A - Adequate

BARCELONA

Methods	Generation of allocation scedule: adequate. Allocation concealment: sealed envelopes (no mention on serial numbering or opaqueness). Blinding: placebo - identical in appearance, smell, and taste. Follow-up: 10 UDCA treated patients and 21 placebo treated patients discontinued.
Participants	Consecutive patients with PBC (compatible liver biopsy, alkaline phosphatases >2 upper normal limit and positive or negative antimitochondrial antibodies; n = 192) from 16 centres in Spain. Patients with negative antimitochondrial antibodies were accepted if there was no evidence of extrahepatic biliary obstruction.
Interventions	Control: placebo. Experimental: UDCA 14-16 mg/kg/day in three divided doses.
Outcomes	Mortality. Liver transplantation. Symptoms. Complications. Liver biochemistry. Liver histology.
Notes	

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Yes	A - Adequate

DALLAS

Methods	Generation of allocation scedule: no data. Allocation concealment: no data, but randomisation was separate at each of the six centres in four stratification groups, involving serum bilirubin level and liver histology stage. Blinding: described as double blind, but placebo only descibed as 'comparable-appearing' and no mention on smell and taste. Follow-up: 2 patients receiving UDCA and 3 placebo withdrew from the trial duing

	placebo controlled period (0-2 year).
Participants	Patients with PBC (n = 151) from six USA centres. Entry criteria were: cholestatic liver disease for at least six months, serum alkaline phosphatases >1.5 times upper normal limit, positive AMA, no biliary obstruction, and liver biopsy compatible with PBC. Excluded were: PBC treatment during the last three months, recurrent bleeds from varices, spontaneous encephalopathy, or diuretic-resistant ascites, serum bilirubin >20 mg/l, pregnancy, age <19 years, or other cliver disease.
Interventions	Control: placebo (2 years) and open-label UDCA (4 years) Experimental: UDCA 10-12 mg/kg/day once at bedtime (Ciba-Geigy Corporation).
Outcomes	Mortality free of liver transplantation. Liver transplantation. Symptoms. Liver biochemistry. Liver histology. UDCA enrichment in bile.
Notes	Three patients randomi- sed to receive placebo had high bile-UDCA concentrations, suggesting UDCA intake. All patients were offered open label UDCA following completion of the first 2-year of the trial.

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Unclear	B - Unclear

FRANKFURT

Methods	Generation of allocation schedule: adequate. Allocation concealment: adequate. Blinding: placebo identical in appearance, smell, and taste.
Participants	Patients with PBC (n = 20) from Germany. PBC defined as at least three of the following: alkaline phosphatases >1.7 times upper normal limit, gamma-glutamyl transferase >5.0 times upper normal limit, Immunoglobulin M > 2.0 times upper normal limit, positive AMA plus no obstruction of the extrahepatic biliary tract. Exclusion criteria were: oesophageal varices, pancreatitis, cardiac failure, renal failure, pregnancy, age <03 years, PBC treatment within the previous four weeks, and alcohol or drug abuse.
Interventions	Control: placebo. Experimental: UDCA 10 mg/kg/day, divided into two doses.
Outcomes	Mortality. Symptoms.

Outcomes	Liver biochemistry. Liver histology.
Notes	

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Yes	A - Adequate

GÖTEBORG

Methods	Generation of allocation schedule: adequate. Allocation concealment: adequate, sealed envelopes. Patients were stratified into symptomatic/asymptomatic Blinding: described as 'double-blind', and placebo looked identical to UDCA, but details on taste and smell not given. Follow-up: 8 patients receving UDCA and 7 placebo withdrew.
Participants	Patients with PBC (n = 116) from six centres in Sweden. PBC defined as: chronic cholestatic liver disease of more than six months' duration with histology typical of or compatible with PBC plus at least two of the following: positive anti-mitochodrial antibodies, alkaline phosphatases >1.5 times the upper reference value, and/or IgM >1.5 times the upper reference value during the year preceding the entry into the study.
Interventions	Control: placebo. Experimental: 500 mg UDCA (~7.7 mg/kg/day).
Outcomes	Mortality. Liver transplantation. Symptoms - pruritus, fatigue, ascites, jaundice. Liver biochemistry and bile acids. Histology - portal inflammation, spill-over, interface hepatitis, bile duct proliferation, portal fibrosis. Quality of life.
Notes	At 24 months, 32 of 49 patients allocated to placebo and still remaining in the study were switched to UDCA and 42 of 52 patients allocated to UDCA and still remaining in the study continued with UDCA. Anti-hepatitis C virus tests not performed.

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Yes	A - Adequate

HELSINKI

Methods	Generation of allocation schedule: adequate, random numbers in blocks of six. Allocation concealment: adequate, central. Patients were 'randomly stratified according to bilirubin' to intervention arm. Blinding: placebo identical looking and film-coated (considered adequate). Follow-up: np patients receiving UDCA and 8 placebo withdrew.
Participants	Patients with PBC (n = 90) from four centres in Finland. PBC defined as: elevated alkaline phosphatases, liver biopsy compatible with PBC, and positive AMA. End-stage PBC and patients treated with drugs that might affect prognosis were excluded.
Interventions	Control: placebo. Experimental 1: UDCA 12-15 mg/kg/day in two doses. Experimental 2: colchicine 1 mg/day.
Outcomes	Death. Liver transplantation. Symptoms. Liver biochemistry. Liver histology.
Notes	

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Yes	A - Adequate

MANCHESTER

Methods	Randomisation: information being sought - desribed as randomised. Blinding: 'placebo' employed, but it is not known if it was indeed double blind. Follow-up: not described.
Participants	Patients with PBC (n = 28) form UK. Diagnostic criteria (data being sought).
Interventions	Control: placebo. Experimental 1: UDCA 10mg/kg/day. Experimental 2: colchicine 1 mg/day. Experimental 3: UDCA plus colchicine.
Outcomes	Mortality (being sought) Liver transplantation (being sought). Serum aspartate aminotransferase, alanine aminotransferase, gamma-glutamyl transpeptidase, bilirubin and albumin. Serum alkaline phosphatases. Serum procollagen peptide. Galactose elimination capacity. Bromosulfophtalin excretion.

Notes	No exact data on number of patients randomised to each arm. No data given seperatedly on mortality and liver transplantation. Information being sought.

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	No	C - Inadequate

MAYO-I

Methods	Generation of the allocation schedule: adequate, computer. Allocation concealment: adequate, patients stratified for centre, histological stage, serum bilirubin, and esophageal varices using 'a blocked, randomised assignment schedule'. Blinding: 'double-blind, and placebo looked and smelled identical to UDCA, but placebo was sweet and UDCA bitter. However, only one patient broke the code. Follow-up: five voluntary withdrawals in UDCA arm and 13 voluntary withdrawals in the placebo arm.
Participants	Patients with PBC (n = 180) enrolled from four USA centres. However, 162 patients (90%) came from one centre. PBC defined as: chronic cholestatic liver disease for at least six months, a serum alkaline phosphatases level >1.5 times upper normal limit, antimitochondrial antibody positivity, absence of biliary obstruction, and liver biopsy compatible with PBC. Excluded were: PBC-drug treatment in preceeding 3 months, anticipated need for liver transplantation within one year, recurrent variceal hemorrhage, spontaneous encephalopaty, or diuretic resistant ascites, pregnancy, age <18 or >70 years, or other coexistent liver disease.
Interventions	Control: placebo. Experimental: UDCA at a dose of 13-15mg/kg/day in four diveded doses.
Outcomes	Composite end point consisting of death, transplant, toxicity, and voluntary withdrawal. Death. Liver transplantation. Symptoms. Autoimmune conditions. Liver biochemistry. Liver histology. Adverse events, including weight gain.
Notes	Patients originally receiving placebo switched to UDCA after four years and followed for an andditional eight years..

Risk-of-bias table

Item	Judgment	Description

Allocation concealment?	Yes	A - Adequate

MEXICO CITY

Methods	Randomisation: no data. Allocation of concealment: unclear. Blinding: 'placebo' used.
Participants	Patients with PBC (n = 28) from one centre in Mexico.
Interventions	Control: placebo. Experimental: UDCA (data being sought).
Outcomes	Serum cholesterol.
Notes	

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Unclear	B - Unclear

MILAN

Methods	Generation of allocation schedule: adequate, patients were randomised by each center according to a computer generated list. Allocation concealment: no data. Blinding: described as double-blind, and placebo was 'identical in appearance', but smell and taste not mentioned. Follow-up: 5 patients receiving UDCA and 1 placebo dropped out.
Participants	Patients with PBC (n = 88) from seven centres in Italy. PBC defined as: positive AMA and liver bioposy compatible with PBC. If one of these were missing, patients could enter provided they had three of the following: seum alkaline phosphatases >2.0 times upper normal limit, immuneglobulin M >280 mg/l, pruritus, serum bilirubin > 2 mg/l, and/or a positive Schyrimer's test plus absence of extrahepatic obstruction.
Interventions	Control: placebo. Experimental: UDCA 500 mg daily in two dived doses at mealtime (~8.7 mg/kg/day; range 5.4-11-6 mg/kg/day).
Outcomes	Symptoms. Liver biochemistry. Serum bile acids. Serum cholesterol.
Notes	Patients switched onto UDCA at the end of the trial.

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Unclear	B - Unclear

NEWARK-II

Methods	Generation of randomisation schedule: no data. Allocation concealment: no data. Blinding: described as double-blind, but no mention of appearance, smell, and taste. Follow-up: no patients withdrew.
Participants	Patients with PBC (n = 19) enrolled from one centre in USA. Inclusion criteria: PBC confirmed by liver biopsy and supporting clinical tests. Exclusion criteria: extrahepatic biliary obstruction.
Interventions	Control: placebo. Experimental: UDCA 10 mg/kg/day.
Outcomes	Mortality. Symptoms. Liver biochemistry.
Notes	

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Unclear	B - Unclear

NEWCASTLE

Methods	Generation of allocation schedule: adequate, based on a list of random numbers. Allocation concealment: adequate, patients were enterd into the trial in pairs according to clinical stratification. Sealed envolopes were kept and opened by the pharmacy once a pair of matching patients were identified indicating 'treatment A' for one patient and 'treatment B' for the other. Blinding: placebo 'identical looking', but was neither matched for taste nor smell.
Participants	Patients with PBC (n = 46) from one centre in UK. PBC defined as: clinically and histologically compatible with PBC, positive AMA, abnormal liver function tests, and no medication wihtin six months of study entry.
Interventions	Control: placebo. Experimental: UDCA ~10mg/kg/day (mean actual dose (+/-SD): 11.4+/-0.9 mg/kg/day.
	Mortality.

Outcomes	Liver transplantation. Symptoms. Liver biochemistry. Liver histology. Quality of life.
Notes	

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Yes	A - Adequate

TAIPEI

Methods	Generation of randomisation schedule: adequate, table of random numbers. Allocation concealment: no data. Blinding: described as double-blind, and placebo and UDCA were identical looking, but no data on smell and taste. Follow-up: no patients withdrew.
Participants	Patients with PBC (n = 12) from one centre in Taiwan. PBC defined as: elevated serum alkaline phosphatases and gamma-glutamyl transferase with lack of large bile duct abnormalities, positive AMA, with elevated immunoglobulin M, G or A, and liver biopsy compatibale with PBC. Exclusion criteria were: previous PBC treatment.
Interventions	Control: placebo. Experimental 1: UDCA 12-15 mg/kg/day in two doses. Experimental 2: colchicine 1 mg/day.
Outcomes	Mortality. Symptoms. Liver biochemistry.
Notes	All patients swiched to UDCA on completion of the six months cross-over trial.

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Unclear	B - Unclear

TOKYO

Methods	Generation of allocation schedule: no data. Allocation concealment: adequate, allocation by a single monitor according to a randomisation scheme (1:1). Blinding: UDCA and placebo with identical appearance (size and color), but taste and smell not mentioned.

	Follow-up: 4 patients receiving UDCA and 3 placebo dropped out.
Participants	Patients with PBC (n = 49) from 13 departments in Japan. PBC was diagnosed clinically and histologically. Patients with severe symptoms or having received other medications for their PBC within the last three months were excluded. Placebo female/male: 20/4. UDCA female/male: 24/1.
Interventions	Control: placebo. Experimental: UDCA
Outcomes	Symptoms (itching). Complications (oesophageal varices). Liver biochemistry. Serum cholesterol. Serum bile acids.
Notes	

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Yes	A - Adequate

TORONTO

Methods	Generation of allocation schedule: no data. Concealment of allocation: adequate, separately at each center by the study pharmacist stratified for symptomatic/asymptomatic. Blinding: described as double-blind, and the placebo tablets were identical and 'equally bitter tasting', this was confirmed by the research coordinator. Follow-up: 13 patients receiving UDCA and 19 placebo withdrew.
Participants	Of 408 patients assessed, 222 patients with PBC were randomised (1:1) during a 26 months period. Inclusion criteria were: positive AMA, serum alkaline phosphatases >1.0 times upper normal limit, liver biopsy compatible with PBC, and age >18 years. Patients were excluded if they were on liver transplant list, needed to take enzyme-inducing drugs, were pregnant, or had a severe coexisting condition that was likely to affect survival within five years of study entry.
Interventions	Control: placebo. Experimental: UDCA 14mg/kg/day swallowed with the evening meal.
Outcomes	Mortality. Liver transplantation. Symptoms - pruritus, fatique. Liver biochemistry and bile acids. Histology.

Notes	Patients offered UDCA at the end of the trial.

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Yes	A - Adequate

VILLEJUIF

Methods	Generation of allocation schedule: no data. Allocation concealment: adequate, 'patients were randomised by each center in blocks of four to drug package containing UDCA or placebo capsules'. Blinding: described as double-blind, and placebo was 'identical in appearance', but smell and taste are not mentioned. Placebo was made of starch and lactose. Follow-up: 5 patients receving UDCA and 6 placebo withdrew
Participants	Patients with PBC (n = 146) from 22 centres in France and Canada. PBC defined as: liver biopsy compatible with PBC, serum alkaline phosphatases >2.0 upper normal limit, and positive AMA. Exclusion criteia were: PBC treatment within last sixmonths, serum bilirubin >150 µmol/l, serum albumin <25 g/l, past or active bleeding oesophageal varices, extrahepatic obstruction, excessive alcohol consumption, or positive hepatitis B surface antigen.
Interventions	Control: placebo. Experimental: UDCA 13-15 mg/kg/day.
Outcomes	Mortality. Liver transplantation. Symptoms. Liver biochemistry. Liver histology.
Notes	All patients treated for two years with placebo were offered UDCA and further followed-up for another twoyears together with patients continuing on UDCA. One patient, included in the publications of the study up to 1993, was excluded from the 1994 publication due to a raised serum bilirubin at entry, violating the entry criteria.

Risk-of-bias table

Item	Judgment	Description
Allocation concealment?	Yes	A - Adequate

Footnotes

Characteristics of excluded studies

Angulo 1999

Reason for exclusion	This is not a randomised trial, but a comparison of liver histology of 16 UDCA trated patients from one RCT to the liver histology of 51 patients from another RCT.

Angulo 1999 a

Reason for exclusion	There is no placebo or no intervention group in this RCT, which compares low- (5-7 mg/kg/day), standard- (13-15 mg/kg/day), and high- (23-25 mg/kg/day) doses of UDCA in 155 patients with PBC. The improvements in alkaline phosphatases, aspartate aminotransferase, Mayo risk score, and biliary UDCA enrichment were significantly greater in the standard- and high-dose groups compared to the low-dose group, but not between the standard- and high-dose group. No significant effects were noted on symptoms with any dose.

Bateson 1998

Reason for exclusion	This is a case series of 40 PBC patients with symptomatic disease treated with UDCA. The results were compared to 12 historic UDCA-untreated PBC patients.

Brodanova 1997

Reason for exclusion	This is a case series of 13 PBC patients treated with UDCA.

Cauch-Dudek 1998

Reason for exclusion	This is a case series of 88 patients with PBC evaluating fatigue. A self rated fatigue. Severity score did not correlate with UDCA use.

Crippa 1995

Reason for exclusion	The study is not randomised, but compares 18 UDCA treated PBC patients to eight untreated PBC patients.

Crosignani 1996

Reason for exclusion	This is a dose-response study examining the effects of three doses of tauro-UDCA in 24 patients with PBC.

Eisenburg 1988

Reason for exclusion	This is a case series of 21 PBC patients during UDCA administration.

Ferri 1993

Reason for exclusion	This is a controlled comparison of UDCA with tauro-UDCA for PBC.

Grippa 1995

Reason for exclusion	This is a non-randomised study comparing 18 UDCA treated PBC patients to eight UDCA-untreated PBC patients.

Ideo 1990

Reason for exclusion	Out of three PBC patients treated with UDCA (600 mg/day), UDCA was stopped in one of these patiens 'randomly selected'.

Ikeda 1996

Reason for exclusion	This is a randomised trial comparing UDCA plus colchicine versus UDCA alone in 22 patients with PBC.

Kehagioglou 1991

Reason for exclusion	The study is not described as randomised, but compares 16 PBC patients treated with UDCA (14 mg/kg/day for a mean period of 22 months (range 3-35 months) to a control group consisting of 10 PBC patients treated with placebo.

Kim 1997

Reason for exclusion	This is a case series of eight UDCA-treated PBC patiens who lacked antimitochondrial antibodies.

Kneppelhout 1992

Reason for exclusion	This is a case series of 19 patients with PBC during UDCA administration.

Krzeski 1999

Reason for exclusion	This is a case series of 60 PBC patients treated with UDCA.

Larghi 1997

Reason for exclusion	This is a randomised trial with crossover design comparing UDCA versus tauro-UDCA.

Leuschner 1996

Reason for exclusion	This randomised trial compared UDCA plus prednisolone versus UDCA plus placebo for PBC.

LONDON 1998

	This trial compared placebo to different doses of URSO (300 mg/day, 600 mg/day, 900 mg/day and 1200 mg/day) in 23 biopsy proven early stage PBC patients. There is no mention of randomisation. Patients were followed for eight

Reason for exclusion	weeks with a four week washout period between doses. A significant trend toward normalising of abnormal liver function tests was observed together with a significant increase in lethargy, irrespective of UDCA dose, compared to placebo.

Lotterer 1990

Reason for exclusion	This is a case series of seven PBC patients during UDCA administration.

Matsuzaka 1994

Reason for exclusion	This is a case series of three PBC patients during UDCA administration.

Matsuzaki 1990

Reason for exclusion	This is a case series of ten PBC patients during UDCA administration.

MAYO-II 1997

Reason for exclusion	This trial randomised 150 PBC patients to three doses of UDCA (5-7 mg/kg/day; 13-15 mg/kg/day; 22-25 mg/kg/day) and followed the patients for one year. No differences were observed between the medium and the high dose with respect to liver biochemistry changes, buth both these dose groups had significantly greater improvement of liver biochemistry compared to the low dose group. Clinical events such as death, transplantation, or complications of liver disease were rare and were not differnt between the three dose groups.

NEWARK-I

Reason for exclusion	The study is not randomised. The study included only four patients with PBC and apparently these were treated first with placebo for three months and then with UDCA (10-15 mg/kg/day) for three-six months. No major outcome variables are reported.

NEWARK-III

Reason for exclusion	This study investigated biochemical features, including biliary bile acids, in 14 patients with PBC using a paired design. First, all patients received placebo for three months. Then, the patients were treated with 900 mg UDCA (10-12 mg/kg/day) for six months (n=11) to 12 months (n=8). The latter patients were then treated with placebo for three months and restarted on UDCA for another 12 months. Due to the paired design the observed improvements may be due to the fluctuating course of PBC.

Ogino 1993

Reason for exclusion	This is a case series of 28 PBC patients treated with UDCA and compared to seven PBC patiens not treated with UDCA.

Okuyama 1988

Reason for exclusion	This is a study of a single PBC patient during UDCA administration.

Osuga 1989

Reason for exclusion	This is a case series of eight PBC patients during UDCA administration.

Peridigoto 1992

Reason for exclusion	This is a study of three PBC patiens during UDCA administration.

Podda 1989

Reason for exclusion	This is a randomised trial examining three doses of UDCA in PBC patients and patients with primary sclerosing cholangitis and chroinic hepatitis.

Poupon 1987

Reason for exclusion	This is a case series of 15 PBC patients during UDCA administration.

Poupon 1989

Reason for exclusion	This study is not randomised.

Poupon 1996

Reason for exclusion	This is a randomised trial comparing UDCA plus colchicine versus UDCA in 74 patients with PBC.

Schonfeld 1997

Reason for exclusion	This is a case series of 15 PBC patients during UDCA administration.

Shibata 1992

Reason for exclusion	This is a case series of 12 PBC patients during UDCA administration.

Stiehl 1990

Reason for exclusion	This is a case series of 29 patients with PBC during UDCA administration.

Taha 1994

Reason for exclusion	This is a case series of patients with PBC during different drug administrations (cholestyramine, wash out, UDCA, and UDCA plus cholestyramine).

Takezaki 1991

Reason for exclusion	This is a study of a single PBC patient during UDCA administration.

Toda 1998

Reason for exclusion	No placebo or no intervention group are included. The trial compares the efficacy of three doses of UDCA (150 mg/day; 600 mg/day; 900 mg/day) in 82 PBC patients for 24 months.

Unoura 1990

Reason for exclusion	Not a randomised trial, but compares 16 UDCA treated PBC-patients to eight patients without UDCA treatment.

Van de Meeberg 1996

Reason for exclusion	No placebo or no intervention group. Five patients treated 'in random order' with 10 mg UDCA/kg/day in either a single or in three divided doses - no difference in liver biochemistry improvement.

Van Hoogstraten 1998

Reason for exclusion	This RCT compares 10 versus 20 mg UDCA/kg/day during six months in 61 PBC patients. Liver biochemistry improved in PBC patients receiving 20 mg/kg/day compared to a dose of 10 mg/kg/day.

Verma 1999

Reason for exclusion	This cross-over RCT compares different doses of UDCA in twenty-four biopsy-proven early-stage PBC patients (one male, 23 female), who received five doses of UDCA (0, 300, 600, 900, 1200 mg/day) each for eight weeks with four-week washout periods between doses. Symptoms (pruritus, fatigue, diarrhoea) were assessed on a four-point scale (none, mild, moderate, severe). Liver function tests were performed using conventional methods, and serum bile acids were measured using gas liquid chromatography. The dose of 900 mg/day produced the greatest enrichment of UDCA in serum bile acids; although there was no difference in the enrichment of UDCA between the different doses. There was a trend towards normalization of the abnormal LFTs in a dose-dependent manner (for y-glutamyl transferase (yGT), alkaline phosphatase (ALP), alanine transaminase (ALT) and IgM). Multi-factorial analysis showed that UDCA treatment, irrespective of dose, was significantly better than placebo for all the variables. The 900 and 1200 mg doses were better than both 300 and 600 mg using gamma-glutamyltranspeptidase and total bilirubin as variables, better than 300 mg using alkaline phosphatases and IgM as variables, and better than 600 mg using albumin as a variable. No variables showed a significant difference between 900 and 1200 mg. The study concluded that the optimum dose of UDCA is 900 mg/day (equivalent to 13.5 mg/kg/day). This trial is excluded due to the cross-over design and due to the fact that it did not provide any data on the

	primary outcome variables.

Wirth 1994

Reason for exclusion	This is a case series of 14 patients with PBC examined before and during UDCA administration.

Wirth 1995

Reason for exclusion	This is a case series of 22 patients with PBC, hwo have their subtypes of antimitochondrial antibodies examined and related to response to UDCA administration.

Wolfhagen 1994

Reason for exclusion	No randomisation, combination therapy with UDCA and prednisone in seven patients.

Yamazaki 1992

Reason for exclusion	This is a study of a single PBC patient with eosinophilic infiltration.

Yamazaki 1996

Reason for exclusion	This is a case series of 38 PBC patients, of which 55 per cent exhibited eosinophilia. The eosinophilia was reduced during UDCA treatment.

Yokomori 1996

Reason for exclusion	this is a study of a single patient with PBC and pruritus responding to treatment with UDCA and cholestyramine.

Footnotes

Characteristics of studies awaiting assessment

Footnotes

Characteristics of ongoing studies

Footnotes

Summaries of findings

Additional tables

1 Search strategy

Database	Searching period	Search term
The Cochrane Hepato-Biliary Group Controlled Trials Register	1948 to March 2006	#1= 'primary biliary cirrhosis' and 'ursodeoxycholic acid'
The Cochrane Central Register of Controlled Trials in The Cochrane Library	Issue 1, 2006	#1 = LIVER CIRRHOSIS BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = pbc #5 = #1 or #2 or #3 or #4 #6 = URSODEOXYCHOLIC ACID: MESH #7 = DEOXYCHOLIC ACID: MESH #8 = 'ursodeoxycholic acid' or 'UDCA' #9 = #6 or #7 or #8 #10 = #5 and #9
PubMed	Until March 2006	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = PBC #5 = #1 or #2 or #3 or #4 #6 = URSODEOXYCHOLIC ACID: MESH #7 = DEOXYCHOLIC ACID: MESH #8 = 'ursodeoxycholic*' or 'UDCA' #9 = deoxycholic* #10 = #6 or #7 or #8 or #9 #11 = #5 and #10 #12 = random* or placebo* or blind* or meta-analysis #13 = #11 and #12
MEDLINE	January 1966 to March 2006	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = PBC #5 = #1 or #2 or #3 or #4 #6 = URSODEOXYCHOLIC ACID: MESH #7 = DEOXYCHOLIC ACID: MESH

		#8 = 'ursodeoxycholic*' or 'UDCA' #9 = deoxycholic* #10 = #6 or #7 or #8 or #9 #11 = #5 and #10 #12 = random* or placebo* or blind* or meta-analysis #13 = #11 and #12
EMBASE	January 1980 to March 2006	#1 = PRIMARY-BILIARY-CIRRHOSIS: MESH #2 = BILIARY-CIRRHOSIS: MESH #3 = primary and biliary and cirrhosis #4 = primary biliary cirrhosis #5 = PBC #6 = #1 or #2 or #3 or #4 or #5 #7 = URSODEOXYCHOLIC ACID: MESH #8 = DEOXYCHOLIC ACID: MESH #9 = 'ursodeoxycholic*' or 'UDCA* #10 = deoxycholic* #11 = #7 or #8 or #9 or #10 #12 = #6 and #11 #13 = random* or placebo* or blind* or meta-analysis #14 = #12 and #13
Chinese Biochemical CD Database	January 1979 to March 2006	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = PBC #5 = #1 or #2 or #3 or #4 #6 = URSODEOXYCHOLIC ACID: MESH #7 = DEOXYCHOLIC ACID: MESH #8 = 'ursodeoxycholic*' or 'UDCA' #9 = deoxycholic* #10 = #6 or #7 or #8 or #9 #11 = #5 and #10 #12 = random* or placebo* or blind* or meta-analysis #13 = #11 and #12
LILACS	1982 to March 2006	#1 = (primary and biliary and cirrhosis) or (primary biliary cirrhosis) #2 = primary biliary cirrhosis #3 = ursodeoxycholic acid
SCI-EXPANDED	1945 to March 2006	Sarah can help with??

	2000	

Footnotes

2 Summary of characteristics of the included trials

Trial	Risk of bias	UDCA dose*	Trial duration	Severity of PBC#¤
ATHENS	High	13.5	92.4	0.6400
BARCELONA	Low	15.5	63.6	0.2708
DALLAS	High	11.5	24.0	0.6689
FRANKFURT	Low	10.0	9.0	0.1500
GOTEBORG	Low	7.7	24.0	0.3350
HELSINKI	Low	13.5	24.0	0.3333
MANCHESTER	High	10.0	15.0	0.3200
MAYO-I	Low	14.0	48.0	0.6833
MILAN	High	8.7	12.0	0.4950
NEWARK-II	High	10.0	6.0	0.6666
NEWCASTLE	Low	10.0	24.0	0.8261
TAIPEI	High	9.2	3.0	0.5833
TOKYO	High	9.2	6.0	0.3795
TORONTO	High	14.0	24.0	0.5270
VILLEJUIF	High	14.0	24.0	0.4658
* UDCA dose in mg/kg/day				
# PBC: primary biliary cirrhosis				
¤ proportion of patients with stage III or IV at entry; or symptomatic patients at entry.				

Footnotes

3 UDCA effects on mortality and pre-defined covariates

Covaiates	Coefficient	95% CI	P-value
Risk of bias (low vs. high)	0.07	-0.56 to 0.71	0.82
UDCA dose	-0.14	-0.42 to 0.14	0.34
Trial duration	0.01	0.01 to 0.02	0.003
Severity of PBC*	-2.66	-5.11 to -0.20	0.03
*PBC: primary biliary cirrhosis			

Footnotes

4 UDCA effects on mortality or liver transplantation and pre-defined covariates

Covariates	Coefficient	95% CI	P-value
Risk of bias (low vs. high)	0.37	-0.35 to 1.09	0.32
UDCA dose	-0.10	-0.29 to 0.09	0.28
Trial duration	0.01	-0.02 to 0.03	0.08
Severity of PBC	-1.04	-3.19 to 1.11	0.34

Footnotes

5 Bayesian mete-regression: trial duration

Node	Median	95% CrI
Delt*	0.71	0.39 to 1.29
Gamma¤	0.03	0.01 to 0.05
* Delt representing UDCA effect (OR) adjusted for baseline risk and trial duration	¤ Gamma representing coefficient of trial duration.	

Footnotes

6 Bayesian meta-regression: disease severity

Node	Median	95% CrI
		0.43 to

Delt*	0.80	0.43 to 1.46
Gamma¤	-0.67	-4.26 to 2.75
* Delt representing UDCA effect (OR) adjusted for baseline risk and disease severity	¤ Gamma representing coefficient of disease severity	

Footnotes

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Data and analyses

1 UDCA versus placebo or no intervention

Outcome or Subgroup	Studies	Participants	Statistical Method	Effect Estimate
1.1 Mortality	14	1391	Odds Ratio (M-H, Fixed, 95% CI)	0.97 [0.62, 1.51]
1.2 Mortality or liver transplantation	15	1419	Odds Ratio (M-H, Fixed, 95% CI)	0.90 [0.65, 1.26]
1.3 Liver transplantation	14	1391	Odds Ratio (M-H, Fixed, 95% CI)	0.80 [0.50, 1.29]
1.4 Pruritus	5	438	Odds Ratio (M-H, Fixed, 95% CI)	0.93 [0.63, 1.39]
1.5 Pruitus score	3	271	Mean Difference (IV, Fixed, 95% CI)	-0.20 [-0.44, 0.05]
1.6 Fatigue	3	373	Odds Ratio (M-H, Fixed, 95% CI)	0.76 [0.49, 1.17]
1.7 Jaundice	2	198	Odds Ratio (M-H, Fixed, 95%	0.32 [0.12, 0.87]

			CI)	
1.8 Portal pressure	1	30	Mean Difference (IV, Fixed, 95% CI)	0.80 [-2.18, 3.78]
1.9 Development of varices	3	318	Odds Ratio (M-H, Fixed, 95% CI)	0.54 [0.25, 1.19]
1.10 Bleeding varices	4	451	Odds Ratio (M-H, Fixed, 95% CI)	0.53 [0.20, 1.43]
1.11 Hepatic encephalopathy	2	302	Odds Ratio (M-H, Fixed, 95% CI)	0.38 [0.06, 2.60]
1.12 Ascites	4	500	Odds Ratio (M-H, Fixed, 95% CI)	0.41 [0.18, 0.92]
1.13 Variceal bleeding, ascites, and/or encephalopathy	1	56	Odds Ratio (M-H, Fixed, 95% CI)	1.30 [0.31, 5.47]
1.14 S-bilirubin (µmol/l) - about six months	6	674	Mean Difference (IV, Fixed, 95% CI)	-10.30 [-15.48, -5.13]
1.15 S-alkaline phosphatases (IU/l) - about six months	6	595	Mean Difference (IV, Random, 95% CI)	-359.08 [-525.05, -193.11]
1.16 S-gamma-glutamyl transpeptidase (IU/l) - about six months	4	395	Mean Difference (IV, Fixed, 95% CI)	-257.82 [-318.28, -197.36]
1.17 S-aspartate aminotransferase (IU/l) - about six months	5	575	Mean Difference (IV, Random, 95% CI)	-35.45 [-53.08, -17.81]
1.18 S-alanine aminotransferase (IU/l) - about six months	5	325	Mean Difference (IV, Random, 95% CI)	-47.66 [-76.90, -18.42]
1.19 S-albumin (g/l) - about six months	2	280	Mean Difference (IV, Fixed, 95% CI)	0.10 [-0.14, 0.33]
1.20 S-cholesterol (total) (mmol/l) - about six months	5	461	Mean Difference (IV, Fixed, 95% CI)	-0.54 [-0.85, -0.24]
1.21 Plasma immunoglobulin M (g/l) - about six months	4	446	Mean Difference (IV, Fixed, 95% CI)	-1.25 [-1.85, -0.64]
1.22 Prothrombin index	2	338	Mean Difference (IV, Fixed, 95% CI)	1.18 [-1.15, 3.50]
1.23 Liver biopsy findings - dichotomous variables	6		Odds Ratio (M-H, Fixed, 95% CI)	Subtotals only
1.23.1 Worsening of histological stage	5	351	Odds Ratio (M-H, Fixed, 95% CI)	0.69 [0.44, 1.08]
1.23.2 Worsening of fibrosis	1	139	Odds Ratio (M-H, Fixed, 95%	0.82 [0.41, 1.65]

			CI)	
1.23.3 Florid duct lesion	1	115	Odds Ratio (M-H, Fixed, 95% CI)	0.80 [0.32, 2.02]
1.24 Liver biopsy findings - continous variables	1		Mean Difference (IV, Fixed, 95% CI)	Subtotals only
1.24.1 Histological stage	1	84	Mean Difference (IV, Fixed, 95% CI)	-0.54 [-0.91, -0.17]
1.24.2 Portal inflammation	1	84	Mean Difference (IV, Fixed, 95% CI)	-0.57 [-0.95, -0.19]
1.24.3 Piecemeal necrosis	1	84	Mean Difference (IV, Fixed, 95% CI)	-0.56 [-0.98, -0.14]
1.24.4 Lobular necrosis	1	84	Mean Difference (IV, Fixed, 95% CI)	-0.30 [-0.66, 0.06]
1.24.5 Ductular proliferation	1	489	Mean Difference (IV, Fixed, 95% CI)	-0.23 [-0.46, -0.00]
1.24.6 Cholestasis	1	84	Mean Difference (IV, Fixed, 95% CI)	-0.08 [-0.28, 0.12]
1.25 Liver biopsy findings - continous variables	1		Mean Difference (IV, Fixed, 95% CI)	Subtotals only
1.25.1 Bile duct/portal tract	1	84	Mean Difference (IV, Fixed, 95% CI)	0.23 [0.10, 0.36]

2 Adverse events - UDCA versus placebo or no intervention

Outcome or Subgroup	Studies	Participants	Statistical Method	Effect Estimate
2.1 Serious adverse events	10	990	Odds Ratio (M-H, Fixed, 95% CI)	3.07 [0.12, 77.41]
2.2 Non-serious adverse events	11	1149	Odds Ratio (M-H, Fixed, 95% CI)	1.74 [1.10, 2.75]

3 Influence of missing data - UDCA versus placebo or no intervention

Outcome or Subgroup	Studies	Participants	Statistical Method	Effect Estimate
3.1 Mortality - completed patient's course plus case scenarios	14		Odds Ratio (M-H, Random, 95% CI)	Subtotals only
3.1.1 Completed patient's course analysis	14	1247	Odds Ratio (M-H, Random, 95% CI)	0.93 [0.58, 1.48]
3.1.2 Assuming bad outcome	14	1391	Odds Ratio (M-H, Random, 95% CI)	0.77 [0.53, 1.11]
3.1.3 Assuming good outcome	14	1391	Odds Ratio (M-H, Random,	0.98 [0.62, 1.56]

			95% CI)	
3.1.4 Extreme case scenario favouring UDCA	14	1391	Odds Ratio (M-H, Random, 95% CI)	0.27 [0.15, 0.49]
3.1.5 Extreme case scenario favouring control	14	1391	Odds Ratio (M-H, Random, 95% CI)	2.38 [1.52, 3.71]
3.2 Mortality or liver transplantation - completed patient's course plus case scenarios	15		Odds Ratio (M-H, Random, 95% CI)	Subtotals only
3.2.1 Completed patient's course analysis	15	1275	Odds Ratio (M-H, Random, 95% CI)	0.88 [0.61, 1.27]
3.2.2 Assuming bad outcome	15	1419	Odds Ratio (M-H, Random, 95% CI)	0.83 [0.58, 1.19]
3.2.3 Assuming good outcome	15	1419	Odds Ratio (M-H, Random, 95% CI)	0.94 [0.67, 1.32]
3.2.4 Extreme case scenario favouring UDCA	15	1419	Odds Ratio (M-H, Random, 95% CI)	0.40 [0.24, 0.65]
3.2.5 Extreme case scenario favouring control	15	1419	Odds Ratio (M-H, Random, 95% CI)	1.81 [1.25, 2.63]
3.3 Mortality - uncertain interval	14	28	Odds ratio (IV, Fixed, 95% CI)	1.03 [0.80, 1.33]
3.4 Mortality or liver transplantation - uncertain interval	15	30	Odds ratio (IV, Fixed, 95% CI)	0.89 [0.64, 1.25]

4 UDCA-UDCA versus placebo/no intervention-UDCA

Outcome or Subgroup	Studies	Participants	Statistical Method	Effect Estimate
4.1 Mortality	14	1391	Odds Ratio (M-H, Fixed, 95% CI)	0.97 [0.68, 1.37]
4.2 Mortality or liver transplation	15	1419	Odds Ratio (M-H, Fixed, 95% CI)	0.81 [0.62, 1.05]
4.3 Liver transplantation	14	1391	Odds Ratio (M-H, Fixed, 95% CI)	0.70 [0.50, 0.98]

Figures

Sources of support

Internal sources

- The Copenhagen Trial Unit, Centre for Clinical Intervention Research, H:S Rigshospitalet, Denmark
- Copenhagen Hospital Corporation, Denmark

External sources

- S.C. Van Foundation, Denmark

Feedback

1 Ursodyeoxycholic acid for primary biliary cirrhosi

Summary

It would be helpful if the Comment had a sentence on what the substantive change is between the original article and the update so its significance, or lack thereof, is apparent.Thank you for your consideration.

I certify that I have no affiliations with or involvement in any organisation or entity with a direct financial interest in the subject matter of my criticisms.

Karyn Driessen, CA, USA
11.06.2003

Reply

Thank you very much for showing your interest in our review and for your comment.

The changes that occurred in our review between the version published in Issue I, 2003 (and previous issues) and in Issue II, 2003 were of no material importance to the data or conclusions of the review. The only encompassed minor stylistic changes as well as addition of an extra reference in the Background section.

Our original text in the Background was:

"Primary biliary cirrhosis (PBC) is a rather rare, chronic liver disease of unknown etiology. It was first comprehensively described by Ahrens and co-workers in 1950 (Ahrens 1950)."

This was changed into:

"Primary biliary cirrhosis (PBC) is a rather rare, chronic liver disease of unknown etiology. It was first comprehensively described around 1950 (MacMahon 1949; Ahrens 1950)."

Therefore, the review was not marked as 'Updated', we only changed the date of last amendment.

Your comment has made me realise the importance of keeping track of all changes, no matter how small. We shall remember that when we update our review in late 2003.

Christian Gluud
The Copenhagen Trial Unit
H:S Rigshospitalet

I certify that I have no affiliations with or involvement in any organisation or entity with a direct financial interest in the subject matter of my criticisms.

11.06.2003

Contributors

Christian Gluud, Erik Christensen.

Appendices

Appendix 1B

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Ursodeoxycholic Acid for Patients With Primary Biliary Cirrhosis: An Updated Systematic Review and Meta-Analysis of Randomized Clinical Trials Using Bayesian Approach as Sensitivity Analyses

Yan Gong, M.D., M.I.H.,[1] Zhibi Huang, M.P.H.,[2] Erik Christensen, M.D., Dr. Med.Sci.,[3] and Christian Gluud, M.D., Dr. Med.Sci.[1]

[1]*The Cochrane Hepato-Biliary Group, Copenhagen Trial Unit, Center for Clinical Intervention Research, Department 7102, Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark;* [2]*Department of Epidemiology, Institute of Public Health, Copenhagen University, Copenhagen, Denmark; and* [3]*Clinic of Internal Medicine I, H:S Bispebjerg Hospital, Copenhagen University Hospital, Copenhagen, Denmark*

OBJECTIVES: Ursodeoxycholic acid (UDCA) is used for primary biliary cirrhosis (PBC), but the beneficial effects remain controversial.

METHODS: We performed an updated systematic review to evaluate the benefits and harms of UDCA in patients with PBC. We included randomized clinical trials evaluating UDCA *versus* placebo or no intervention in patients with PBC. The primary outcomes, mortality and mortality or liver transplantation, were reported as relative risk (RR) with 95% confidence interval (CI). Meta-regression was used to investigate the associations between UDCA effects and the trial's risk of bias, UDCA dose, duration, and PBC severity at trial entry. We used Bayesian meta-analytic approaches as sensitivity analyses.

RESULTS: Sixteen randomized clinical trials (1,447 patients) evaluating UDCA *versus* placebo or no intervention were identified. Over half of the trials had high risk of bias. Comparing with placebo or no intervention, UDCA did not significantly affect mortality (RR 0.97, 95% CI 0.67–1.42) and mortality or liver transplantation (RR 0.92, 95% CI 0.71–1.21). The findings were supported by the Bayesian meta-analyses. Meta-regression analyses suggested that UDCA effects seem to be associated with patient's disease severity and trial duration. UDCA did not improve pruritus, fatigue, autoimmune conditions, liver histology, or portal pressure. UDCA seemed to improve biochemical variables, such as serum bilirubin, and ascites and jaundice, but the findings were based on few trials with sparse data. The use of UDCA was significantly associated with adverse events, mainly weight gain.

CONCLUSIONS: This updated systematic review did not demonstrate any benefit of UDCA on mortality and mortality or liver transplantation in patients with PBC.

(Am J Gastroenterol 2007;102:1799–1807)

INTRODUCTION

Primary biliary cirrhosis (PBC) is an uncommon and slowly progressive autoimmune disease of the liver that primarily affects middle-aged women. It was first comprehensively described around 1950 (1, 2). Over the last 30 yr, substantial increases in the prevalence of PBC have been observed (3). PBC is now a frequent cause of liver morbidity, and the patients are significant users of health resources, including liver transplantation (4). Fatigue and pruritus are the most common presenting symptoms (5). The diagnosis of PBC is currently based on the following triad: the presence of detectable antimitochondrial antibodies in serum, elevation of liver enzymes (most commonly alkaline phosphatases) for more than 6 months, and characteristic liver histological changes in the absence of extrahepatic biliary obstruction (6).

Bile duct destruction leads to the retention of hydrophobic bile acids within the liver cell. This likely contributes to the gradual deterioration in liver function observed in patients with PBC. Ursodeoxycholic acid (UDCA), the epimer of chenodeoxycholic acid, increases the rate of transport of intracellular bile acids across the liver cell and into the canaliculus in patients with PBC (7). UDCA is the only drug approved for PBC by the Food and Drug Administration. Doses of 13–15 mg/kg/day cause significant improvements in liver biochemistry and immunoglobulin levels and reduce titers

of antimitochondrial antibodies (8, 9). However, the effect of UDCA on mortality and histological progression remains controversial (10, 11). Since 2001, several randomized clinical trials have been published with the results of longer-term follow-up on patients' survival (12–14). We, therefore, re-evaluated the effects of UDCA in patients with PBC by updating our systematic review on the topic (11).

METHODS

We conducted the meta-analysis following our protocol (15) and the recommendations from the Cochrane Collaboration (16). We included and reviewed all randomized clinical trials assessing the effects of UDCA *versus* placebo or no intervention in patients with PBC, irrespective of blinding, language, year of publication, and publication status (15).

We searched for randomized trials in The Cochrane Hepato-Biliary Group Controlled Trials Register (17), The Cochrane Central Register of Controlled Trials in The Cochrane Library, MEDLINE, EMBASE, Science Citation Index-Expanded, The Chinese Biomedical CD Database, LILACS, and references of identified studies. The last search was performed in January 2007.

The primary outcome measures were mortality and mortality or liver transplantation. Secondary outcome measures were liver transplantation, pruritus, fatigue, clinical symptoms, liver biochemistry, liver biopsy, quality of life, adverse events (excluding mortality and liver transplantation), and cost-effectiveness.

In accordance with empirical evidence (18–20), we assessed the methodological quality of the trials. Trials with low risk of bias were the ones meeting the adequacy of three components: generation of the allocation sequence, allocation concealment, and blinding (18–20). Trials with high risk of bias were ones having one or more of these components regarded as inadequate or unclear.

We performed meta-analyses with Review Manager 4.2 (http://www.cochrane.dk). We analyzed data by random-effects (21) and fixed-effect (22) models. We presented binary outcome measures as relative risk (RR) with 95% confidence interval (CI) and continuous outcome measures as weighted mean difference (WMD) with 95% CI. Heterogeneity was explored by χ^2 test with significance set at $P < 0.10$. The degree of heterogeneity was measured by I^2 (23) and between-trial variance was estimated by the method of moments (21). The larger the I^2 and the moment-based between-trial variance, the larger degree of heterogeneity is present. We performed a meta-regression analysis with STATA (Intercooled STATA 8.0, Stata Corp., College Station, TX), which examined the effect size of UDCA in relation to the risk of bias, UDCA dosage, trial duration (treatment and follow-up), and severity of PBC at entry. We explored publication bias and other bias according to Begg's and Egger's methods (24, 25) with STATA.

We conducted the following sensitivity analyses to investigate the robustness of our main analyses on primary outcomes: (a) The influence of missing data: the missing data could be due to patient dropouts or lost to follow-up. We used an uncertainty method to allow for missing data (26). (b) Bayesian meta-analytic approach with WinBUGS (version 1.4.1, Medical Research Council, Biostatistics Unit, Cambridge, UK), in which Markov chain Monte Carlo with Gibbs sampling was applied. This approach is able to account for uncertainty of all relevant sources of variability in the random-effects model. The analog of a classical estimate is the marginal posterior median and the analog of a classical confidence interval is the credibility interval (CrI) (27). We used odds ratio (OR) as the summary statistic. For the ease of comparison, we reported the Bayesian results together with results from the classical meta-analysis presented as OR. (c) Bayesian meta-regression to estimate the UDCA effects adjusted for underlying risk. The underlying risk is a convenient and clinically relevant trial-level measure, which can be interpreted as a summary of a number of unmeasured patient characteristics (28). We also use this approach to investigate the relationship between one specific covariate (*e.g.*, UDCA dosage, trial duration, or disease severity of patients at entry) and the effects of UDCA adjusted for underlying risk.

RESULTS

We identified 863 references through electronic and hand searches. We excluded 762 duplicates or clearly irrelevant references and the remaining 101 references referred to 16 randomized clinical trials with 1,447 patients. Two of the 16 trials were published as abstracts only (29, 30), of which the De la Mora *et al.* trial (30) contained no extractable data with

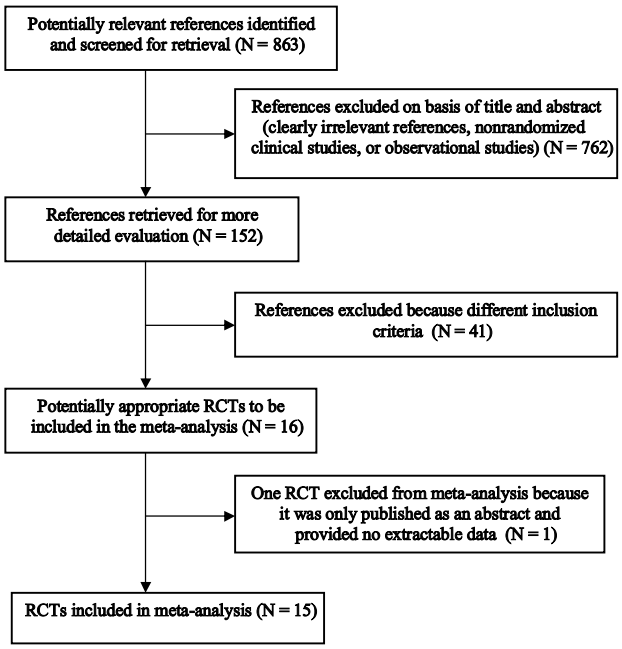

Figure 1. Flow diagram of trial selection.

Table 1. Characteristics of Included Trials of UDCA for Patients With PBC

Study ID	Risk of Bias	UDCA* Dose (mg/kg/day)†	Trial Duration‡ (months)	PBC§ Severity (%)¶	Notes
Athens 2002	High	13.5	92	64	14/43 control patients were crossed over to UDCA at their own request at a median of 3.5 yr (range 2–8 yr) after entry. The authors did both intention-to-treat analysis and treatment-as-received analysis.
Barcelona 2000	Low	15.5	64	27	None
Dallas 2004	High	11.5	24	67	Three patients randomized to receive placebo had high bile UDCA concentrations, suggesting UDCA intake. All patients were offered open-label UDCA following completion of the first 2 yr of the trial.
Frankfurt 1989	Low	10.0	9	15	None
Göteborg 1997	Low	7.7	24	34	At 24 months, 32 of 49 patients allocated to placebo and still remaining in the trial were switched to UDCA and 42 of 52 patients allocated to UDCA and still remaining in the trial continued with UDCA. Antihepatitis C virus tests not performed.
Helsinki 1995	Low	13.5	24	33	None
Manchester 1994	High	10.0	15	32	No exact data on number of patients randomized to each arm. No data given separately on mortality and liver transplantation.
Mayo-I 1994	Low	14.0	48	68	Patients originally receiving placebo switched to UDCA after 4 yr and followed for an additional 8 yr.
Milan 1993	High	8.7	12	50	Patients switched onto UDCA at the end of the trial.
Newark-II 1991	High	10.0	6	67	None
Newcastle 1994	Low	10.0	24	83	None
Taipei 1993	High	9.2	3	58	All patients switched to UDCA on completion of the 6 months crossover trial.
Tokyo 1990	High	9.2	6	38	None
Toronto 1994	High	14.0	24	53	Patients offered UDCA at the end of the trial.
Villejuif 1991	High	14.0	24	47	All patients treated for 2 yr with placebo were offered UDCA and further followed up for another 2 yr together with patients continuing on UDCA. One patient, included in the publications of the study up to 1993, was excluded from the 1994 publication due to a raised serum bilirubin at entry, violating the entry criteria.

*UDCA = ursodeoxycholic acid; †UDCA dose = average of the reported range; ‡Trial duration = includes treatment and follow-up; §PBC = primary biliary cirrhosis; ¶PBC severity = proportion of patients with stage III or IV at entry or with symptoms at entry.

28 patients (Fig. 1). Consequently, a summary of the 15 trials, *i.e.*, risk of bias, UDCA dose, trial duration, the percentage of patients with advanced PBC or presenting symptoms at entry, is given in Table 1. In the follow-up period, seven trials continued UDCA-treated patients on open-label UDCA (UDCA→UDCA) and offered open-label UDCA to all or some patients originally given placebo (placebo→UDCA) (8, 12–14, 31–33). Compared to the first version of this systematic review published in 2001 (11), the present review contains updated data on mortality and liver transplantation from three trials (12, 14, 34) and on adverse events from one trial (14) due to the new publications.

Mortality

Mortality data from 14 trials were combined. UDCA had no significant effects on mortality (RR 0.97, 95% CI 0.67–1.42, $I^2 = 0\%$, Fig. 2). In the UDCA group 45/699 (6.4%) patients died *versus* 46/692 (6.6%) patients in the control group. The moment-based estimate of between-trial variance is 0.042.

To take the missing data into account, we used the uncertainty method to estimate the UDCA effect on mortality (26). The result was consistent with the main finding above (RR 1.08, 95% CI 0.68–1.70). The Bayesian meta-analysis results (median OR 0.89, 95% CrI 0.50–1.49) also supported the main analysis presented as OR with 95% CI (OR 0.97, 95% CI 0.62–1.51). When adjusted for underlying risks the median OR was 0.82 and 95% CrI was 0.43–1.51 (Table 2).

In a meta-regression model we included risk of bias of the trials, UDCA dose, trial duration, and severity of PBC at entry as covariates and the effects of UDCA on mortality as a dependent variable. The model identified trial duration and severity of PBC as two covariates that might have associations with the effects of UDCA (Table 3). The moment-based estimate of between-trial variance changed from 0.042 to 0. Bayesian meta-regression was also used for sensitivity analysis to estimate the influence of the trial duration and disease severity on UDCA effect (Table 2).

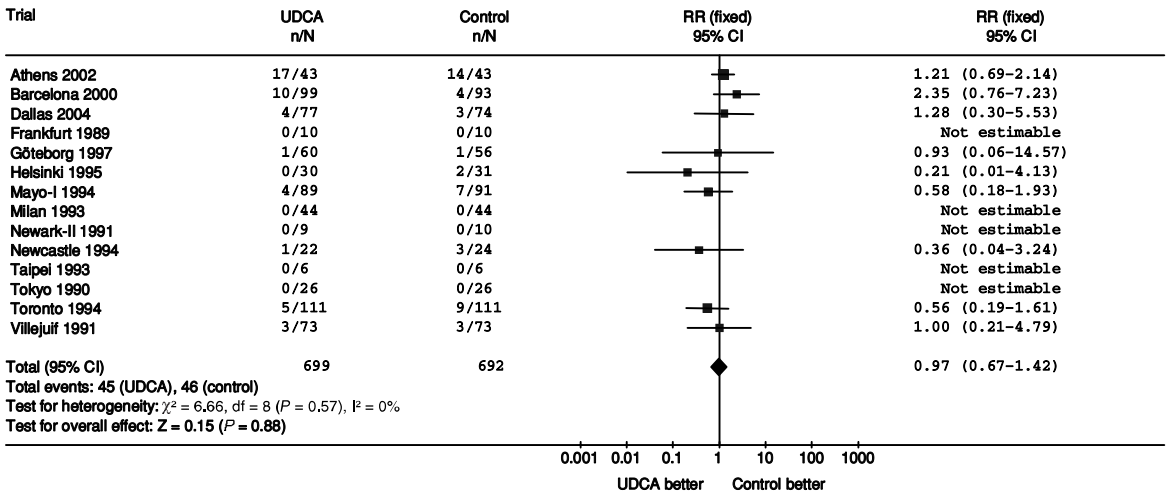

Figure 2. Forest plot of effect of UDCA on mortality. Abbreviations: CI = confidence interval; n = number of patients with outcome; N = number of participants at risk; df = degrees of freedom; I^2 = the percentage of total variation across studies that is due to heterogeneity rather than chance.The result and its 95% CI are represented by a diamond, with the relative risk (95% CI) and its statistical significance given alongside. Squares or diamonds to the left of the solid vertical line indicate benefit with UDCA, but this is conventionally significant ($P < 0{\cdot}05$) only if the horizontal line or diamond does not overlap the solid vertical line.

Analysis of data from the extended follow-up during UDCA→UDCA *versus* placebo→UDCA into the analyses demonstrated a RR of 0.97 with 95% CI 0.73–1.30. It compared 76 deaths in 699 patients (10.9%) originally randomized to UDCA with 78 deaths in 692 patients (11.3%) originally randomized to placebo or no intervention.

Mortality or Liver Transplantation

Combining the results of 15 trials demonstrated no significant effects on mortality or liver transplantation; neither UDCA nor placebo was favored (RR 0.92, 95% CI 0.71–1.21, Fig. 3). In the UDCA group 83/713 (11.6%) patients died or were transplanted *versus* 89/706 (12.6%) patients in the control group.

Taking missing data into consideration, UDCA effect on the composite outcome was estimated as RR 1.05 with 95% CI 0.75–1.48. The Bayesian analysis (median OR 0.84, 95% CrI 0.53–1.30) supported the main analysis presented as OR with 95% CI (OR 0.90, 95% CI 0.65–1.26). When adjusted for baseline risk, the median OR is 0.77 with 95% CrI 0.43–1.37.

In the classical meta-regression model and Bayesian meta-regression, no covariate seems to be significantly associated with the effect of UDCA on this outcome (data not shown).

Including data from the extended follow-up for UDCA→UDCA *versus* placebo/no intervention→UDCA demonstrated a RR of 0.86 with 95% CI 0.71–1.03. It compared 146 deaths or liver transplantations in 713 patients (20.5%) originally randomized to UDCA with 169 deaths or liver transplantations in 706 patients (23.9%) originally randomized to placebo or no intervention.

Liver Transplantation

Combining the results of the 14 trials demonstrated no significant effects on liver transplantation favoring UDCA (RR 0.82, 95% CI 0.53–1.26). In the UDCA group 34/699 (5.0%) patients had liver transplantation *versus* 41/692 (5.9%) patients in the control group.

Table 2. Bayesian Estimate of UDCA Effect on Mortality Presented as Posterior Median OR When Including One of Three Trial-Level Covariates, in Comparison to No Covariate, and the Influence of Covariates Presented as Posterior Median Coefficient, Both Applied to Mortality Data from 14 Trials on UDCA *versus* Placebo or No Intervention in Patients with PBC

	Posterior Median OR (95% Credibility Interval)	Posterior Median Coefficient (95% Credibility Interval)
No covariate	0.89 (0.50–1.49)	Not applicable
Underlying risk	0.82 (0.43–1.51)	0.10 (−0.62–0.65)
Trial duration (yr)	0.71 (0.39–1.29)	0.03 (0.01–0.05)
*PBC severity (%)	0.80 (0.43–1.46)	−0.67 (−4.26–2.75)

*PBC = primary biliary cirrhosis.

Table 3. Meta-Regression Analysis: UDCA Effects on Mortality for Predefined Trial-Level Covariates, *i.e.*, Risk of Bias, UDCA Dose, Trial Duration, and PBC Severity at Entry

	Coefficient	95% Confidence Interval	*P* Value
Risk of bias (low compared to high)	0.07	−0.56–0.71	0.82
UDCA* dose (mg/kg/day)	−0.14	−0.42–0.14	0.34
Trial duration (yr)	0.01	0.01–0.02	0.003
PBC† severity (%)	−2.66	−5.11 to −0.20	0.03

* UDCA = ursodeoxycholic acid; †PBC = primary biliary cirrhosis.

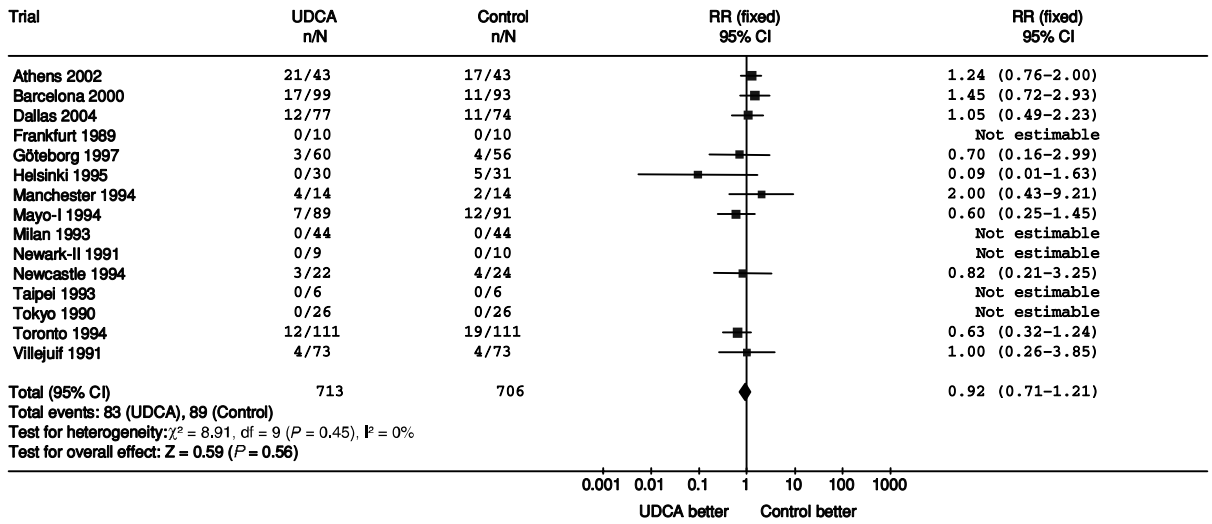

Figure 3. Forest plot of effect of UDCA on mortality or liver transplantation. Abbreviations: CI = confidence interval; n = number of patients with outcome; N = number of participants at risk; df = degrees of freedom; I^2 = the percentage of total variation across studies that is due to heterogeneity rather than chance. The result and its 95% CI are represented by a diamond, with the relative risk (95% CI) and its statistical significance given alongside. Squares or diamonds to the left of the solid vertical line indicate benefit with UDCA, but this is conventionally significant ($P < 0{\cdot}05$) only if the horizontal line or diamond does not overlap the solid vertical line.

Pruritus, Fatigue, Jaundice, and Other Clinical Symptoms

UDCA did not significantly influence either the number of patients with pruritus (RR 0.97, 95% CI 0.78–1.19, 5 trials) or the pruritus score (WMD −0.20, 95% CI −0.44 to 0.05, 3 trials). Fatigue was not significantly improved by UDCA (RR 0.90, 95% CI 0.76–1.06, 3 trials). Two trials reporting the number of patients with jaundice led to a significant effect favoring UDCA (RR 0.35, 95% CI 0.14–0.90) (33, 35). In most trials information on autoimmune conditions was sparse. However, the Mayo-I trial (36) evaluated the autoimmune conditions during the UDCA and placebo periods and did not find any significant effect of UDCA on associated sicca syndrome, Raynaud's phenomenon, arthritis, or Hashimoto's thyroiditis—neither on disappearance of conditions present at entry nor acquisition of new conditions.

Neither portal pressure (WMD 0.8 mmHg, 95% CI −2.2 to 3.8 mmHg, 1 trial), varices (RR 0.59, 95% CI 0.29–1.17, 3 trials), bleeding varices (RR 0.55, 95% CI 0.21–1.41, 4 trials), nor hepatic encephalopathy (RR 0.39, 95% CI 0.06–2.56, 2 trials) were significantly improved by UDCA. The number of patients developing ascites was significantly lower in the UDCA group compared with the control group (RR 0.42, 95% CI 0.19–0.93, 4 trials).

Liver Biochemistry

UDCA intervention led to some improvements on liver biochemistry (Table 4). Only one trial reported s-albumin concentrations (32) and one prothrombin index (33). The two variables were not significantly affected by UDCA.

Liver Histology

There were no significant effects of UDCA on histological stage (RR 0.78, 95% CI 0.57–1.06, random, 5 trials), fibrosis (RR 0.88, 95% CI 0.57–1.38, 1 trial), or florid duct lesions (RR 0.84, 95% CI 0.40–1.76, 1 trial). About half of the patients in the Barcelona trial observed statistically significant improvements in histological stage, portal inflammation, and piecemeal necroses in the UDCA group, but not regarding ductular proliferation or cholestasis. The placebo group had significantly fewer bile ducts per portal tract (9).

Quality of Life

None of the trials examined specific quality-of-life scales. Two trials evaluated symptoms using visual analog scales, (31, 37) and neither showed any significant difference between the UDCA and placebo group.

Adverse Events

Only Battezzati *et al.* reported one serious adverse event in the UDCA group, while the other trials only reported nonserious adverse events (32). UDCA led to a significantly higher incidence of adverse events (OR 1.74, 95% CI 1.10–2.75, 11 trials), mainly weight gain (38). Patients in the UDCA group gained an average of 3.6 kg ± 6.5%, which was significantly greater than the average of 0.6 kg ± 6.9% gained in the placebo group ($P = 0.04$) (38).

Publication Bias and Other Biases

Neither the Egger's nor the Begg's graphs and their tests on the mortality data provided evidence for asymmetry (Egger's test $P = 0.47$, Begg's test $P = 0.83$).

Table 4. Effects of UDCA on Liver Biochemistry

	WMD*	95% Confidence Interval	*P* Value	Number of Trials Analyzed
Bilirubin (μmoL/L)	−10	−16 to −5	<0.001	6
Alkaline phosphatases (IU/L)	−359	−525 to −193	<0.001	6
Gamma-glutamyl transpeptidase (IU/L)	−258	−318 to −197	<0.001	4
Aspartate aminotransferase (IU/L)	−36	−53 to −18	<0.001	5
Alanine aminotransferase (IU/L)	−48	−77 to −18	<0.001	5
Total cholesterol (mmoL/L)	−0.5	−0.8 to −0.2	<0.001	5
Plasma immunoglobulin M (g/L)	−1.3	−1.9 to −0.6	<0.001	4

*Weighted mean difference.

DISCUSSION

Our updated systematic review analyzed data from 15 randomized clinical trials assessing the effects of UDCA against placebo or no intervention for patients with PBC. With the inclusion of updated data from 2001 to January 2007, the present systematic review did not demonstrate any benefit of UDCA on mortality and mortality or liver transplantation. Thus, it supports and extends the main findings of the Goulis *et al.* meta-analysis (10) and our previous Cochrane systematic review (11). Moreover, the potential effects of UDCA on mortality seem to be associated with trial duration and disease severity: the longer the trial, the less effects of UDCA (if any); the more severe the patients are affected, the more effects of UDCA (if any). These findings are in direct contrast to the common claim that UDCA ought to be started early in less diseased patients in order to show its "full effect" (5, 39). There have been no new data on liver biochemistry and clinical symptoms since 2001, and we confirm a reduction in liver biochemistry, jaundice, and ascites following UDCA intervention. However, these results are based on few trials with sparse data. Trial selection bias and outcome reporting bias should, therefore, be considered. UDCA is generally well tolerated in patients with PBC.

There were no statistical signs of publication bias or other bias. This review analyzed 15 trials involving 1,447 patients. This is a low number of patients (40). The median length of trial duration was 2 yr. This is not sufficiently long considering that the estimated median survival of a patient with PBC is 10–15 yr (41). It is, therefore, difficult to detect a significant difference on mortality based on the trials, most of which have low statistical power. Furthermore, nine of the 15 trials had high risk of bias in terms of methodological quality. In general, trials with high risk of bias overestimate intervention effects (18–20). If the same overestimation is valid for the included trials, the prospects for UDCA in PBC may look even worse.

This systematic review did not demonstrate any benefit favoring UDCA on our predefined primary outcomes: mortality and mortality or liver transplantation. This observation is in contrast to some previous attempts to aggregate data from studies assessing UDCA interventions for PBC (42–44). However, Simko *et al.* (42) included nonrandomized studies in their meta-analysis. Such studies are more liable to bias. Poupon *et al.* included only three and five out of the 15 randomized clinical trials in their meta-analyses, respectively (43, 44). Such meta-analyses run the risk of trial selection bias—"cherry picking" (45).

Our main findings using a classical meta-analytic approach are consistent with the results using Bayesian approaches. In our review, the 95% Bayesian CrIs for both mortality and mortality or liver transplantation cover 1.0, indicating absence of significant intervention effect. Therefore, it strengthens the robustness of our main findings.

A common criticism about meta-analyses is that they combine information from trials with very different patient characteristics and designs, regarded as sources of heterogeneity. Therefore, it is justified to estimate the "true" UDCA effect after adjusting for important trial-level covariates. One important trial-level covariate is "underlying risk," *i.e.*, the average risk of an event (*e.g.*, mortality) for a patient at randomization. The "true" UDCA effect on mortality after adjusting the different underlying risks, by using a Bayesian approach, is estimated as median OR 0.82 with 95% CrI 0.43–1.51, and the "true" UDCA effect on mortality or liver transplantation is estimated as median OR 0.77 with 95% CrI 0.43–1.37. These results, taking underlying risk into consideration, support our unadjusted main meta-analyses.

We also considered other important and predefined trial-level covariates, including trial risk of bias, UDCA dose, trial duration, and severity of PBC. The classical meta-regression model showed that UDCA effect on mortality may be associated with trial duration and patients' disease severity at entry: the longer the trial, the less effects of UDCA (if any); the more severe PBC, the more effects of UDCA (if any). The moment-based estimate of between-trial variance is zero when the covariates are included, a change from 0.042 when no covariates are included. So the heterogeneity across the included trials seems largely explained by these two characteristics. The relationship between UDCA effect and trial duration is also supported by Bayesian meta-regression, which included "trial duration" as a covariate.

The previous Lancet meta-analysis (10) and our Cochrane systematic review (11) were mainly criticized for including many trials of only 2-yr duration and with very heterogeneous lengths of follow-up (5, 46). Given the updated evidence from

randomized clinical trials and analyses on longer follow-up data, our present review does not seem to support long-term UDCA intervention, which was suggested in observational studies (47, 48). Furthermore, estimation of UDCA's effect on mortality by Bayesian meta-analyses, adjusting for different length of trial duration and the above-mentioned underlying risk (OR 0.71, 95% CrI 0.39–1.29), has been consistent with the estimation from unadjusted pooled results (OR 0.89, 95% CrI 0.50–1.49). The adjusted result did not suggest any benefit of UDCA on mortality, even assuming that the trials have the same duration and underlying risk.

The relationship between UDCA effect and patients' severity of PBC was indicated in the classical meta-regression, meaning that UDCA's effect on mortality (if any) is more likely to be observed in patients with more severe PBC. This indication is supported by an analysis combining the raw data of three large clinical trials, in which a survival benefit of UDCA was observed in patients with moderate-to-severe disease, but not in those with mild disease (43). However, this relationship was not supported by our Bayesian meta-regression, which included "severity" as a covariate. Therefore, whether the UDCA intervention effect (if any) is related to the severity of PBC or not should be further investigated. Despite the uncertainty, the UDCA effect adjusting for the PBC severity and the above-mentioned underlying risk (OR 0.80, 95% CrI 0.43–1.46) has been consistent with the unadjusted pooled results (OR 0.89, 95% CrI 0.50–1.49). The adjusted result did not suggest any benefit of UDCA on mortality, even assuming that the trials have the same percentage of advanced patients and same level of underlying risk.

We noticed that the number of patients with ascites was significantly less in the UDCA group than in the placebo group. This observation originates from only four trials, and one may fear risk of publication bias and other bias. This observation could also be due to a play of chance, considering that many comparisons have been made without correction of the significance level. Furthermore, the diagnosis of ascites was clinically based; hence more susceptible to bias. Moreover, in our review, UDCA has not been found to decrease portal pressure and s-albumin, which are important in the pathogenesis of ascites. Accordingly, our observation needs confirmation.

It is interesting to know if UDCA could slow the histological progression. We were not able to identify any convincible benefits of UDCA on histology. The possibility that UDCA may still delay progression from early stage disease to late stage disease and then ultimately prolong survival cannot be proven or disproved with the trials completed. Only one trial found significant effects on liver histology (9). It observed positive effects on a number of histological variables, *e.g.*, the histological stage. This finding may also be a spurious one. Only about half of the randomized patients had a follow-up liver biopsy. Furthermore, as the trial showed a trend towards a higher mortality and liver transplantation rate in the UDCA group, this could have led to removal of some of the more seriously affected livers from the UDCA group; probably making those having a biopsy look relatively less affected. Such subgroup results should be interpreted cautiously (49–51). On the other hand, the finding of the Barcelona trial is interesting and should stimulate more clinical research into the effect of UDCA on progression of fibrosis in PBC and eventually cirrhosis development (9).

UDCA was generally well tolerated. We observed that UDCA was associated with nonserious adverse events, mostly weight gain. This finding ensued from new data from the Mayo-I trial (38). However, it is at present unclear if this weight gain should be considered a beneficial or a harmful effect and it needs further study. The effect ought to be mentioned to the patient before considering starting UDCA. Other nonserious adverse events included mild gastrointestinal disorders like diarrhea, nausea, vomiting, etc.

It has been claimed that UDCA is a cost-effective therapy for PBC (52). However, this claim rests on extrapolation from the results of two selected randomized clinical trials (8, 14). It is evident that cost-effectiveness analyses ought to be performed on the basis of all available high-quality evidence and not just on the selected. Considering the annual cost of UDCA of about $2,500 (52) and the findings of the present review, we challenge the conclusion drawn by Pasha *et al.* that UDCA is cost-effective for PBC.

Consistent with previous meta-analyses and reviews (10, 11), this updated systematic review did not demonstrate any benefit of UDCA on mortality and mortality or liver transplantation in patients with PBC. On the other hand, UDCA improved biochemical outcomes. This seems to place clinicians and researchers in a dilemma: if therapeutic decisions are based on clinical outcomes (*e.g.*, mortality), there is insufficient evidence to support the use of UDCA in PBC, but if based on nonvalidated "surrogate" outcomes (*e.g.*, s-bilirubin level), there is evidence favoring the UDCA interventions for the disease (53). This dilemma was reflected in a survey regarding the use of UDCA for PBC among Danish doctors (54), who had very different answers to the question of why they prescribed UDCA for PBC patients. Sixteen percent of the doctors thought UDCA reduced mortality, 27% thought UDCA reduced morbidity, and 23% thought it benefited "surrogate" outcomes (54, 55).

The Mayo Risk Score Model has identified several prognostic biomarkers for PBC, *e.g.*, serum bilirubin. These biomarkers may respond to intervention and are predictive of survival. But they do not necessarily predict clinical benefit of the intervention in question because "a perfect correlation does not a surrogate make" (56). In the absence of validated surrogate outcomes in UDCA for PBC, confirmatory trials assessing the UDCA effect should only be based on clinical outcomes, *e.g.*, survival. We believe that such clinical outcomes-based evaluation will benefit patients in the long run (53).

We also realize that the challenge of performing a new trial on intervention for PBC is high. The estimated median survival of PBC is 10–15 yr. To spend 15 yr planning and

carrying out a trial for each new potential treatment for PBC would consume many patients' lifetimes, not to mention the expense and difficulty of retaining patients in such a long study (57). Nevertheless, there are at least an estimated one million patients with PBC worldwide. Therefore, it is possible to conduct large trials with appropriate statistical power, if international groups of PBC investigators collaborate. Such large trials do not need to be conducted for more than 2–4 yr.

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Reprint requests and correspondence: Yan Gong, M.D., M.I.H., The Cochrane Hepato-Biliary Group, Copenhagen Trial Unit, Center for Clinical Intervention Research, Department 33.44, Rigshospitalet, Copenhagen University Hospital, Blegdamsvej 9, 2100 Copenhagen, Denmark.

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CONFLICT OF INTEREST

Guarantor of the article: Yan Gong, M.D., M.I.H.
Specific author contributions: Zhibi Huang performed part of the statistical analyses. Erik Christensen and Christian Gluud validated selection of trials and contributed to the drafting and editing of the review.
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Potential competing interests: None.

Appendix 2A

D-penicillamine for primary biliary cirrhosis (Review)

Gong Y, Frederiksen SL, Gluud C

This is a reprint of a Cochrane review, prepared and maintained by The Cochrane Collaboration and published in *The Cochrane Library* 2007, Issue 1

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ABSTRACT

Background
D-penicillamine is used for patients with primary biliary cirrhosis due to its hepatic copper decreasing and immunomodulatory potentials. The results from randomised clinical trials have been inconsistent.

Objectives
To systematically review the beneficial and harmful effects of D-penicillamine for patients with primary biliary cirrhosis.

Search strategy
We identified trials through electronic searches of *The Cochrane Hepato-Biliary Group Controlled Trials Register* (September 2003), *The Cochrane Central Register of Controlled Trials* on *The Cochrane Library* (Issue 3, 2003), *MEDLINE* (January 1966 to September 2003), *EMBASE* (January 1980 to September 2003), *The Chinese Biomedical CD Database* (January 1979 to August 2003), and *LILACS* (1982 to 2003); through manual searches of bibliographies; and by contacting authors of the trials and pharmaceutical companies.

Selection criteria
We included randomised clinical trials comparing D-penicillamine with placebo/no intervention or other control intervention irrespective of language, year of publication, and publication status.

Data collection and analysis
Two reviewers independently assessed the methodological quality of the trials and extracted data, validated by a third reviewer. The primary outcomes were 1) mortality and 2) a combination of those who died or underwent liver transplantation. We analysed dichotomous outcomes as relative risk (RR) with 95% confidence interval (CI) by a fixed effect model and a random effects model. We investigated sources of heterogeneity by subgroup analyses and tested the robustness of our findings by sensitivity analyses.

Main results
We included seven trials randomising 706 patients with primary biliary cirrhosis. D-penicillamine compared with placebo/no intervention tended to increase mortality (RR 1.34, 95% CI 1.09 to 1.64, fixed; RR 1.46, 95% CI 0.85 to 2.50, random). However, there was substantial heterogeneity. No significant differences were detected regarding the risks of mortality or liver transplantation, pruritus, liver complications, progression of liver histological stage, or the levels of liver biochemical variables (except alanine aminotransferase). D-penicillamine versus placebo/no intervention significantly increased the risk of adverse events (RR 3.11, 95% CI 2.33 to 4.16, fixed; RR 4.18, 95% CI 1.38 to 12.69, random).

Authors' conclusions
D-penicillamine did not appear to reduce the risk of mortality, but significantly increased the occurrences of adverse events in patients with primary biliary cirrhosis. We do not support the use of D-penicillamine for patients with primary biliary cirrhosis.

PLAIN LANGUAGE SUMMARY

D-penicillamine did not reduce the risk of mortality of patients with primary biliary cirrhosis but increased the occurrences of adverse events

Primary biliary cirrhosis is an uncommon, chronic liver disease of unknown etiology. D-penicillamine, a cupruretic drug, has been tested in randomised clinical trials and is used to treat patients with primary biliary cirrhosis. After combining results from seven trials, D-penicillamine did not appear to improve survival of patients. D-penicillamine was associated with a four-time increase of adverse events. There were no significant differences between D-penicillamine and placebo/no intervention with respect to clinical changes, liver histology, and liver biochemistry.

BACKGROUND

Primary biliary cirrhosis is an uncommon chronic progressive liver disease of unknown etiology. Ninety per cent of patients with primary biliary cirrhosis are females, and the majority are diagnosed after the age of 40 years (James 1981). Primary biliary cirrhosis is classically defined on the basis of the triad: antimitochondrial antibodies, found in over 95 per cent patients with primary biliary cirrhosis (Fregeau 1989; Lacerda 1995; Invernizzi 1997; Turchany 1997; Mattalia 1998); abnormal liver function tests that are typically cholestatic (raised activity of alkaline phosphatases are the most frequently seen abnormality); and characteristic liver histological changes (Scheuer 1967) without extrahepatic biliary obstruction (Kaplan 1996). Patients may either be diagnosed during a symptomatic phase (with common symptoms as pruritus, fatigue, jaundice, liver enlargement, signs of portal hypertension, sicca complex, and scleroderma-like lesions) when survival is decreased or during an asymptomatic phase when the prognosis is relatively favourable (Beswick 1985; Balasubramaniam 1990). However, 40 to 100 per cent of the asymptomatic patients will subsequently develop symptoms of primary biliary cirrhosis (Nyberg 1989; Metcalf 1996; Prince 2000).

Although the etiology remains unknown, primary biliary cirrhosis is in many respects analogous to the graft-versus-host syndrome in which the immune system is sensitised to foreign proteins. Most primary biliary cirrhosis patients have increased expression of class II human leukocyte antigen (HLA) histocompatibility on bile duct cells (Ballardini 1984; Van den Oord 1986). The bile duct epithelium in these patients is infiltrated with cytotoxic T-cells (Yamada 1986). Lacrimal and pancreatic glands, for example, with a high concentration of HLA class II antigens on their epithelium, may be involved in the disease process (Epstein 1982).

Patients with primary biliary cirrhosis are administered many drugs. Ursodeoxycholic acid, a bile acid, is the most extensively used drug (Verma 1999). However, a meta-analysis and a systematic Cochrane review were unable to demonstrate any significant effect of ursodeoxycholic acid on mortality or liver transplantation (Goulis 1999; Gluud 2002). Over the years, a number of other drugs have been evaluated for primary biliary cirrhosis. Attempts to treat primary biliary cirrhosis using immune-modulating and other agents such as azathioprine (Heathcote 1976; Christensen 1985), prednisolone (Mitchison 1992), chlorambucil (Hoofnagle 1986), cyclosporine (Wiesner 1990), colchicine (Warnes 1987; Vuoristo 1995; Poupon 1996), or methotrexate (Kaplan 1991; Lindor 1995) have resulted in clinical effects that have not led to widespread acceptance of these drugs in patients with primary biliary cirrhosis (Kaplan 1994).

D-penicillamine is a cupruretic drug known for its efficacy in treating Wilson's disease (Sternlieb 1964; Deiss 1971). Primary biliary cirrhosis is also associated with increased hepatic levels of copper. Therefore, the major rationale for evaluating D-penicillamine in primary biliary cirrhosis was its ability to induce cupruresis. In addition, D-penicillamine has other pharmacologic actions of potential benefit, including antifibrogenic effect, ability to decrease circulating immune complexes, and inhibitory effect on lymphocyte function (Nimni 1972; Epstein 1979; Lipsky 1980). There are about 2,500,000 patients with primary biliary cirrhosis in the world (Kim 2000). At least 2.8 per cent of these patients are probably being treated with D-penicillamine according to UK experience (Verma 1999). This means that about 70,000 primary biliary cirrhosis patients around the world may receive D-penicillamine as treatment. This figure may even be larger as we think that physicians in UK are conservative - at least when compared to other European physicians regarding interventions for alcoholic liver disease (Gluud 1993).

Conflicting reports concerning the effects of D-penicillamine in the treatment of primary biliary cirrhosis have been published. Earlier reports showed that D-penicillamine was a promising drug, improving survival in patients with primary biliary cirrhosis and having relatively few side-effects (Triger 1980; Epstein 1981; Taal 1983). Several later studies showed that D-penicillamine did decrease hepatic levels of copper, but it did not have a beneficial effect on symptoms related to primary biliary cirrhosis, hepatic biochemistries, histologic progression, or survival. In addition, D-penicillamine was associated with up to a 46 per cent incidence of major toxicity, most commonly proteinuria, allergic drug reaction, and rarely bone marrow depression (Matloff 1982; Neuberger 1985; Dickson 1985; Bodenheimer 1985). We have been unable to identify meta-analyses or systematic reviews on the beneficial and harmful effects of D-penicillamine in patients with primary biliary cirrhosis.

OBJECTIVES

To systematically assess the beneficial and harmful effects of D-penicillamine in patients with primary biliary cirrhosis.

CRITERIA FOR CONSIDERING STUDIES FOR THIS REVIEW

Types of studies

We included randomised clinical trials irrespective of language, year of publication, and publication status. We excluded studies using quasi-randomisation (eg, allocation by date of birth).

Types of participants

Patients with primary biliary cirrhosis, ie, patients having at least two of the following: elevated serum activity of alkaline phosphatases (or other markers of intrahepatic cholestasis), and/or a positive result for serum mitochondrial antibody, and/or liver biopsy findings diagnostic for or compatible with primary biliary cirrhosis.

Types of intervention

D-penicillamine at any dose compared with placebo, no intervention, another active drug, or other dose of D-penicillamine. Co-interventions were allowed as long as both intervention arms of the randomised clinical trial received similar co-interventions.

Types of outcome measures

The primary outcome measures were:

- Mortality.
- A combination of mortality or liver transplantation.

The secondary outcome measures were:

- Liver transplantation.
- Pruritus: number of patients without improvement of pruritus and/or pruritus score.
- Fatigue: number of patients without improvement of fatigue and/or fatigue score.
- Liver complications: number of patients developing variceal bleeding, ascites, hepatic encephalopathy, jaundice, hepato-renal syndrome, or sicca complex.
- Liver biopsy findings: deterioration of liver histological stage or score.
- Liver biochemistry: serum (s)-bilirubin; s-alkaline phosphatases; s-gamma-glutamyltransferase; s-aspartate aminotransferase; s-alanine aminotransferase; s-albumin; s-cholesterol (total); plasma immunoglobulin M, etc.
- Adverse events. The adverse event is defined as any untoward medical occurrence in a patient in either of the two arms of the included randomised clinical trials, which did not necessarily have a causal relationship with the treatment, but did, however, result in a dose reduction, discontinuation of treatment, or registration of the advent as an adverse event/side effect. The adverse events are subdivided into non-serious adverse events as well as serious adverse events according to the ICH-GCP guidelines (ICH-GCP 1997). A serious adverse event is any event that leads to death, is life-threatening, requires inpatient hospitalisation or prolongation of existing hospitalisation, results in persistent or significant disability or congenital anomaly/birth defect, or any important medical event which may jeopardize the patient or requires intervention to prevent it.
- Quality of life: a broad concept that includes physical functioning (ability to carry out activities of daily living such as self-care and walking around), psychological functioning (emotional and mental well-being), social functioning (social relationships and participation in social activities), and perception of health, pain, and overall satisfaction with life.
- Cost-effectiveness: the estimated costs connected with the interventions were to be weighed against any possible health gains.

SEARCH METHODS FOR IDENTIFICATION OF STUDIES

See: Cochrane Hepato-Biliary Group methods used in reviews.

Relevant randomised clinical trials were identified by searching *The Cochrane Hepato-Biliary Group Controlled Trials Register* (September 2003), *The Cochrane Central Register of Controlled Trials* on *The Cochrane Library* (Issue 3, 2003), *MEDLINE* (January 1966 to September 2003), and *EMBASE* (January 1980 to September 2003), *The Chinese Biomedical CD Database* (January 1979 to August 2003), and *LILACS* (1982 to 2003). See 'Table 01' for the search strategies applied to the individual electronic databases.

Further trials were identified by reading the reference lists of the identified studies. We wrote to the principal authors of the identified randomised clinical trials and to researchers active in the field to inquire about additional randomised clinical trials they might know of. We also wrote to the pharmaceutical companies that sponsored D-penicillamine in the identified trials in order to obtain any unidentified or unpublished randomised clinical trial.

METHODS OF THE REVIEW

The meta-analyses were performed following the published protocol and the recommendations given by the Cochrane Reviewers' Handbook (Alderson 2003).

Trials selection

Identified trials were listed and two contributors (YG and SLF) independently evaluated whether the trials fulfilled the inclusion criteria. Excluded trials were listed in 'Characteristics of excluded studies' with the reasons for exclusion. Disagreements were resolved by discussion.

Data extraction

YG and SLF independently extracted data onto a standard paper form, and CG validated the data extraction. We wrote to the authors of the included trials and asked them to specify the data of interest if those data were not reported clearly in their reports.

Assessment of methodological quality of included trials

The methodological quality of the randomised clinical trials was assessed using four components (Schulz 1995; Moher 1998; Kjaergard 2001):

Generation of the allocation sequence

- Adequate, if the allocation sequence was generated by a computer or random number table. Drawing of lots, tossing of a coin, shuffling of cards, or throwing dice will be considered as adequate if a person who was not otherwise involved in the recruitment of participants performed the procedure;
- Unclear, if the trial was described as randomised, but the method used for the allocation sequence generation was not described;
- Inadequate, if a system involving dates, names, or admittance numbers were used for the allocation of patients. These studies are known as quasi-randomised and were excluded from the present review.

Allocation concealment

- Adequate, if the allocation of patients involved a central independent unit, on-site locked computer, identically appearing numbered drug bottles or containers prepared by an independent pharmacist or investigator, or sealed envelopes;
- Unclear, if the trial was described as randomised, but the method used to conceal the allocation was not described;
- Inadequate, if the allocation sequence was known to the investigators who assigned participants or if the study was quasi-randomised. Such studies were excluded from the present review.

Blinding (or masking)

- Adequate, if the trial was described as double blind and the method of blinding involved identical placebo or active drug;
- Unclear, if the trial was described as double blind, but the method of blinding was not described;
- Not performed, if the trial was not double blind.

Follow-up

- Adequate, if the numbers and reasons for dropouts and withdrawals in all intervention groups were described or if it was specified that there were no dropouts or withdrawals;
- Unclear, if the report gave the impression that there had been no dropouts or withdrawals, but this was not specifically stated;
- Inadequate, if the number or reasons for dropouts and withdrawals were not described.

Characteristics of patients

Number of patients randomised; patient inclusion and exclusion criteria; mean (or median) age; sex ratio; number of patients lost to follow-up; drop-outs; withdrawals.

Characteristics of interventions

Type, dose, and form of D-penicillamine intervention; type of intervention in the control group and collateral interventions; trial duration.

Characteristics of outcomes

All outcomes were extracted from each included trial.

We analysed mortality and/or liver transplantation at maximum follow-up. We analysed other outcomes, which were repeatedly observed on patients (like liver biochemistry, clinical symptoms, etc.) at maximum follow-up.

Statistical methods

We intended to include parallel group and cross-over trials. For cross-over trials, we only intended to include data from the first period. We used the statistical package (RevMan Analyses 1.0.2) provided by The Cochrane Collaboration. We presented dichotomous data as relative risk (RR) with 95% confidence interval (CI) and continuous outcome measures by weighted mean differences (WMD) with 95% CI. All analyses for primary outcomes were performed according to the intention-to-treat principle, which means that participants in the trials should have been analysed in the groups to which they were randomised, regardless of whether they received or adhered to the allocated intervention.

We examined intervention effects by using both a random effects model (DerSimonian 1986) and a fixed effect model (DeMets 1987) with the significant level set at P-value ≤ 0.05. If the results of the two analyses led to the same conclusion, we presented only the results of the fixed effect analysis. In case of discrepancies of the two models, we reported the results of both models. We explored the presence of statistical heterogeneity by chi-squared test with significance set at P-value ≤ 0.10 and measured the quantities of heterogeneity by I^2. However, due to possible few anticipated trials and the relative large number of outcomes going to be assessed, we interpreted significant results with caution.

Subgroup analysis and sensitivity analysis

We performed subgroup analyses, in which trials were grouped according to the methodological quality of the included trials, dosage of D-penicillamine, and duration of treatment and follow-up. The high methodological quality was confined to adequate generation of the allocation sequence, allocation concealment, blinding, and follow-up. The difference between the estimates of two subgroups was estimated according to Altman 2003.

Regarding the primary outcome measure, ie, mortality, we included patients with incomplete or missing data in the sensitivity analyses by imputing them (Hollis 1999):

- Available case analysis: data on only those whose results are known, using as denominator the total number of patients who completed the trial;
- Assuming poor outcome: dropouts from both the D-penicillamine and control groups had the primary outcomes;
- Assuming good outcome: none of the dropouts from the D-penicillamine and control groups had the primary outcomes;
- Extreme case favouring D-penicillamine: none of the dropouts from the D-penicillamine-group but the dropouts from the control group had the primary outcomes;
- Extreme case favouring control: all dropouts from the D-penicillamine-group but none from the control group had the primary outcomes.

For secondary outcomes, we adopted 'available case analysis'. Therefore, in the review, the number of patients in the denominator changed according to the secondary outcomes investigated.

Bias exploration

Funnel plot was used to provide a visual assessment of whether treatment estimates are associated with study size. The performance of the available methods of detecting publication bias and other biases (Begg 1994; Egger 1997; Macaskill 2001) vary with the magnitude of the treatment effect, the distribution of study size, and whether a one- or two-tailed test is used (Macaskill 2001). Therefore, we used the most appropriate method, which has a good trade-off in the sensitivity and specificity, based on characteristics of the trials included in this review.

DESCRIPTION OF STUDIES

We identified a total of 178 references through electronic searches of *The Cochrane Hepato-Biliary Group Controlled Trials Register* (n = 26), *The Cochrane Central Registerof Controlled Trials* on *The Cochrane Library* (n = 28), *MEDLINE* (n = 29), *EMBASE* (n = 51), *The Chinese Biomedical CD Database* (n = 43), and *LILACS* (n = 1). We excluded 143 duplicates and clearly irrelevant references by reading abstracts. Accordingly, 35 references were retrieved for further assessment. Of these, we excluded three because they were non-randomised clinical studies or observational studies. The remaining 32 references referred to seven randomised clinical trials involving 706 patients with primary biliary cirrhosis, which fulfilled our inclusion criteria ('Characteristics of included studies' table). The year of publication of these trials ranged from year 1981 to 1985. The Bassendine 1982 trial was published as an abstract only, while the other six trials were published as full papers.

The mean age of the patients was about 51 years. The majority of the patients were females (female/male: 495/53) in the trials reporting gender distribution. The Bassendine 1982 trial and the Taal 1983 trial did not report the baseline histological status of primary biliary cirrhosis. Data from the other five trials showed that more patients had stage III or IV than stage I or II (stage III or IV /stage I or II: 443/168).

Of the seven trials, six trials compared D-penicillamine with placebo/no intervention. One trial compared 750 mg/day D-penicillamine with 250 mg/day D-penicillamine (Bodenheimer 1985). The Bassendine 1982 trial had three groups of comparisons: D-penicillamine 1g/day, 250 mg/day, and no intervention group. We extracted data from the group of 1g/day versus no intervention, which was the most commonly used dosage. All of the remaining five trials employed placebo as control intervention. The D-penicillamine dosage of 1g/day was applied in four trials (Bassendine 1982; Matloff 1982; Taal 1983; Dickson 1985), 1.2 g/day in the Neuberger 1985 trial, and 0.6 g/day in the Epstein 1981 trial. The duration of treatment and follow-up varied from 1.5 to 9 years.

METHODOLOGICAL QUALITY

Generation of the allocation sequence was adequate in one trial (Dickson 1985) and unclear in the other six. Allocation concealment was adequate in two trials (Dickson 1985; Matloff 1982) and unclear in the other five. Blinding was adequate in five trials, was considered inadequate in the Neuberger 1985 trial, and was not performed in the Bassendine 1982 trial. It should be noted that the description of the control in the trials reporting double blinding was not sufficient since all the trials claiming to be double blind only stated the use of identical in appearance placebo tablets, but did not address smell and taste. Follow-up was adequate in five trials, but considered inadequate in two trials (Bodenheimer 1985; Epstein 1981). In total, 90 patients (17%) were lost to follow-up: 79 patients in D-penicillamine and 11 in control group. In the Neuberger 1985 trial, 35 (36%) patients in the D-penicillamine group and 7 (8%) in the control group were lost to follow-up. None of the trials reported a sample size estimate. No trials reported that they used intention-to-treat analyses. Overall, only the Dickson 1985 trial was viewed as a high methodological quality trial, ie, having adequate generation of allocation, allocation concealment, blinding, and follow-up.

RESULTS

D-penicillamine versus placebo/no intervention

Mortality

Six trials (628 patients) provided data to estimate the risk of mortality of D-penicillamine versus placebo/no intervention (Comparison 01-01; Comparison 01-02). The mortality risk was 1.46

(95% CI 0.85 to 2.50) by the random effects model and 1.34 (95% 1.09 to 1.64) by the fixed effect model. The trials had significant heterogeneity (I^2 = 77.5%).

We performed sensitivity analyses to assess the impact of missing data. The 'assuming poor outcome' showed a significant harmful effect of D-penicillamine on mortality. However, the 'assuming good outcome' analyses did not detect a significant difference of mortality between D-penicillamine and placebo/no intervention (RR 0.95, 95% CI 0.71 to 1.26). The 'extreme case favouring control' showed a significant harmful effect of D-penicillamine on mortality (RR 1.92, 95% CI 1.51 to 2.43, fixed, RR 2.13, 95%CI 1.16 to 3.90, random). The 'extreme case favouring D-penicillamine' showed a significant beneficial effect of D-penicillamine (RR 0.66, 95% CI 0.51 to 0.86). We also performed 'available case analysis', in which we did not find a significant difference between D-penicillamine and placebo (RR 1.08, 95% 0.82 to 1.43).

We performed subgroup analyses according to different methodological quality, dosages of D-penicillamine, duration of treatment and follow-up, and histological stages (Comparison 01-05 to 11). The estimate of intervention effect were significantly different in the subgroup analyses of generation of allocation sequence (P = 0.03), allocation concealment (P = 0.04), blinding (P = 0.007), and follow-up (P = 0.008). The subgroup analyses stratifying the trials into three dosages of D-penicillamine (1.2 g/day, 1 g/day, or 0.6 g/day) did not show a clear increasing trend towards harmful effects of D-penicillamine along with increased dosage (Comparison 01-09), although the lowest dose had the lowest harm profile. The trial using dosage of 0.6 g/day showed a significant difference from the trials with 1 g/day (P = 0.04) and with 1.2 g/day (P = 0.005), while the comparison between 1 g/day and 1.2 g/day did not achieve significance. The risks of mortality in the trials with short-term treatment and follow-up (shorter than three years) had a significant difference with the trials with long-term treatment and follow-up (longer than three years) (P = 0.003).

Mortality or liver transplantation

Only one trial (Neuberger 1985) reported the number of patients who underwent liver transplantation (RR 0.93, 95% CI 0.06 to 14.63). Accordingly, the relative risk of mortality or liver transplantation was 1.33 (95% CI 1.09 to 1.63) in the fixed effect model and 1.45 (95% CI 0.85 to 2.48) in the random effects model (Comparison 01-13,01-14).

Pruritus, fatigue, and liver complications

Neuberger 1985 observed a marginal beneficial effect of D-penicillamine on pruritus (RR 0.57, 95% CI 0.33 to 0.99) (Comparison 01-15). Evidence about fatigue was not located. For liver complication, no significant differences were found with respect to gastrointestinal bleeding and ascites (Comparison 01-16)

Liver histological and biochemical outcomes

Data from three trials with 149 patients estimated the effects of D-penicillamine on liver histology (Epstein 1981; Matloff 1982; Taal 1983). D-penicillamine did not retard the progression of liver histological stage (RR 0.96, 95% CI 0.58 to 1.58) but D-penicillamine had a significant beneficial effect on inflammatory activity in the Epstein 1981 trial (RR 0.50, 95% CI 0.26 to 0.94, one trial).

Matloff 1982 provided data on liver biochemical outcomes presented as mean changes from values for each patient before randomization and showed no significant differences between D-penicillamine and placebo except for alanine aminotransferase.

Adverse events

All the seven trials reported adverse events in both groups. In the D-penicillamine group, 139 (43%) patients had adverse events (types of adverse events in Table 02) versus 44 (15%) patients treated with placebo/no intervention (RR 4.18, 95% CI 1.38 to 12.69, random; RR 3.11, 95% CI 2.33 to 4.16, fixed, I^2 = 93.2%) (Comparison 01-23, 24). In the sensitivity analysis after excluding the Taal 1983 trial, which had the smallest sample size (24 patients)and the highest placebo response rate (85 per cent), the RR changed to 3.69 (95% CI 2.62 to 5.19) and I^2went down to 49.7%. We were unable to distinguish between serious and non-serious adverse events due to insufficient reporting.

Quality of life and cost-effectiveness

None of the trials examined specific quality-of-life scales or outcomes regarding cost-effectiveness.

High-dose D-penicillamine versus low-dose D-penicillamine

In the Bassendine 1982 trial, the risk of mortality tended to be lower with a high-dose than with a low-dose D-penicillamine, although this difference is not significant (RR 0.26, 95% CI 0.06 to 1.05). More patients in the high-dose group than in the low-dose group tended to develop adverse events (RR 1.99, 95% CI 0.81 to 4.89). The Bodenheimer 1985 trial only reported the total number of deaths in the two groups, and more patients in the high-dose group had improvement of histological progression than in the low-dose group.

Bias exploration

We did not perform funnel plot analysis and did not apply the three statistical methods to detect publication bias and other biases because the power of those would have been low and inconsistent because of the small number of included trials.

DISCUSSION

We found that D-penicillamine tended to have a detrimental effect on mortality of patients with primary biliary cirrhosis. The meta-analysis also showed that the use of D-penicillamine significantly increased the occurrences of adverse events.

Our systematic review on D-penicillamine versus placebo/no intervention analysed only six trials involving 628 patients. This is

a low number of patients (Ioannidis 2001). None of the trials reported a sample size estimate. The loss during follow-up was relatively high in the D-penicillamine group. The methodological trial quality was generally low, which makes it hard to interpret this sample of trials. Generally, low methodological quality trials overestimate significantly intervention effects (Schulz 1995; Moher 1998; Kjaergard 2001). If the same overestimation is valid for the present sample of trials, the prospects for D-penicillamine for primary biliary cirrhosis look even worse, ie, the harmful effects could be even larger. On the other hand, we cannot preclude that such low-doses D-penicillamine may have beneficial effects because only a few trials have been performed with low-doses. In addition, most of the trials have shorter follow-up than the estimated median survival of 10 to 15 years (Prince 2002). Therefore, it is difficult to detect a significant difference on mortality.

Heterogeneity is an important aspect of a meta-analysis. Heterogeneity can occur because of an artefact of the summary measures used and of trial design features such as duration of follow-up, reliability of outcome measures, or methodological quality of the trial. It may also be due to real variations in the treatment effect, such as the underlying risk of the patients in the different trials, intervention timing or intensity, co-intervention, or the outcome measurement and timing. Although the ideal way to study causes of true variations is within trials rather than between, in most situations we had to do with a trial level investigation in the present meta-analyses (Glasziou 2002).

Regarding mortality, we found 'severe' heterogeneity (Higgins 2002) across the trials and also discrepancy between the fixed effect analysis and the random effects analysis. In the fixed effect analysis, we detected a significant harmful effect of D-penicillamine, while in the random effects analysis, no significant difference was found. Due to the low number of trials, which did not allow us to perform a meaningful meta-regression, we performed subgroup analyses according to the methodological quality, dosage of D-penicillamine, and duration of treatment and follow-up. Bearing in mind the observational nature of the subgroup analyses, we found that only the unclear/inadequate follow-up tended to underestimate the beneficial effects of D-penicillamine. This finding is in contrast to previous studies (Schulz 1995; Moher 1998; Kjaergard 2001), probably because too low number of trials were included to perform any meaningful subgroup analyses.

We found that D-penicillamine had no significant effect on reducing the risk of mortality compared to placebo/no intervention. The pooled estimate from high quality trials also support this finding. The analyses of the four scenarios, which took the impact of missing data into consideration, showed that patients taking D-penicillamine were more likely to have higher risk of mortality compared to patients taking placebo or getting no intervention. The 'assuming poor outcome' showed the significantly harmful effect of D-penicillamine, while the 'assuming good outcome' did not catch any significant difference between D-penicillamine and placebo/no intervention.

The subgroup analysis showed that the risk of mortality seemed to increase by dosage. This observation, however, was not supported by the Bassendine 1982 trial, where the patients taking high dose of D-penicillamine had lower risk of mortality than patients on low dose. Since the ideal way to study causes of true variation is within trials rather than between, and the purpose and nature of this meta-analysis was not to study the dose-response, the relationship between the effect of D-penicillamine and dosage is not clear.

It is presumed that high-risk groups will have more to gain from an intervention and may therefore experience sufficient benefit to outweigh the harms. Whether the severity of primary biliary cirrhosis was related to the treatment effect of D-penicillamine is not confirmed in this review. There was lack of trials to be included and also the possible relationship was not indicated in many of the trials.

Only Neuberger et al reported the number of patients having clinical changes (Neuberger 1985), which revealed that there were no significant differences on the state of pruritus, gastrointestinal bleeding, or ascites between D-penicillamine and placebo. Although the remaining trials did not report the exact data, they all claimed that no consistent clinical improvement in either the D-penicillamine or placebo group had been found (Dickson 1985; Matloff 1982; Taal 1983).

Data from three trials enabled us to meta-analyse the effects of D-penicillamine on liver histology and we found that the rate of liver histological progression neither favoured D-penicillamine nor favoured placebo/no intervention (Epstein 1981; Matloff 1982; Taal 1983). There is a significant beneficial effect of D-penicillamine regarding histological inflammatory activity (Epstein 1981). However, the effect is only marginally significant and based on only one trial with a small sample size of patients.

The report by Matloff et al allowed us to extract data on liver biochemical variables, which resulted in no significant differences except for alanine aminotransferase (Matloff 1982). This finding was replicated in the Neuberger 1985 trial in which alanine aminotransferase was the only significant difference among the various liver biochemical variables. Epstein 1981and Bassendine 1982 found a beneficial effect of D-penicillamine in reducing the levels of aspartate aminotransferase and immunoglobulin. Dickson 1985 did not detect any significant effect, and Taal 1983 found that D-penicillamine significantly decreased immunoglobulin M and G levels. Thus, the inconsistent findings across the trials weakened the conclusion of beneficial effect of D-penicillamine on liver biochemical variables at large.

Six out of seven trials reported on adverse events and showed that the risk of adverse events in the D-penicillamine group was, on average, four times higher than the placebo/no intervention group both in random effects and fixed effect models. Most of the adverse

events were proteinuria, gastrointestinal upset, rash, cytopenia, etc.

In the meta-analyses of adverse events, we also found severe heterogeneity across the trials. Although the results from the fixed effect and random effects models indicated that the use of D-penicillamine highly increased the occurrences of adverse events, investigation for sources of heterogeneity was necessary. We found that I^2decreased to zero (no statistical heterogeneity) when changing the RR to the odds ratio (OR). However, the selection of a summary measure on the basis of minimising heterogeneity is a somewhat data derived approach since it generates spurious, over-optimistic findings. It is theoretically possible that important sources of heterogeneity could be missed if the strategy of using the summary with the smallest heterogeneity statistic is universally applied (Deeks 2001a; Deeks 2002). Considering that the selection of a summary measure being argued on the grounds of consistency of effect, ease of interpretation, and mathematical properties, we left RR as the summary measure in the analysis of adverse events.

For meta-analyses of RR, the proportional weights, given to trials estimating the same effect with the same sample size, increase with increasing event rates. The relationship becomes particularly strong when the event rates are above 50 per cent (Deeks 2001b). In this respect, we scrutinized the event rates in the included trials and we noticed that the Taal 1983 trial had the smallest sample size (24 patients), but surprisingly the highest placebo response rate, 85 per cent. It was offered the second most weight, 23 per cent in the analysis of RR, whereas the weight of 2 per cent was used in the analysis of OR. Hence, we performed a sensitivity analysis excluding the Taal 1983 trial, and it resulted in the RR of 3.69 (95% CI 2.62 to 5.19) with the acceptable moderate heterogeneity (I^2 = 49.7%). Therefore, our conclusion, that the use of D-penicillamine was accociated with the increase of adverse events, was consolidated.

AUTHORS' CONCLUSIONS

Implications for practice

D-penicillamine did not significantly reduce the risk of mortality of patients with primary biliary cirrhosis. Furthermore, we found a significant increase of adverse events when comparing patients taking D-penicillamine with those on placebo/no intervention. Hence, we are against using D-penicillamine for patients with primary biliary cirrhosis.

Implications for research

We do not recommend further randomised clinical trials aiming at establishing the value of D-penicillamine in the treatment of primary biliary cirrhosis, at least not with the dosages employed in previous trials. The possibility that low doses may offer beneficial effects cannot be excluded. Investigators ought to report their trials according to the CONSORT Statement (www.consort-statement.org).

POTENTIAL CONFLICT OF INTEREST

None known.

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* *Indicates the major publication for the study*

TABLES

Characteristics of included studies

Study	Bassendine 1982
Methods	Generation of allocation sequence: unclear. Allocation concealment: unclear. Blinding: not performed. Follow-up: adequate, four patients in the high-dose D-penicillamine, five patients in the low-dose D-penicillamine, and none in control were lost to follow-up.
Participants	Country: UK. Mean age: not reported. Female/Male: not reported. PBC stage status: not reported.
Interventions	D-penicillamine 1g/day (n = 19) D-penicillamine 250 mg/day (n = 22) No intervention (n = 19) Mean period of follow-up: 37 months.

Characteristics of included studies (*Continued*)

	Analysed duration of trial: 3 years.
Outcomes	1. Mortality. 2. Liver biochemical variables. 3. Adverse events.
Notes	1. Side effects required withdrawal of D-penicillamine in nine patients. 2. It was only published as an abstract. 3. Correspondence sent to the author on 2 December 2003. No reply was received by 20 June 2004.
Allocation concealment	B – Unclear

Study	**Bodenheimer 1985**
Methods	Generation of allocation sequence: unclear. Allocation concealment: unclear. Blinding: adequate, identical placebo. Follow-up: inadequate, 26 patients in both groups were lost to follow-up.
Participants	Place: USA. Mean age: 52 in high-dose group and the same in low-dose group. Female/Male:51/5. PBC stage status: 10 with stage I or II in high-dose group; 8 with stage I or II in low-dose group. 20 with stage III or IV in high dose group; 18 with stage III or IV in low dose group. Inclusion criteria: 1. A history of chronic cholestatic liver disease. 2. Liver biopsies were compatible with PBC.
Interventions	High-dose group (n = 30): 250 mg/day increased gradually until 750 mg/day was achieved. Low-dose group (n = 26): 250 mg/day. Mean period of treatment and follow-up: three years.
Outcomes	1. Mortality data (only total number for two groups). 2. Liver test results. 3. Liver biopsy findings. 4. Adverse effects.
Notes	1. Liver test results were analysed as logarithms due to log-normal distribution of data and reported as per cent change. Therefore, it is not possible for us to extract the data. 2. Correspondence sent to the author on 2 December 2003. Reply was received on 13 February 2004. No additional information were added.
Allocation concealment	D – Not used

Study	**Dickson 1985**
Methods	Generation of allocation sequence: adequate, a table of random numbers. Allocation concealment: adequate, a central pharmacist. Blinding: adequate, identical placebo. Follow-up: adequate, 24 patients in the D-penicillamine and no patient in the placebo group withdrew from this trial.
Participants	Country: USA. Mean age: not reported, but 43% patients in D-penicillamine and 54% in placebo not older than 50 years. Female/Male: 200/27. PBC stage status: 3 and 4. Inclusion criteria: 1. Established liver disease of more than six months' duration. 2. Raised alkaline phosphatases more than 2.5 times the normal level. 3. AMA titer greater than 1:10.

Characteristics of included studies (*Continued*)

	4. Liver biopsy diagnostic of, or consistent with PBC.
Interventions	D-penicillamine 250 mg/day for 2 weeks increased by 250 mg/day every 2 weeks until 1 g/day (n = 111) Placebo (the administration same as D-penicillamine) (n = 116) Median period of follow-up: 5 years. Analysed duration of trial: 10 years.
Outcomes	1. Survival analysis. 2. Clinical and biochemical changes. 3. Histologic results. 4. Toxicity.
Notes	1. Survival data at 5 years were available to be extracted only. 2. Correspondence sent to the author on 2 December 2003. No reply was received by 20 June 2004.
Allocation concealment	A – Adequate

Study	**Epstein 1981**
Methods	Generation of allocation sequence: unclear. Allocation concealment: unclear. Blinding: adequate, identical placebo. Follow-up: inadequate.
Participants	Country: UK. Mean age: not reported, instead, median age: 52 years in D-penicillamine, 54 years in placebo. Female/Male: not reported. PBC stage status: 18 with stage 1 or 2 and 37 with stage 3 or 4 in D-penicillamine; 9 with stage 1 or 2 and 23 with stage 3 or 4 in placebo. Inclusion criteria: 1. Liver test pointed to cholestasis. 2. AMA test positive. 3. Normal extrahepatic bile ducts by cholangiography. 4. Liver histology either diagnostic of or highly suggestive of PBC.
Interventions	D-penicillamine: over 8 to 10 weeks from 150 mg/day to 600 mg/day (n = 61). Placebo (n = 37). Median period follow-up: 33 months. Analysed duration of trial: 6 years.
Outcomes	1. Survival data. 2. Liver biochemical variables. 3. Liver histology.
Notes	1. The trial has recruited 98 patients, but data on adverse events were only reported for 87 patients (55 in D-penicillamine and 32 in placebo group). 2. Because of expected withdrawals due to D-penicillamine drug reactions, the randomisation was weighted to allow a 3:2 ratio of D-penicillamine to placebo treated patients. 2. Correspondence sent to the author on 2 December 2003. No reply was received by 20 June 2004.
Allocation concealment	B – Unclear

Study	**Matloff 1982**
Methods	Generation of allocation sequence: unclear. Allocation concealment: adequate, a study monitor. Blinding: adequate, identical placebo. Follow-up: adequate, nine patients in the D-penicillamine group and no patients in the placebo group were lost to follow-up.

Characteristics of included studies (*Continued*)

Participants	Country: USA. Mean age: 51.5 years in D-penicillamine, 51.5 years in placebo. Female/Male: 48 /4. PBC stage status: 14 patients with advanced disease in D-penicillamine, 13 in placebo. Inclusion criteria: 1. A history of chronic cholestatic liver disease. 2. Raised alkaline phosphatases. 3. patent extrahepatic bile ducts. 4. Liver specimen diagnostic of or consistent with primary biliary cirrhosis. 5. AMA test positive.
Interventions	D-penicillamine 1g/day (n = 26). Placebo: identical placebo (n = 26). Total treatment duration: 28 months.
Outcomes	1. Survival data. 2. Liver histology. 3. Liver biochemical variables. 4. Adverse events.
Notes	1. Because a high incidence of side effects was noted in the first 39 patients, the last 13 patients were begun on a dose of 250 mg per day, which was gradually increased to 1g per day over a six-week period. 2. Correspondence sent to the author on 2 December 2003. Reply was received on 19 December 2003. No additional information was added.
Allocation concealment	A – Adequate

Study	**Neuberger 1985**
Methods	Generation of allocation sequence: unclear. Allocation concealment: unclear. Blinding: inadequate. Follow-up: adequate, 35 patients in the D-penicillamine and seven patients in the placebo group were lost to follow-up.
Participants	Country: UK, Spain, Denmark. Mean age: not reported. Female/Male: 173/16. PBC stage status: 12% patients in D-penicillamine and 15% in placebo with stage 1; 40% in D-penicillamine and 37% in placebo with stage 2; 24% in D-penicillamine and 21% in placebo with stage 3; 24% in D-penicillamine and 27% in placebo with stage 4. Inclusion criteria: 1. A clinical and histological picture compatible with that of primary biliary cirrhosis. 2. Raised alkaline phosphatase in the absence of evidence of extrahepatic biliary obstruction.
Interventions	D-penicillamine 1.2 g/day, increased from 300 mg by 300 mg each fortnight until 1.2 g (n = 98) Placebo, taken in the same way (n = 91). Analysed duration of trial: 4 years.
Outcomes	1. Clinical features. 2. Liver biochemical variables. 3, Liver histology. 4. Survival data. 5. Adverse events.
Notes	1. It was an international multicentre (3 centres) trial. 2. Correspondence with the author 2 December 2003. Reply was received on 3 December 2003. No additional information was added.
Allocation concealment	B – Unclear

Study	Taal 1983
Methods	Generation of allocation sequence: unclear. Allocation concealment: unclear. Blinding: adequate, identical placebo. Follow-up: adequate, 2 in the D-penicillamine and 4 in the placebo group were lost to follow-up.
Participants	Country: Netherlands. Median age: 51 years in D-penicillamine, 48 years in placebo. Female/Male: 23/1. PBC stage status: not reported. Inclusion criteria: 1. Raised serum alkaline phosphatases. 2. AMA test positive. 3. A liver biopsy showing lymphoplasmacellular infiltrates with destruction of interlobular bile ducts or a paucity of bile ducts, and no demonstrable abnormalities of the extrahepatic bile ducts on the cholangiogram. 4. Only symptomatic patients (fatigue, pruritus, and/or jaundice).
Interventions	D-penicillamine 1 g/day (increased from 250 mg every month until 1 g for the first 6 months. After that, decreased to 500 mg/day for the remaining 6 months (n = 11). Placebo taken in the same way (n = 13). Duration of treatment: 1 year Duration of post-treatment follow-up: 0.5 year. Analysed duration of trial: 1.5 years.
Outcomes	1. Survival data. 2. PBC-related symptoms. 3. Liver biochemical variables. 4. Liver histological variables. 5. Adverse events.
Notes	1. It involved two centres. 2. Correspondence sent to the author on 2 December 2003. Reply was received on 3 December 2003. No additional information was added.
Allocation concealment	B – Unclear

PBC: primary biliary cirrhosis
AMA: antimitochondrial antibody

Characteristics of excluded studies

Study	Reason for exclusion
Gupta 1982	An observational study, examing for three years the serum levels of immune complexes from 88 patient with primary biliary cirrhosis, treated with D-penicillamine.
Savolainen 1983	Non-randomised clinical study.
Triger 1980	Non-randomised clinical study.

ADDITIONAL TABLES

Table 01. Search strategy for identification of studies

Database	Period	Search strategy
The Cochrane Hepato-Biliary Group Controlled Trials Register	September 2003	#1= 'PRIMARY BILIARY CIRRHOSIS' and 'D-PENICILLAMINE'
The Cochrane Central Register of Controlled Trials on The Cochrane Library	Issue 3, 2003	#1 = LIVER CIRRHOSIS BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = pbc #5 = #1 or #2 or #3 or #4 #6 = PENICILLAMINE: MESH #7 = CHELATING AGENTS: MESH #8 = penicillamine #9 = chelating next agent* #10 = #6 or #7 or #8 or #9 #11 = #5 and #10
MEDLINE	January 1966 to September 2003	#1 = Liver-Cirrhosis-Biliary: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = PBC #5 = #1 or #2 or #3 or #4 #6 = Penicillamine: MESH #7 = Chelating-Agents: MESH #8 = penicillamine #9 = chelating agent* #10 = #6 or #7 or #8 or #9 #11 = #5 and #10 #12 = random* or placebo* or blind* or meta-analysis #13 = #11 and #12
EMBASE	January 1980 to September 2003.	#1 = primary-biliary-cirrhosis: MESH #2 = biliary-cirrhosis: MESH #3 = primary and biliary and cirrhosis #4 = primary biliary cirrhosis #5 = PBC #6 = #1 or #2 or #3 or #4 or #5 #7 = penicillamine: MESH #8 = chelating-agent: MESH #9 = penicillamine #10 = chelating agent* #11 = #7 or #8 or #9 or #10 #12 = #6 and #11 #13 = random* or placebo* or blind* or meta-analysis #14 = #12 and #13
Chinese Biochemical CD Database	January 1979 to August 2003	#1 = Liver-Cirrhosis-Biliary: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = PBC

Table 01. Search strategy for identification of studies (*Continued*)

Database	Period	Search strategy
		#5 = #1 or #2 or #3 or #4 #6 = Penicillamine: MESH #7 = Chelating-Agents: MESH #8 = penicillamine #9 = chelating agent* #10 = #6 or #7 or #8 or #9 #11 = #5 and #10 #12 = random* or placebo* or blind* or meta-analysis #13 = #11 and #12
LILACS	1982 to 2003	#1 = (primary and biliary and cirrhosis) or (primary biliary cirrhosis) #2 = primary biliary cirrhosis #3 = penicillamine

Table 02. Adverse events in the included trials

Trials	D-penicillamine	Control
Bassendine 1982	Proteinuria, rash, 'lupus' syndrome, myasthenia, thrombocytopenia.	None.
Dickson 1985	Hypersensitivity, cytopenia, arthralgias, linchen planus, loss of taste, proteinuria.	Cytopenia, arthralgias, linchen planus, dysgeusia, proteinuria.
Epstein 1981	Rashes, proteinuria, neutropenia.	None.
Matloff 1982	Goodpasture-like syndrome, myasthenia, proteinuria, linchen planus, arthralgias, splenomegaly, rash, loss of taste, stomatitis.	Proteinuria.
Neuberger 1985	Rash, proteinuria, thrombocytopenia, arthralgia, gastrointestinal upset, leucopenia, asthma, pemphigoid, loss of taste, psychosis, palpitations, non-compliance.	Proteinuria, gastrointestinal upset, headaches, non-compliance, neurological complications.
Taal 1983	Exanthema, gastrointestinal upset, loss of taste.	Exanthema, gastrointestinal upset.
Bodenheimer 1985	In 750 mg/day (high-dose) group: Fever, rash, arthralgia, loss of taste, mouth ulcers, nausea, haemolysis, thrombocytopenia, neutropenia, pulmonary fibrosis, albuminuria, neuropathy. In 250 mg/day (low-dose) group: Fever, rash, arthralgia, loss of taste, mouth ulcers, thrombocytopenia, neutropenia, pulmonary fibrosis, albuminuria, neuropathy.	

ANALYSES

Comparison 01. D-penicillamine versus placebo/no intervention

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Mortality (expressed as relative risk) - fixed effect model	6	628	Relative Risk (Fixed) 95% CI	1.34 [1.09, 1.64]
02 Mortality (expressed as relative risk) - random effects model	6	628	Relative Risk (Random) 95% CI	1.46 [0.85, 2.50]
05 Subgroups of methodological quality - generation of allocation sequence - mortality			Relative Risk (Fixed) 95% CI	Subtotals only
06 Subgroups of methodological quality - allocation concealment - mortality	6	628	Relative Risk (Fixed) 95% CI	1.34 [1.09, 1.64]
07 Subgroups of methodological quality - blinding - mortality	6	628	Relative Risk (Fixed) 95% CI	1.34 [1.09, 1.64]
08 Subgroups of methodological quality - follow-up - mortality	6	628	Relative Risk (Fixed) 95% CI	1.34 [1.09, 1.64]
09 Subgroups of dosage - mortality			Relative Risk (Fixed) 95% CI	Subtotals only
10 Subgroups of treatment and follow-up duration - mortality	6	628	Relative Risk (Fixed) 95% CI	1.34 [1.09, 1.64]
11 Subgroups of PBC histological stage - mortality			Relative Risk (Fixed) 95% CI	Subtotals only
12 Sensitivity analyses - mortality			Relative Risk (Fixed) 95% CI	Subtotals only
13 Mortality or liver transplantation - fixed effect model	6	628	Relative Risk (Fixed) 95% CI	1.33 [1.09, 1.63]
14 Mortality or liver transplantation - random effects model	6	628	Relative Risk (Random) 95% CI	1.45 [0.85, 2.48]
15 Patients without improvement of pruritus	1	189	Relative Risk (Fixed) 95% CI	0.57 [0.33, 0.99]
16 Patients without improvement of liver complications			Relative Risk (Fixed) 95% CI	Subtotals only
17 Liver histology			Relative Risk (Fixed) 95% CI	Subtotals only
18 Bilirubin (µmol/L)	1	29	Weighted Mean Difference (Fixed) 95% CI	49.00 [-43.44, 141.44]
19 Alkaline phosphatases (IU/L)	1	29	Weighted Mean Difference (Fixed) 95% CI	-62.50 [-294.67, 169.67]
20 Aspartate aminotransferase (IU/L)	1	30	Weighted Mean Difference (Fixed) 95% CI	-38.00 [-79.82, 3.82]
21 Alanine aminotransferase (IU/L)	1	22	Weighted Mean Difference (Fixed) 95% CI	-45.00 [-75.11, -14.89]
22 Albumin (g/dL)	1	29	Weighted Mean Difference (Fixed) 95% CI	-0.50 [-1.04, 0.04]
23 Adverse event - fixed effect model	6	617	Relative Risk (Fixed) 95% CI	3.11 [2.33, 4.16]
24 Adverse event - random effects model	6	617	Relative Risk (Random) 95% CI	4.18 [1.38, 12.69]

25 Adverse event - excluding Taal 1983 trial	5	593	Relative Risk (Fixed) 95% CI	3.69 [2.62, 5.19]

Comparison 02. High-dose D-penicillamine versus low-dose D-penicillamine

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Mortality	1	41	Relative Risk (Fixed) 95% CI	0.26 [0.06, 1.05]
02 Patients without improvement of liver histological progression	1	34	Relative Risk (Fixed) 95% CI	0.63 [0.31, 1.29]
03 Adverse event	1	40	Relative Risk (Fixed) 95% CI	1.99 [0.81, 4.89]

INDEX TERMS

Medical Subject Headings (MeSH)

Chelating Agents [*adverse effects]; Liver Cirrhosis, Biliary [*drug therapy; mortality]; Penicillamine [*adverse effects]; Randomized Controlled Trials

MeSH check words

Humans

COVER SHEET

Title	D-penicillamine for primary biliary cirrhosis
Authors	Gong Y, Frederiksen SL, Gluud C
Contribution of author(s)	YG performed the searches, selected trials for inclusion, wrote to authors and pharmaceutical companies, performed data extraction and data analyses, and drafted the protocol and the systematic review. SLF modified the search strategy, extracted data, and revised the protocol and the systematic review. CG formulated the idea of this review and revised the protocol, solved discrepancy of data extraction, validated data analyses, and revised the review.
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What's New	Information not supplied by author
Date new studies sought but none found	Information not supplied by author
Date new studies found but not yet included/excluded	Information not supplied by author
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Contact address	Dr Yan Gong Copenhagen Trial Unit

Centre for Clinical Intervention Research, Copenhagen University Hospital
Dept. 7102, Blegdamsvej 9
H:S Rigshospitalet
Copenhagen
DK-2100
DENMARK
E-mail: ygong@ctu.rh.dk
Tel: +45 3545 7161
Fax: +45 3545 7101

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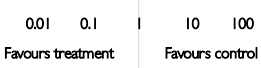
GRAPHS AND OTHER TABLES

Analysis 01.01. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 01 Mortality (expressed as relative risk) - fixed effect model

Review: D-penicillamine for primary biliary cirrhosis
Comparison: 01 D-penicillamine versus placebo/no intervention
Outcome: 01 Mortality (expressed as relative risk) - fixed effect model

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Bassendine 1982	11/19	2/19		1.9	5.50 [1.40, 21.56]
Dickson 1985	44/111	46/116		43.4	1.00 [0.73, 1.38]
Epstein 1981	18/61	16/37		19.2	0.68 [0.40, 1.17]
Matloff 1982	16/26	3/26		2.9	5.33 [1.76, 16.13]
Neuberger 1985	53/98	29/91		29.0	1.70 [1.19, 2.41]
Taal 1983	2/11	4/13		3.5	0.59 [0.13, 2.64]
Total (95% CI)	326	302		100.0	1.34 [1.09, 1.64]

Total events: 144 (D-penicillamine), 100 (Control)
Test for heterogeneity chi-square=22.27 df=5 p=0.0005 I^2 =77.5%
Test for overall effect z=2.82 p=0.005



Analysis 01.02. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 02 Mortality (expressed as relative risk) - random effects model

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

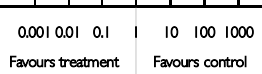
Outcome: 02 Mortality (expressed as relative risk) - random effects model

Study	D-penicillamine n/N	Control n/N	Relative Risk (Random) 95% CI	Weight (%)	Relative Risk (Random) 95% CI
Bassendine 1982	11/19	2/19		9.8	5.50 [1.40, 21.56]
Dickson 1985	44/111	46/116		24.2	1.00 [0.73, 1.38]
Epstein 1981	18/61	16/37		21.0	0.68 [0.40, 1.17]
Matloff 1982	16/26	3/26		12.5	5.33 [1.76, 16.13]
Neuberger 1985	53/98	29/91		23.8	1.70 [1.19, 2.41]
Taal 1983	2/11	4/13		8.7	0.59 [0.13, 2.64]
Total (95% CI)	326	302		100.0	1.46 [0.85, 2.50]

Total events: 144 (D-penicillamine), 100 (Control)

Test for heterogeneity chi-square=22.27 df=5 p=0.0005 I^2 =77.5%

Test for overall effect z=1.37 p=0.2



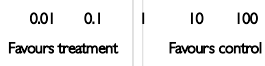
Analysis 01.05. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 05 Subgroups of methodological quality - generation of allocation sequence - mortality

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 05 Subgroups of methodological quality - generation of allocation sequence - mortality

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Adequate generation of allocation sequence					
Dickson 1985	44/111	46/116		100.0	1.00 [0.73, 1.38]
Subtotal (95% CI)	111	116		100.0	1.00 [0.73, 1.38]
Total events: 44 (D-penicillamine), 46 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=0.00 p=1					
02 Unclear or inadequate generation of allocation sequence					
Bassendine 1982	11/19	2/19		3.4	5.50 [1.40, 21.56]
Epstein 1981	18/61	16/37		34.0	0.68 [0.40, 1.17]
Matloff 1982	16/26	3/26		5.1	5.33 [1.76, 16.13]



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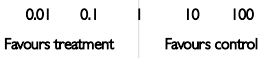
Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Neuberger 1985	53/98	29/91		51.3	1.70 [1.19, 2.41]
Taal 1983	2/11	4/13		6.3	0.59 [0.13, 2.64]
Subtotal (95% CI)	215	186		100.0	1.60 [1.23, 2.08]
Total events: 100 (D-penicillamine), 54 (Control)					
Test for heterogeneity chi-square=19.22 df=4 p=0.0007 I² =79.2%					
Test for overall effect z=3.48 p=0.0005					



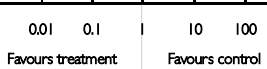
Analysis 01.06. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 06 Subgroups of methodological quality - allocation concealment - mortality

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 06 Subgroups of methodological quality - allocation concealment - mortality

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Adequate allocation concealment					
Dickson 1985	44/111	46/116		43.4	1.00 [0.73, 1.38]
Matloff 1982	16/26	3/26		2.9	5.33 [1.76, 16.13]
Subtotal (95% CI)	137	142		46.3	1.27 [0.94, 1.72]
Total events: 60 (D-penicillamine), 49 (Control)					
Test for heterogeneity chi-square=8.60 df=1 p=0.003 I² =88.4%					
Test for overall effect z=1.55 p=0.1					
02 Unclear or inadequate allocation concealment					
Bassendine 1982	11/19	2/19		1.9	5.50 [1.40, 21.56]
Epstein 1981	18/61	16/37		19.2	0.68 [0.40, 1.17]
Neuberger 1985	53/98	29/91		29.0	1.70 [1.19, 2.41]
Taal 1983	2/11	4/13		3.5	0.59 [0.13, 2.64]
Subtotal (95% CI)	189	160		53.7	1.40 [1.06, 1.84]
Total events: 84 (D-penicillamine), 51 (Control)					
Test for heterogeneity chi-square=13.20 df=3 p=0.004 I² =77.3%					
Test for overall effect z=2.39 p=0.02					
Total (95% CI)	326	302		100.0	1.34 [1.09, 1.64]
Total events: 144 (D-penicillamine), 100 (Control)					
Test for heterogeneity chi-square=22.27 df=5 p=0.0005 I² =77.5%					
Test for overall effect z=2.82 p=0.005					

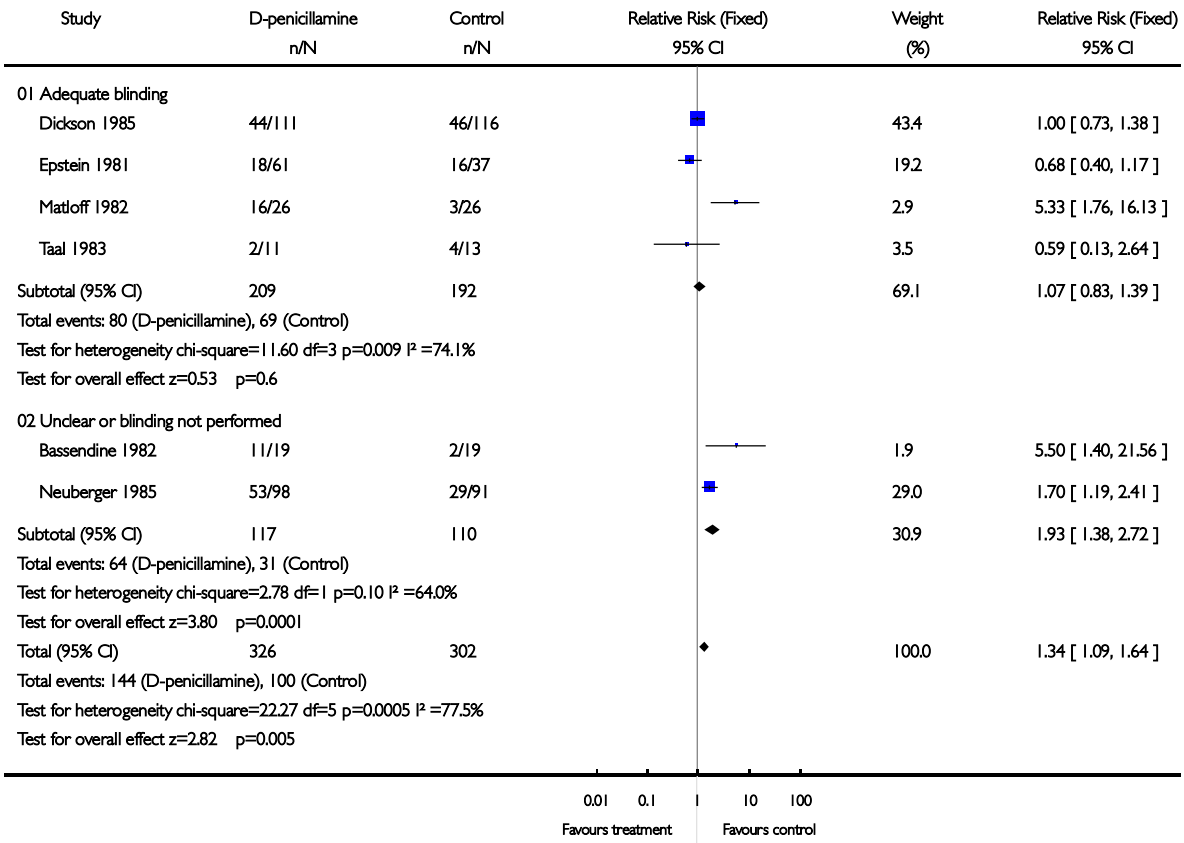


Analysis 01.07. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 07 Subgroups of methodological quality - blinding - mortality

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 07 Subgroups of methodological quality - blinding - mortality



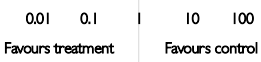
Analysis 01.08. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 08 Subgroups of methodological quality - follow-up - mortality

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 08 Subgroups of methodological quality - follow-up - mortality

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Adequate follow-up					
Bassendine 1982	11/19	2/19		1.9	5.50 [1.40, 21.56]
Dickson 1985	44/111	46/116		43.4	1.00 [0.73, 1.38]
Matloff 1982	16/26	3/26		2.9	5.33 [1.76, 16.13]
Neuberger 1985	53/98	29/91		29.0	1.70 [1.19, 2.41]
Taal 1983	2/11	4/13		3.5	0.59 [0.13, 2.64]
Subtotal (95% CI)	265	265		80.8	1.50 [1.20, 1.87]
Total events: 126 (D-penicillamine), 84 (Control)					
Test for heterogeneity chi-square=16.58 df=4 p=0.002 I² =75.9%					
Test for overall effect z=3.55 p=0.0004					
02 Unclear or inadequate follow-up					
Epstein 1981	18/61	16/37		19.2	0.68 [0.40, 1.17]
Subtotal (95% CI)	61	37		19.2	0.68 [0.40, 1.17]
Total events: 18 (D-penicillamine), 16 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.40 p=0.2					
Total (95% CI)	326	302		100.0	1.34 [1.09, 1.64]
Total events: 144 (D-penicillamine), 100 (Control)					
Test for heterogeneity chi-square=22.27 df=5 p=0.0005 I² =77.5%					
Test for overall effect z=2.82 p=0.005					



Analysis 01.09. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 09 Subgroups of dosage - mortality

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 09 Subgroups of dosage - mortality

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 D-penicillamine 1.2 g/day					
Neuberger 1985	53/98	29/91		100.0	1.70 [1.19, 2.41]
Subtotal (95% CI)	98	91		100.0	1.70 [1.19, 2.41]
Total events: 53 (D-penicillamine), 29 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=2.95 p=0.003					
02 D-penicillamine 1 g/day					
Bassendine 1982	11/19	2/19		3.7	5.50 [1.40, 21.56]
Dickson 1985	44/111	46/116		83.8	1.00 [0.73, 1.38]
Matloff 1982	16/26	3/26		5.6	5.33 [1.76, 16.13]
Taal 1983	2/11	4/13		6.8	0.59 [0.13, 2.64]
Subtotal (95% CI)	167	174		100.0	1.38 [1.04, 1.84]
Total events: 73 (D-penicillamine), 55 (Control)					
Test for heterogeneity chi-square=14.79 df=3 p=0.002 I^2 =79.7%					
Test for overall effect z=2.21 p=0.03					
03 D-penicillamine 0.6 g/day					
Epstein 1981	18/61	16/37		100.0	0.68 [0.40, 1.17]
Subtotal (95% CI)	61	37		100.0	0.68 [0.40, 1.17]
Total events: 18 (D-penicillamine), 16 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.40 p=0.2					

Analysis 01.10. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 10 Subgroups of treatment and follow-up duration - mortality

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 10 Subgroups of treatment and follow-up duration - mortality

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Long-term treatment and long-term follow-up					
Dickson 1985	44/111	46/116		43.4	1.00 [0.73, 1.38]
Epstein 1981	18/61	16/37		19.2	0.68 [0.40, 1.17]
Neuberger 1985	53/98	29/91		29.0	1.70 [1.19, 2.41]
Subtotal (95% CI)	270	244		91.6	1.15 [0.93, 1.43]
Total events: 115 (D-penicillamine), 91 (Control)					
Test for heterogeneity chi-square=9.09 df=2 p=0.01 I^2 =78.0%					
Test for overall effect z=1.31 p=0.2					
02 Short-term treatment and short-term follow-up					
Bassendine 1982	11/19	2/19		1.9	5.50 [1.40, 21.56]
Matloff 1982	16/26	3/26		2.9	5.33 [1.76, 16.13]
Taal 1983	2/11	4/13		3.5	0.59 [0.13, 2.64]
Subtotal (95% CI)	56	58		8.4	3.37 [1.70, 6.66]
Total events: 29 (D-penicillamine), 9 (Control)					
Test for heterogeneity chi-square=6.36 df=2 p=0.04 I^2 =68.6%					
Test for overall effect z=3.49 p=0.0005					
Total (95% CI)	326	302		100.0	1.34 [1.09, 1.64]
Total events: 144 (D-penicillamine), 100 (Control)					
Test for heterogeneity chi-square=22.27 df=5 p=0.0005 I^2 =77.5%					
Test for overall effect z=2.82 p=0.005					

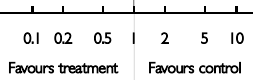
Analysis 01.11. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 11 Subgroups of PBC histological stage - mortality

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 11 Subgroups of PBC histological stage - mortality

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 PBC stage III or IV					
Dickson 1985	44/111	46/116		78.5	1.00 [0.73, 1.38]
Epstein 1981	5/37	10/23		21.5	0.31 [0.12, 0.79]
Subtotal (95% CI)	148	139		100.0	0.85 [0.63, 1.15]
Total events: 49 (D-penicillamine), 56 (Control)					
Test for heterogeneity chi-square=5.38 df=1 p=0.02 I^2 =81.4%					
Test for overall effect z=1.05 p=0.3					
02 PBC stage I or II					
× Epstein 1981	0/18	0/9		0.0	Not estimable
Subtotal (95% CI)	18	9		0.0	Not estimable
Total events: 0 (D-penicillamine), 0 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect: not applicable					



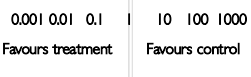
Analysis 01.12. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 12 Sensitivity analyses - mortality

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 12 Sensitivity analyses - mortality

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Available patient course analysis					
Bassendine 1982	2/10	2/19		2.0	1.90 [0.31, 11.54]
Dickson 1985	28/95	25/95		37.0	1.12 [0.71, 1.77]
Epstein 1981	18/61	16/37		29.5	0.68 [0.40, 1.17]
Matloff 1982	7/17	3/26		3.5	3.57 [1.07, 11.93]
Neuberger 1985	18/63	22/84		27.9	1.09 [0.64, 1.85]
× Taal 1983	0/9	0/9		0.0	Not estimable
Subtotal (95% CI)	255	270		100.0	1.08 [0.82, 1.43]



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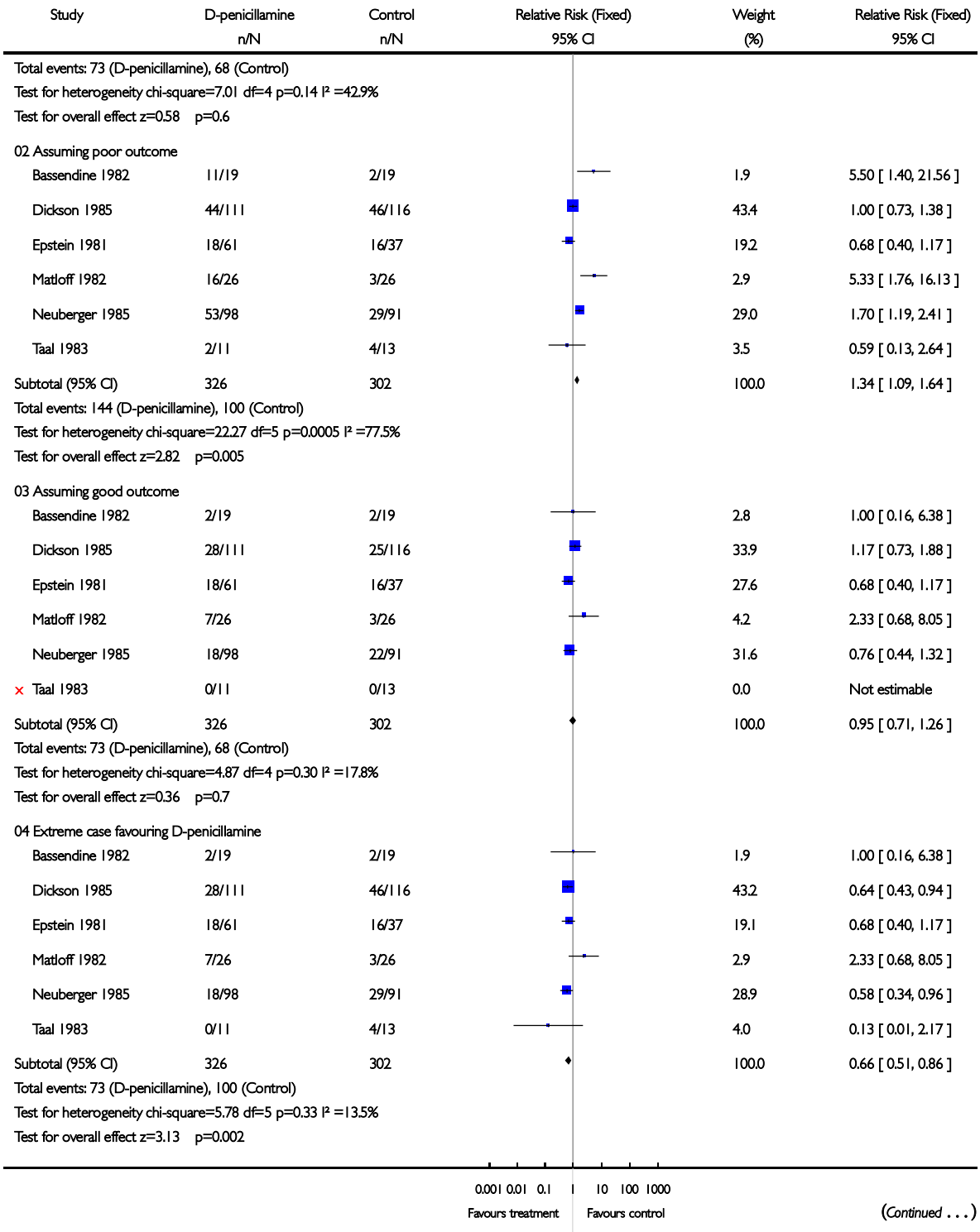

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Total events: 73 (D-penicillamine), 68 (Control)					
Test for heterogeneity chi-square=7.01 df=4 p=0.14 I² =42.9%					
Test for overall effect z=0.58 p=0.6					
02 Assuming poor outcome					
Bassendine 1982	11/19	2/19		1.9	5.50 [1.40, 21.56]
Dickson 1985	44/111	46/116		43.4	1.00 [0.73, 1.38]
Epstein 1981	18/61	16/37		19.2	0.68 [0.40, 1.17]
Matloff 1982	16/26	3/26		2.9	5.33 [1.76, 16.13]
Neuberger 1985	53/98	29/91		29.0	1.70 [1.19, 2.41]
Taal 1983	2/11	4/13		3.5	0.59 [0.13, 2.64]
Subtotal (95% CI)	326	302		100.0	1.34 [1.09, 1.64]
Total events: 144 (D-penicillamine), 100 (Control)					
Test for heterogeneity chi-square=22.27 df=5 p=0.0005 I² =77.5%					
Test for overall effect z=2.82 p=0.005					
03 Assuming good outcome					
Bassendine 1982	2/19	2/19		2.8	1.00 [0.16, 6.38]
Dickson 1985	28/111	25/116		33.9	1.17 [0.73, 1.88]
Epstein 1981	18/61	16/37		27.6	0.68 [0.40, 1.17]
Matloff 1982	7/26	3/26		4.2	2.33 [0.68, 8.05]
Neuberger 1985	18/98	22/91		31.6	0.76 [0.44, 1.32]
× Taal 1983	0/11	0/13		0.0	Not estimable
Subtotal (95% CI)	326	302		100.0	0.95 [0.71, 1.26]
Total events: 73 (D-penicillamine), 68 (Control)					
Test for heterogeneity chi-square=4.87 df=4 p=0.30 I² =17.8%					
Test for overall effect z=0.36 p=0.7					
04 Extreme case favouring D-penicillamine					
Bassendine 1982	2/19	2/19		1.9	1.00 [0.16, 6.38]
Dickson 1985	28/111	46/116		43.2	0.64 [0.43, 0.94]
Epstein 1981	18/61	16/37		19.1	0.68 [0.40, 1.17]
Matloff 1982	7/26	3/26		2.9	2.33 [0.68, 8.05]
Neuberger 1985	18/98	29/91		28.9	0.58 [0.34, 0.96]
Taal 1983	0/11	4/13		4.0	0.13 [0.01, 2.17]
Subtotal (95% CI)	326	302		100.0	0.66 [0.51, 0.86]
Total events: 73 (D-penicillamine), 100 (Control)					
Test for heterogeneity chi-square=5.78 df=5 p=0.33 I² =13.5%					
Test for overall effect z=3.13 p=0.002					

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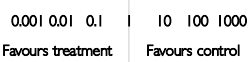
Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
05 Extreme case favouring control					
Bassendine 1982	11/19	2/19		2.8	5.50 [1.40, 21.56]
Dickson 1985	44/111	25/116		33.7	1.84 [1.21, 2.79]
Epstein 1981	18/61	16/37		27.4	0.68 [0.40, 1.17]
Matloff 1982	16/26	3/26		4.1	5.33 [1.76, 16.13]
Neuberger 1985	53/98	22/91		31.4	2.24 [1.49, 3.36]
Taal 1983	2/11	0/13		0.6	5.83 [0.31, 109.99]
Subtotal (95% CI)	326	302		100.0	1.92 [1.51, 2.43]
Total events: 144 (D-penicillamine), 68 (Control)					
Test for heterogeneity chi-square=21.01 df=5 p=0.0008 I² =76.2%					
Test for overall effect z=5.37 p<0.00001					



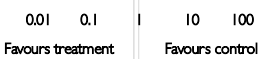
Analysis 01.13. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 13 Mortality or liver transplantation - fixed effect model

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 13 Mortality or liver transplantation - fixed effect model

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Bassendine 1982	11/19	2/19		1.9	5.50 [1.40, 21.56]
Dickson 1985	44/111	46/116		43.0	1.00 [0.73, 1.38]
Epstein 1981	18/61	16/37		19.0	0.68 [0.40, 1.17]
Matloff 1982	16/26	3/26		2.9	5.33 [1.76, 16.13]
Neuberger 1985	54/98	30/91		29.7	1.67 [1.19, 2.36]
Taal 1983	2/11	4/13		3.5	0.59 [0.13, 2.64]
Total (95% CI)	326	302		100.0	1.33 [1.09, 1.63]
Total events: 145 (D-penicillamine), 101 (Control)					
Test for heterogeneity chi-square=22.08 df=5 p=0.0005 I² =77.4%					
Test for overall effect z=2.81 p=0.005					



Analysis 01.14. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 14 Mortality or liver transplantation - random effects model

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

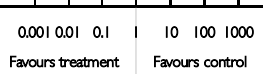
Outcome: 14 Mortality or liver transplantation - random effects model

Study	D-penicillamine n/N	Control n/N	Relative Risk (Random) 95% CI	Weight (%)	Relative Risk (Random) 95% CI
Bassendine 1982	11/19	2/19		9.7	5.50 [1.40, 21.56]
Dickson 1985	44/111	46/116		24.2	1.00 [0.73, 1.38]
Epstein 1981	18/61	16/37		21.0	0.68 [0.40, 1.17]
Matloff 1982	16/26	3/26		12.4	5.33 [1.76, 16.13]
Neuberger 1985	54/98	30/91		23.9	1.67 [1.19, 2.36]
Taal 1983	2/11	4/13		8.6	0.59 [0.13, 2.64]
Total (95% CI)	326	302		100.0	1.45 [0.85, 2.48]

Total events: 145 (D-penicillamine), 101 (Control)

Test for heterogeneity chi-square=22.08 df=5 p=0.0005 I^2 =77.4%

Test for overall effect z=1.36 p=0.2



Analysis 01.15. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 15 Patients without improvement of pruritus

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

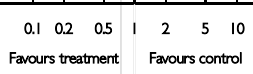
Outcome: 15 Patients without improvement of pruritus

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Neuberger 1985	16/98	26/91		100.0	0.57 [0.33, 0.99]
Total (95% CI)	98	91		100.0	0.57 [0.33, 0.99]

Total events: 16 (D-penicillamine), 26 (Control)

Test for heterogeneity: not applicable

Test for overall effect z=1.98 p=0.05



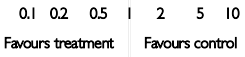
Analysis 01.16. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 16 Patients without improvement of liver complications

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 16 Patients without improvement of liver complications

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Gastrointestinal bleeding					
Neuberger 1985	4/98	8/91		100.0	0.46 [0.14, 1.49]
Subtotal (95% CI)	98	91		100.0	0.46 [0.14, 1.49]
Total events: 4 (D-penicillamine), 8 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.29 p=0.2					
02 Ascites					
Neuberger 1985	14/98	21/91		100.0	0.62 [0.34, 1.14]
Subtotal (95% CI)	98	91		100.0	0.62 [0.34, 1.14]
Total events: 14 (D-penicillamine), 21 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.53 p=0.1					
03 Hepatic encephalopathy					
Subtotal (95% CI)	0	0		0.0	Not estimable
Total events: 0 (D-penicillamine), 0 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect: not applicable					

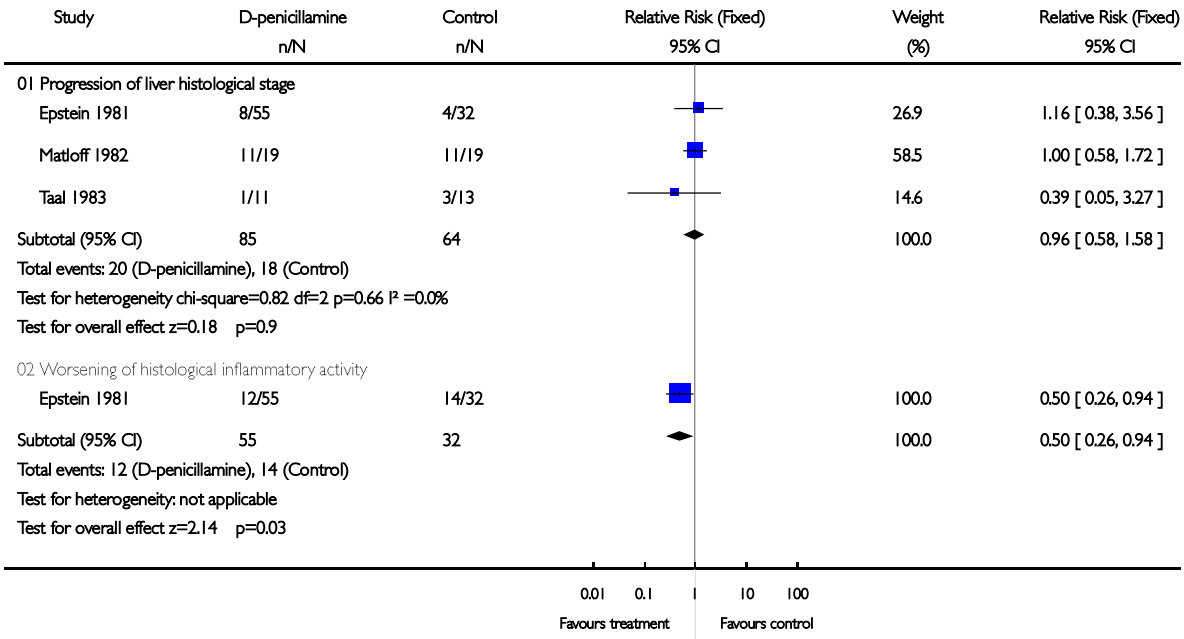


Analysis 01.17. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 17 Liver histology

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 17 Liver histology

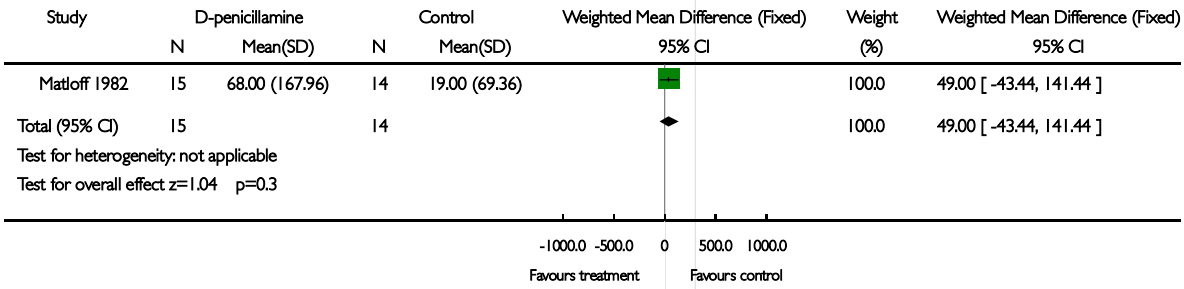


Analysis 01.18. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 18 Bilirubin (ømol/L)

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 18 Bilirubin (µmol/L)

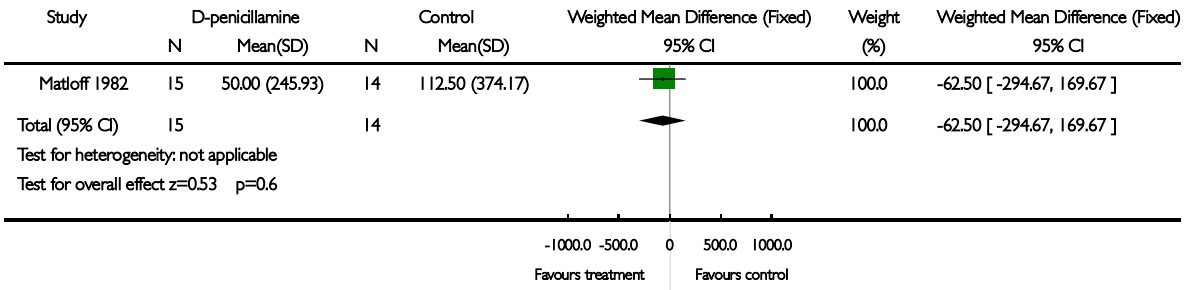


Analysis 01.19. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 19 Alkaline phosphatases (IU/L)

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 19 Alkaline phosphatases (IU/L)

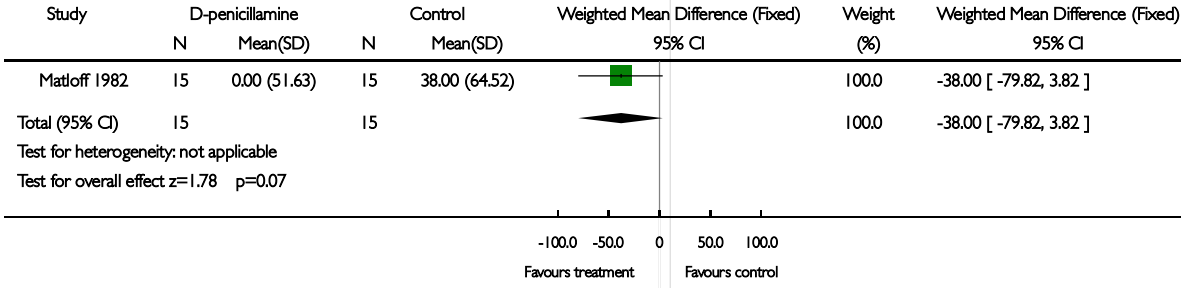


Analysis 01.20. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 20 Aspartate aminotransferase (IU/L)

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 20 Aspartate aminotransferase (IU/L)

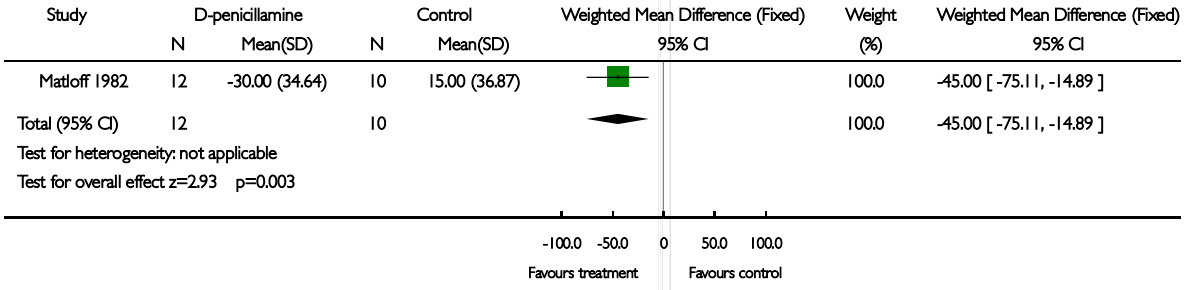


Analysis 01.21. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 21 Alanine aminotransferase (IU/L)

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 21 Alanine aminotransferase (IU/L)

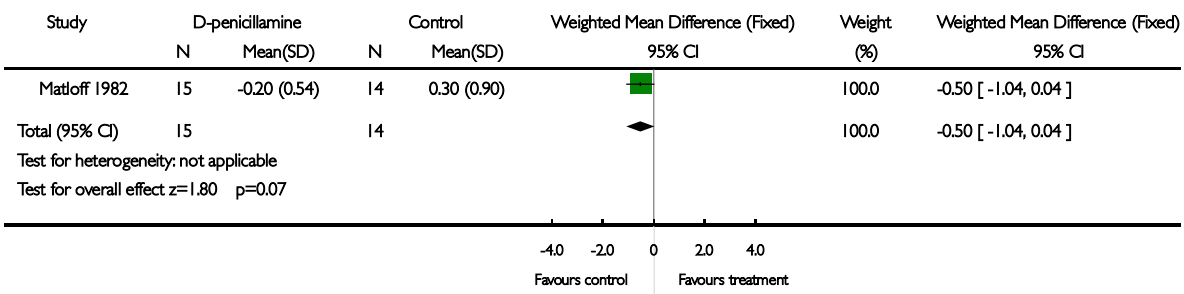


Analysis 01.22. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 22 Albumin (g/dL)

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 22 Albumin (g/dL)

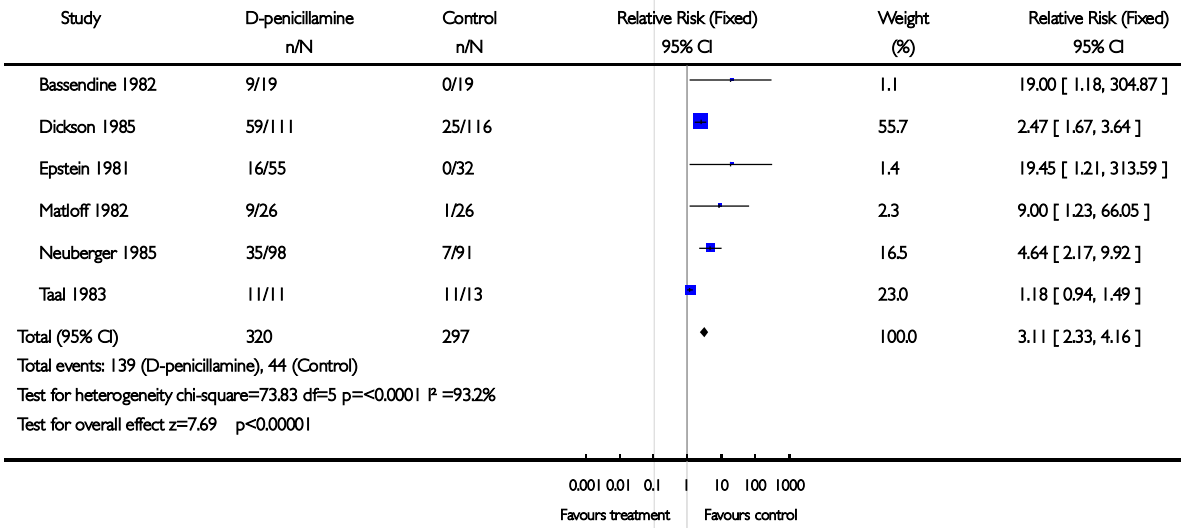


Analysis 01.23. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 23 Adverse event - fixed effect model

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

Outcome: 23 Adverse event - fixed effect model



Analysis 01.24. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 24 Adverse event - random effects model

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

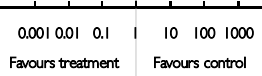
Outcome: 24 Adverse event - random effects model

Study	D-penicillamine n/N	Control n/N	Relative Risk (Random) 95% CI	Weight (%)	Relative Risk (Random) 95% CI
Bassendine 1982	9/19	0/19		9.5	19.00 [1.18, 304.87]
Dickson 1985	59/111	25/116		22.9	2.47 [1.67, 3.64]
Epstein 1981	16/55	0/32		9.5	19.45 [1.21, 313.59]
Matloff 1982	9/26	1/26		13.4	9.00 [1.23, 66.05]
Neuberger 1985	35/98	7/91		21.2	4.64 [2.17, 9.92]
Taal 1983	11/11	11/13		23.4	1.18 [0.94, 1.49]
Total (95% CI)	320	297		100.0	4.18 [1.38, 12.69]

Total events: 139 (D-penicillamine), 44 (Control)

Test for heterogeneity chi-square=73.83 df=5 p=<0.0001 I^2 =93.2%

Test for overall effect z=2.52 p=0.01



Analysis 01.25. Comparison 01 D-penicillamine versus placebo/no intervention, Outcome 25 Adverse event - excluding Taal 1983 trial

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 01 D-penicillamine versus placebo/no intervention

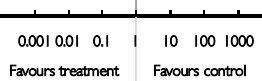
Outcome: 25 Adverse event - excluding Taal 1983 trial

Study	D-penicillamine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Bassendine 1982	9/19	0/19		1.5	19.00 [1.18, 304.87]
Dickson 1985	59/111	25/116		72.3	2.47 [1.67, 3.64]
Epstein 1981	16/55	0/32		1.9	19.45 [1.21, 313.59]
Matloff 1982	9/26	1/26		3.0	9.00 [1.23, 66.05]
Neuberger 1985	35/98	7/91		21.5	4.64 [2.17, 9.92]
Total (95% CI)	309	284		100.0	3.69 [2.62, 5.19]

Total events: 128 (D-penicillamine), 33 (Control)

Test for heterogeneity chi-square=7.95 df=4 p=0.09 I^2 =49.7%

Test for overall effect z=7.46 p<0.00001



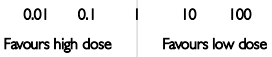
Analysis 02.01. Comparison 02 High-dose D-penicillamine versus low-dose D-penicillamine, Outcome 01 Mortality

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 02 High-dose D-penicillamine versus low-dose D-penicillamine

Outcome: 01 Mortality

Study	High dose n/N	Low dose n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Bassendine 1982	2/19	9/22		100.0	0.26 [0.06, 1.05]
Total (95% CI)	19	22		100.0	0.26 [0.06, 1.05]
Total events: 2 (High dose), 9 (Low dose)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.90 p=0.06					



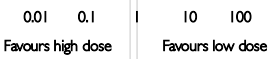
Analysis 02.02. Comparison 02 High-dose D-penicillamine versus low-dose D-penicillamine, Outcome 02 Patients without improvement of liver histological progression

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 02 High-dose D-penicillamine versus low-dose D-penicillamine

Outcome: 02 Patients without improvement of liver histological progression

Study	High dose n/N	Low dose n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Bodenheimer 1985	6/15	12/19		100.0	0.63 [0.31, 1.29]
Total (95% CI)	15	19		100.0	0.63 [0.31, 1.29]
Total events: 6 (High dose), 12 (Low dose)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.26 p=0.2					



Analysis 02.03. Comparison 02 High-dose D-penicillamine versus low-dose D-penicillamine, Outcome 03 Adverse event

Review: D-penicillamine for primary biliary cirrhosis

Comparison: 02 High-dose D-penicillamine versus low-dose D-penicillamine

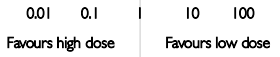
Outcome: 03 Adverse event

Study	High dose n/N	Low dose n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Bassendine 1982	9/19	5/21		100.0	1.99 [0.81, 4.89]
Total (95% CI)	19	21		100.0	1.99 [0.81, 4.89]

Total events: 9 (High dose), 5 (Low dose)

Test for heterogeneity: not applicable

Test for overall effect z=1.50 p=0.1



Appendix 2B

Alimentary Pharmacology & Therapeutics

Systematic review and meta-analysis: D-Penicillamine vs. placebo/no intervention in patients with primary biliary cirrhosis ñ Cochrane Hepato-Biliary Group

Y. GONG, S. L. KLINGENBERG & C. GLUUD

The Cochrane Hepato-Biliary Group, Copenhagen Trial Unit, Centre for Clinical Intervention Research, H:S Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark

Correspondence to:
Dr Y. Gong, The Cochrane Hepato-Biliary Group, Copenhagen Trial Unit, Centre for Clinical Intervention Research, Department 7102, H:S Rigshospitalet, Copenhagen University Hospital, Blegdamsvej 9, 2100 Copenhagen Ø, Denmark.
E-mail: ygong@ctu.rh.dk

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SUMMARY

Background

D-Penicillamine is used for patients with primary biliary cirrhosis due to its ability to decrease hepatic copper and modulate the immune response. The results on effects of D-penicillamine in randomized-clinical trials of primary biliary cirrhosis patients are inconsistent.

Aim

To systematically evaluate the benefits and harms of D-penicillamine for patients with primary biliary cirrhosis.

Methods

We have performed a systematic review with meta-analyses of randomized-clinical trials to evaluate the effects of D-penicillamine for primary biliary cirrhosis. The primary outcomes are mortality and mortality or liver transplantation. We analysed the data by fixed-effect and random-effect models.

Results

Seven randomized trials including 706 patients were analysed. D-Penicillamine was without significant effects on mortality (RR 1.08, 95% CI: 0.82–1.43, $P = 0.56$), mortality or liver transplantation (RR 1.11, 95% CI: 0.74–1.68, $P = 0.62$), pruritus, liver complications, progression of liver histological stage and liver biochemical variables. D-Penicillamine significantly decreased serum alanine aminotransferase activity (weighted mean difference −45 IU/L, 95% CI: −75 to −15, $P < 0.05$) and led to significantly more adverse events (RR 4.18, 95% CI: 1.38–12.69, $P = 0.01$).

Conclusion

D-Penicillamine did not appear to reduce the risk of mortality or morbidity, and led to more adverse events in patients with primary biliary cirrhosis.

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INTRODUCTION

Primary biliary cirrhosis is an uncommon chronic progressive liver disease of unknown aetiology. Over the last 30 years, substantial increases in prevalence have been noted in many countries.[1] Patients with primary biliary cirrhosis have been subjected to several immunosuppressive agents, e.g. prednisolone, azathioprine, chlorambucil, ciclosporin, methotrexate, colchicine and D-penicillamine.[2] The observed clinical effects have not led to widespread acceptance of these drugs.[3]

The major rationale for treating patients with primary biliary cirrhosis is its abilities to induce cupruresis, to inhibit lymphocytes, and to decrease circulating immune complexes.[4–6] There are conflicting reports concerning the effects of D-penicillamine for primary biliary cirrhosis.[7–9] According to a UK survey,[10] nearly 3% of the patients with primary biliary cirrhosis are being treated with D-penicillamine. We, therefore, performed a systematic review and meta-analysis to assess the effects of D-penicillamine in primary biliary cirrhosis.

MATERIALS AND METHODS

Inclusion criteria

We applied The Cochrane Collaboration methodology and followed our predefined, peer-reviewed, published protocol.[11] We only included randomized-clinical trials comparing D-penicillamine with placebo/no intervention in primary biliary cirrhosis patients, irrespective of language, year of publication, or publication status. Patients should have at least two of the following: elevated serum activity of alkaline phosphatases (or other markers of intrahepatic cholestasis), and/or a positive result for serum mitochondrial antibody, and/or liver biopsy findings diagnostic for or compatible with primary biliary cirrhosis. Co-interventions were allowed provided that all intervention groups received similar co-interventions.

Search strategy

We searched for trials in The Cochrane Hepato-Biliary Group Controlled Trials Register, The Cochrane Central Register of Controlled Trials in The Cochrane Library, MEDLINE, EMBASE, Science Citation Index-EXPANDED, The Chinese Biomedical CD Database, LILACS and in references of identified studies. The last search was performed in December 2005. We contacted principal authors and sponsor companies of the identified trials (Eli Lilly, USA and Shionogi & Co., Ltd, Japan) to obtain missing information and additional published or unpublished trials.

Data extraction

Two of the authors (YG and SLK) independently scrutinized all articles and decided which trials to be included. Data from included trials were extracted onto a standard form including three aspects of methodological quality of the trials:[12–14] generation of the allocation sequence, allocation concealment and blinding. Any disagreement about data extraction was resolved by discussion among the authors.

Outcome measures

The primary outcome measures were mortality and mortality or liver transplantation. Our secondary outcome measures were: pruritus, fatigue, liver complications (variceal bleeding, ascites, hepatic encephalopathy, jaundice and hepato-renal syndrome), liver biopsy findings, liver biochemistry variables, adverse events,[15] quality of life and cost-effectiveness.

Data analysis

The meta-analysis was performed with REVIEW MANAGER software (version 4.2) provided by The Cochrane Collaboration (http://www.cochrane.org). We calculated an overall weighted estimate of the relative risk (RR) with a 95% confidence interval (CI) for binary outcomes, and weighted averages of differences between mean values for continuous outcomes. As a sensitivity analysis, we used the uncertainty method to pool the data on primary outcomes in order to allow for missing data because of dropouts.[16] The uncertainty method was developed for incorporating uncertainty, with weights assigned to trials based on uncertainty interval widths. The uncertainty interval for a trial incorporates both sampling error and the potential impact of missing data. We examined intervention effects by a random-effects[17] and a fixed-effect[18] models with the two-sided significance set at $P < 0.05$. We explored the presence of statistical heterogeneity by chi-squared test with significance set at $P < 0.10$ and measured the quantities of heterogeneity by I^2.[19]

We performed subgroup analyses,[20] in which trials were grouped according to the risk of bias, dosage of

D-penicillamine, and duration of treatment and follow-up. We also tried to perform a subgroup analysis as per histological stage, but the data were not reported subgrouped according to histological severity, so we could not evaluate its influence in relation to the effects of D-penicillamine. Trials were considered as low-bias risk trials, if they met two of the three criteria: adequate generation of the allocation sequence, adequate allocation concealment and adequate blinding.[12] Trials not meeting this criterion were considered high-bias risk trials.

RESULTS

Description of the included trials

We identified 178 references through electronic and hand searches. We excluded 146 duplicates and clearly irrelevant references, non-randomized-clinical studies, or observational studies. The remaining 32 references referred to seven randomized-clinical trials including 706 patients (Figure 1). The trial publication with most completed data were regarded as the primary reference, from which data were extracted. Six trials compared D-penicillamine vs. placebo/no intervention.[8, 9, 21–24] Bodenheimer *et al.* compared two different D-penicillamine dosages: 750 mg/day vs. 250 mg/day.[25]

Randomization created comparable intervention groups in the included trials. The baseline characteristics of the patients are summarized in Table 1. The mean age of the patients was 51 years. Most of the patients were women (495 of 548, 90.3%) in the four trials reporting gender. Most of patients had advanced histological stages at entry (stage III or IV/stage I or II: 443 of 168).[8, 22–24] The trial duration, including treatment and follow-up, varied from 1.5 to 10 years. Taal *et al.* explicited the length of treatment and follow-up separately.[9]

Methodological quality of the included studies

The methodological quality of the included trials, including the number of dropouts, is summarized in Table 2. Dickson *et al.* and Matloff *et al.* were regarded as low-bias risk trials.[22, 23] No trials reported sample size estimation. Five trials reported the number of dropouts in D-penicillamine (74 patients) and in control group (16 patients), respectively.[9, 21–24] Bodenheimer *et al.* reported the total number of dropouts in both groups (26 patients).[25] Epstein *et al.* did not report the extractable data on dropouts.[8] There were no trials reporting using intention-to-treat analyses.

Mortality

D-Penicillamine was without significant effects on mortality (RR 1.08, 95% CI: 0.82–1.43, $P = 0.56$, six trials, 525 patients; Figure 2). The RR of mortality allowing for dropouts was 0.92 with an uncertainty interval from 0.61 to 1.38 (Figure 3). The degree of heterogeneity was moderate ($I^2 = 42.9\%$ and 0%, respectively).

We performed subgroup analyses according to methodological quality, dosage of D-penicillamine, duration of treatment and follow-up (shorter or longer than 3 years) and histological stage. The results are summarized in Table 3.

Mortality or liver transplantation

D-Penicillamine did not significantly affect the composite outcome of mortality or liver transplantation (RR 1.09, 95% CI: 0.83–1.43, $P = 0.54$, six trials, 525 patients).

Pruritus, fatigue and liver complications

Neuberger *et al.* observed the benefit of D-penicillamine on pruritus (RR 0.57, 95% CI: 0.33–0.99, $P < 0.05$).[24] Data on fatigue were not extractable. No significant differences between the D-penicillamine and placebo group were found with respect to improvements of gastrointestinal bleeding (RR 0.46, 95% CI: 0.14–1.49, $P = 0.20$, one trial, 189 patients) and ascites (RR 0.62, 95% CI: 0.34–1.14, $P = 0.13$, one trial, 189 patients).

Liver histological and biochemical outcomes

D-Penicillamine did not significantly slow the disease progression. The number of patients advancing to a more severe histological stage did not significantly differ between the D-penicillamine and placebo group (RR 0.96, 95% CI: 0.58–1.58, $P = 0.86$, three trials, 149 patients). However, Epstein *et al.*[8] showed that patients with worsening of inflammatory activity were fewer in the D-penicillamine group than in the placebo group (RR 0.50, 95% CI: 0.26–0.94, $P = 0.03$, one trial, 87 patients). Matloff *et al.*[22] revealed no significant differences on biochemical outcomes between

Table 1. Patient baseline characteristics from included trials of D-penicillamine for patients with primary biliary cirrhosis

Trial	Interventions compared	Mean age (years)	Histological stage	Bilirubin (μM)	Albumin (g/L)	Alkaline phosphatases (IU/L)	Aspartate transaminase (μ/L)	Immunoglobulin M (g/L)
Bassendine *et al.*[21] (UK)	D-Penicillamine 1 g/day ($n = 19$), 250 mg/day ($n = 21$), no intervention ($n = 19$)	Published as an abstract only, and no baseline data could be extracted						
Bodenheimer *et al.*[25] (USA)	D-Penicillamine 750 mg/day ($n = 30$)	52	I & II: 10 III & IV: 20	2.2 ± 0.4 (mg/dL)	Not reported	350 ± 56	Not reported	621 ± 91 (mg/dL)
	D-Penicillamine 250 mg/day ($n = 26$)	52	I & II: 8 III & IV: 18	3.0 ± 0.6 (mg/dL)	Not reported	358 ± 68	Not reported	698 ± 75 (mg/dL)
Dickson *et al.*[23] (USA)	D-Penicillamine 1 g/day ($n = 111$)	43% of patients ≤50	III/IV: 50/61	Not reported	Not reported	Not reported	Not reported	Not reported
	Placebo ($n = 116$)	54% of patients ≤50	III: 53/63	Not reported	Not reported	Not reported	Not reported	Not reported
Epstein *et al.*[8] (UK)	D-Penicillamine 600 mg/day ($n = 61$)	52	I & II: 18 III & IV: 37	32	Not reported	66 (KAU/L)	51	5.1
	Placebo ($n = 37$)	54	I & II: 9 III & IV: 23	24	Not reported	54 (KAU/L)	42	5.1
Matloff *et al.*[22] (USA)	D-Penicillamine 1 g/day ($n = 26$)	51.5	I & II & III: 12 IV: 14	6.1 ± 1.5 (mg/dL)	3.5 ± 0.1 (mg/dL)	22.4 ± 2.9 (bodasnky U)	118 ± 13 (IU)	Not reported
	Placebo ($n = 26$)	51.5	I & II & III: 13 IV: 13	2.1 ± 0.4 (mg/dL)	3.6 ± 0.1 (mg/dL)	21.7 ± 2.0 (bodansky U)	93 ± 13 (IU)	Not reported
Neuberger *et al.*[24] (UK, Spain, Denmark)	D-Penicillamine 1.2 g/day ($n = 98$)	Not reported	I: 12 II: 38 II: 24 IV: 24	23.9	35.4	502	Not reported	3.6
	Placebo ($n = 91$)	Not reported	I: 14 II: 33 II: 19 IV: 25	21	35.2	504	Not reported	3.5
Taal *et al.*[9] (the Netherlands)	D-Penicillamine 1 g/day ($n = 11$)	51	Not reported	20	45	4.4 (×*N**)	2.9 (×*N**)	6.5
	Placebo ($n = 13$)	48	Not reported	21	43	4.7 (×*N**)	3.4 (×*N**)	4.9

* *N* is the upper limit of normal, defined as the 95th percentile of the normal population of the chemical laboratory.

Table 2. Characteristics of included trials of D-penicillamine for patients with primary biliary cirrhosis

Trial	Trial duration (years)	Generation of allocation sequence	Allocation of concealment	Blinding	Number of patients lost to follow-up	
					D-Penicillamine	Control
Bassendine *et al.*[21]	3	Unclear	Unclear	Not performed	4 (21%)*	5 (23%)†
Bodenheimer *et al.*[25]	3	Unclear	Unclear	Adequate‡	26 in total	
Dickson *et al.*[23]	10	Adequate	Adequate	Adequate‡	24 (22%)	0 (0%)
Epstein *et al.*[8]	6	Unclear	Unclear	Adequate‡	Not reported	Not reported
Matloff *et al.*[22]	28 months	Unclear	Adequate	Adequate‡	9 (35%)	0 (0%)
Neuberger *et al.*[24]	4	Unclear	Unclear	Inadequate	35 (36%)	7 (8%)
Taal *et al.*[9]	3	Unclear	Unclear	Adequate‡	2 (18%)	4 (31%)

* D-Penicillamine 1 g/day.
† D-Penicillamine 250 mg/day.
‡ The trials are considered adequate regarding blinding although none of the trials addressed on smell or taste, but only on appearance of the placebo. We are, therefore, not in a position to say that blinding was in fact sufficiently adequate.[8]

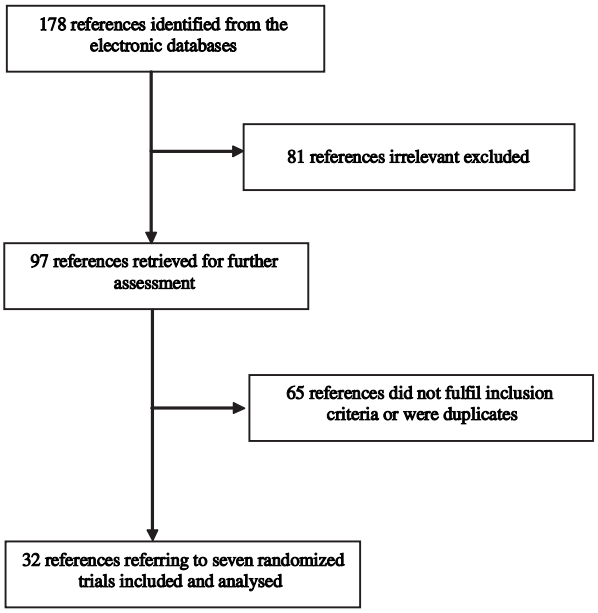

Figure 1. Flow diagram of trial selection.

D-penicillamine and placebo, except for alanine aminotransferase (weighted mean difference −45 IU/L, 95% CI: −75 to −15, $P < 0.05$, one trial, 29 patients).

Adverse events

One hundred and thirty-nine patients (43%) given D-penicillamine had adverse events compared with 44 (15%) patients in control group (RR 4.18, 95% CI: 1.38–12.69, $P = 0.01$, $I^2 = 93.2\%$, six trials, 617 patients; Figure 4 and Table 4). No trials separately reported non-serious or serious adverse events according to the International Conference on Harmonisation – Good Clinical Practise.[15]

Quality of life and cost-effectiveness

None of the trials examined quality of life scales or outcomes regarding cost-effectiveness.

High-dose D-penicillamine vs. low-dose D-penicillamine

In the Bassendine *et al.*'s trial,[21] the risk of mortality tended to be lower in the high-dose group (1 g/day) than in low-dose group (250 mg/day), although the difference was not significant (RR 0.25, 95% CI: 0.06–1.00, $P = 0.05$, one trial, 40 patients). More patients in the high-dose group than in the low-dose group developed adverse events (RR 1.99, 95% CI: 0.81–4.89, $P = 0.13$, one trial, 40 patients).

Bias exploration

Due to the low number of trials included, we did not perform funnel plot analysis and did not apply the statistical methods[26, 27] to detect publication bias and other biases, as the power of those analyses would have been low and inconsistent.

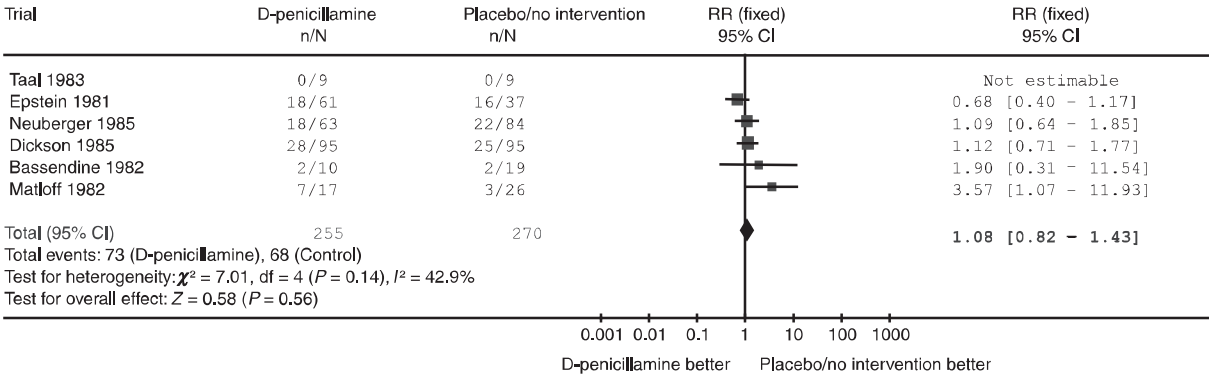

Figure 2. Relative risk (RR) of mortality in primary biliary cirrhosis patients randomized to D-penicillamine vs. placebo/no intervention (complete case analysis). CI, confidence interval; ***n*, number of patients with outcome; *N*, number of participants at risk; d.f., degrees of freedom; χ^2, chi-squared statistic; I^2, the percentage of total variation across studies that is due to heterogeneity rather than chance. Relative risks are plotted (black squares with area proportional to the amount of statistical information in each trial) comparing outcome among participants allocated to D-penicillamine with those allocated to placebo/no intervention, alongside with their 95% CI (horizontal lines). For particular subtotals, the result and its 95% CI are represented by a diamond, with the RR (95% CI) and its statistical significance given alongside. Squares or diamonds to the left of the solid vertical line indicate benefit with D-penicillamine, but this is conventionally significant ($P < 0.05$) only if the horizontal line or diamond does not overlap the solid vertical line.

Table 3. Relative risks of mortality in primary biliary cirrhosis patients randomized to D-penicillamine vs. placebo/no intervention – subgroup analyses

Subgroups	Number of trials	RR	95% CI	Test of interaction test
Methodological quality of trials				
Low-bias risk*	2[22, 23]	1.27	0.94–1.72	$P = 0.04$
High-bias risk†	4[8, 9, 21, 24]	1.40	1.06–1.84	
Dosage of D-penicillamine (g/day)				
1.2	1[24]	1.70	1.19–2.41	$P = 0.04$ between 0.6 and 1 g/day;
1	4[9, 21–23]	1.38	1.04–1.84	$P = 0.005$ between 0.6 and 1.2 g/day;
0.6	1[8]	0.68	0.40–1.17	$P = 0.37$ between 1 and 1.2 g/day
Trial duration (years)				
>3	3[8, 23, 24]	1.15	0.93–1.43	$P = 0.003$
≤3	3[9, 21, 22]	3.37	1.70–6.66	
Histological stage				
I/II	1[8]	Not estimable	Not estimable	Not estimable
III/IV	2[8, 23]	0.85	0.63–1.15	

* Low-bias risk – trials that meet two of the three criteria: adequate generation of allocation sequence, allocation concealment and blinding.
† High-bias risk – trials that did not meet two of the three criteria: adequate generation of allocation sequence, allocation concealment and blinding.

DISCUSSION

We found that D-penicillamine did not appear to reduce the risk of mortality and morbidity in patients with primary biliary cirrhosis. The use of D-penicillamine led to significantly more adverse events.

Our systematic review analysed seven trials including 706 patients. This is a low number of patients.[28] None of the trials reported sample size estimation. Dropouts were more often seen in the D-penicillamine group. The trials' methodological quality was generally low. In general, low methodological quality trials

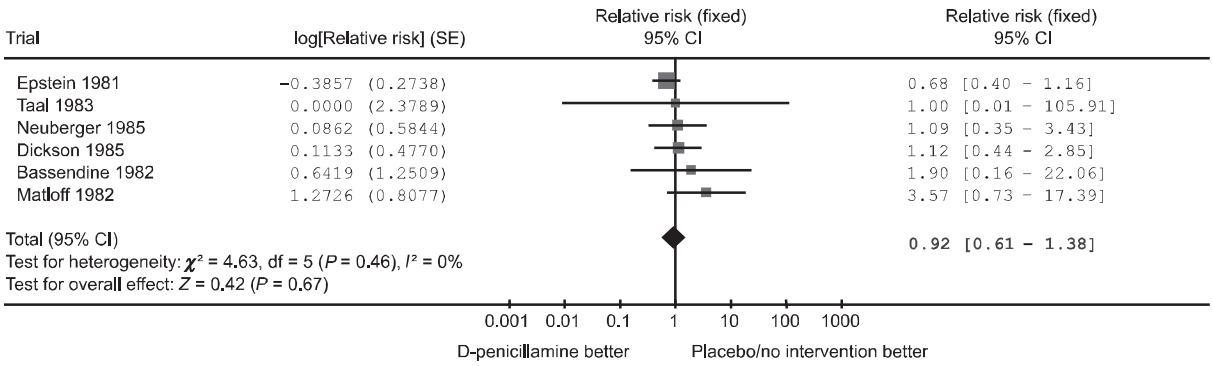

Figure 3. Relative risk (RR) of mortality in primary biliary cirrhosis patients randomized to D-penicillamine vs. placebo/no intervention (uncertainty interval method). CI, confidence interval; ***n*, number of patients with outcome; *N*, number of participants at risk; d.f., degrees of freedom; χ^2, chi-squared statistic; I^2, the percentage of total variation across studies that is due to heterogeneity rather than chance. Relative risks are plotted (black squares with area proportional to the amount of statistical information in each trial) comparing outcome among participants allocated to D-penicillamine with those allocated to placebo/no intervention, alongside with their 95% CI (horizontal lines). For particular subtotals, the result and its** 95% CI are represented by a diamond, with the RR (95% CI) and its statistical significance given alongside. Squares or diamonds to the left of the solid vertical line indicate benefit with D-penicillamine, but this is conventionally significant **($P < 0.05$) only if the horizontal line or diamond does not overlap the solid vertical line.**

Table 4. Adverse events in primary biliary cirrhosis patients randomized to D-penicillamine vs. placebo/no intervention

Trials	D-Penicillamine	Placebo/no intervention
Bassendine *et al.*[21]	Proteinuria, rash, 'lupus' syndrome, myasthenia and thrombocytopenia	None reported
Dickson *et al.*[23]	Hypersensitivity, cytopenia, arthralgias, linchen planus, loss of taste and proteinuria	Cytopenia, arthralgias, linchen planus, dysgeusia and proteinuria
Epstein *et al.*[8]	Rashes, proteinuria and neutropenia	None reported
Matloff *et al.*[22]	Good pasture-like syndrome, myasthenia, proteinuria, linchen planus, arthralgias, splenomegaly, rash, loss of taste and stomatitis	Proteinuria
Neuberger *et al.*[24]	Rash, proteinuria, thrombocytopenia, arthralgia, gastrointestinal upset, leucopoenia, asthma, pemphigoid, loss of taste, psychosis, palpitations and non-compliance	Proteinuria, gastrointestinal upset, headaches, non-compliance and neurological complications
Taal *et al.*[9]	Exanthema, gastrointestinal upset and loss of taste	Exanthema and gastrointestinal upset

significantly overestimate intervention effects.[12–14] If the overestimation is valid for the six trials, the prospects for D-penicillamine for primary biliary cirrhosis may be worse than observed. However, from the subgroup analysis of dosage, we cannot preclude that low-dose D-penicillamine may have benefits. In addition, most of the trials have shorter follow-up than the estimated median survival of primary biliary cirrhosis, i.e. 10–15 years.[29] Therefore, it is difficult to detect a significant difference on mortality. It seems that trials with a duration shorter than 3 years showed D-penicillamine may increase the risk of mortality (RR 3.37, 95% CI: 1.70–6.66). But this indication was suggested by subgroup analysis, which is hypothesis generating in nature. The result could possibly be confounded by other factors. Therefore, the relationship between trial's duration and effects of D-penicillamine (if any) needs to be investigated further.

We found that D-penicillamine did not reduce the risk of mortality. The pooled estimate from high-quality trials supports this finding. The estimate also holds, after increasing uncertainty, to allow for informative missing data due to dropouts. The uncertainty method is based on best–worst case analysis,[11] which

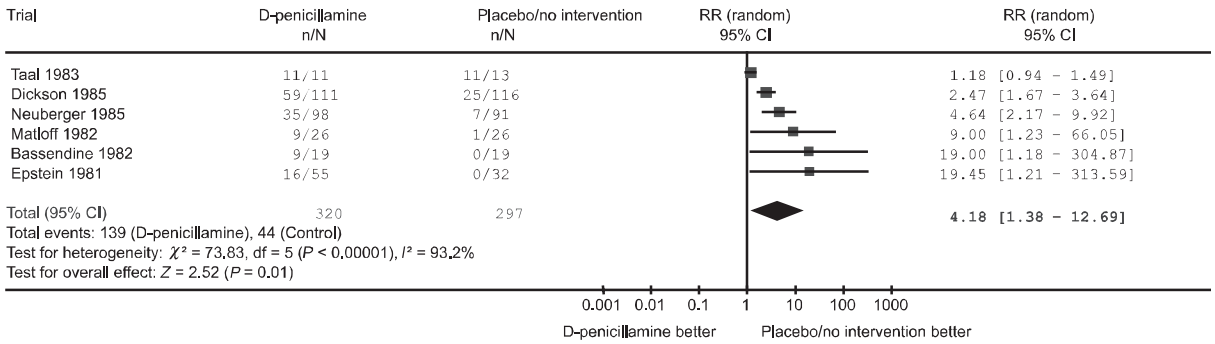

Figure 4. Relative risk (RR) of adverse events in primary biliary cirrhosis patients randomized to D-penicillamine vs. placebo/no intervention. CI, confidence interval; ***n*, number of patients with outcome; *N*, number of participants at risk; d.f., degrees of freedom; χ^2, chi-squared statistic; I^2, the percentage of total variation across studies that is due to heterogeneity rather than chance. Relative risks are plotted (black squares with area proportional to the amount of statistical information in each trial) comparing outcome among participants allocated to D-penicillamine with those allocated to placebo/no intervention, alongside with their 95% CI (horizontal lines). For particular subtotals, the result and its 95% CI are represented by a diamond, with the RR (95% CI) and its statistical significance given alongside. Squares or diamonds to the left of the solid vertical line indicate benefit with D-penicillamine, but this is conventionally significant ($P < 0.05$) only if the horizontal line or diamond does not overlap the solid vertical line.

is one way to handle missing data in meta-analysis. Compared with best–worst case analysis, the uncertainty method has similar ability to capture the true effect, but with narrower intervals and hence better power.[16]

The subgroup analysis showed that the risk of mortality seemed to increase with dose of D-penicillamine. This observation, however, was not supported by the Bassendine *et al.*'s trial, where the patients taking high-dose D-penicillamine had a lower risk of mortality than those taking the low dose.[21] As the ideal way to study causes of true variation is within trials rather than among trials, and the purpose and nature of this meta-analysis was not to study the dose–response relationship, the relationship between the effect of D-penicillamine and dosage is not clear.

It is presumed that patients with advanced disease will have more to gain from an intervention and may therefore experience sufficient benefits to outweigh the harms. However, due to the small number of trials included, we were not able to identify an association between severity of primary biliary cirrhosis (i.e. histological stage) and the effects of D-penicillamine.

Neuberger *et al.* reported extractable data on clinical findings.[24] They revealed no significant differences regarding pruritus, gastrointestinal bleeding, or ascites between D-penicillamine and placebo groups. The other trials claimed that no consistent clinical improvement in either the D-penicillamine or placebo group had been found.[22, 23]

The rate of liver histological progression favoured neither D-penicillamine nor placebo. One trial identified that D-penicillamine reduced histological inflammatory activity.[8] However, the effect is only marginally significant and based on only one trial with a small sample of patients.[8]

The report by Matloff *et al.* reported extractable data on liver biochemical variables, which resulted in no significant differences except D-penicillamine significantly decreasing alanine aminotransferase activity.[22] This finding was replicated in the Neuberger *et al.*'s trial, in which alanine aminotransferase was the only liver biochemical outcome improved by D-penicillamine.[24] Epstein *et al.* and Bassendine *et al.* found the benefit of D-penicillamine in reducing the levels of aspartate aminotransferase and immunoglobulin.[8, 21] Taal *et al.* found that D-penicillamine decreased immunoglobulin M and G levels.[9] The fact that improvements in different biochemical variables were observed in different trials weakened the conclusion of benefit of D-penicillamine on liver biochemical variables, in general.

The adverse events in the D-penicillamine group were, on average, four times more than the placebo/no intervention group. Most of the adverse events were proteinuria, gastrointestinal upset, rash, cytopenia, etc.

We were unable to distinguish between serious and non-serious adverse events due to the insufficient reporting of the trials. For meta-analyses employing RR as effect measurement, the proportional weights increase with increasing event rates, given to trials estimating the same effect with the same sample size. The relationship becomes particularly strong when the event rates are above 50%.[30] In this respect, we scrutinized the event rates in the included trials and noticed that the trial by Taal *et al.* had the smallest sample size (24 patients) but with the highest placebo response rate of 85% (11 of 13).[9] Hence, we performed a sensitivity analysis excluding the Taal *et al.*'s trial[9] and it resulted in the RR of 3.69 (95% CI: 2.62–5.19) with moderate heterogeneity ($I^2 = 49.7\%$). Therefore, our conclusion, that the use of D-penicillamine was associated with significant increase of adverse events, was consolidated.

D-Penicillamine did not appear to reduce the risk of mortality or morbidity and led to significantly more adverse events in patients with primary biliary cirrhosis. Hence, we do not advocate using D-penicillamine for patients with primary biliary cirrhosis. We do not recommend further randomized-clinical trials aiming at establishing the value of D-penicillamine in the treatment of primary biliary cirrhosis, at least not with the dosages employed in the included trials. The possibility that low doses may offer beneficial effects cannot be excluded. Future investigators should report their trials according to the CONSORT Statement (http://www.consort-statement.org).

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This systematic review was carried out using the recommendations of The Cochrane Collaboration and The Cochrane Hepato-Biliary Group. This review was published as a Cochrane review in The Cochrane Library. Cochrane Reviews are regularly updated as new evidence emerges and in response to comments and criticisms. The Cochrane Library should be consulted for the most recent version of the review.

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Appendix 3A

Colchicine for primary biliary cirrhosis (Review)

Gong Y, Gluud C

This is a reprint of a Cochrane review, prepared and maintained by The Cochrane Collaboration and published in *The Cochrane Library* 2007, Issue 1

http://www.thecochranelibrary.com

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Colchicine for primary biliary cirrhosis (Review)

Gong Y, Gluud C

Status: *Commented*

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ABSTRACT

Background
Colchicine has been used for patients with primary biliary cirrhosis because of its immunomodulatory and antifibrotic potential. The therapeutical responses to colchicine in randomised clinical trials were inconsistent.

Objectives
To evaluate the beneficial and harmful effects of colchicine in patients with primary biliary cirrhosis.

Search strategy
We identified trials through electronic searches of *The Cochrane Hepato-Biliary Group Controlled Trials Register*, *The Cochrane Central Register of Controlled Trials* on *The Cochrane Library*, *MEDLINE*, *EMBASE (September 2003)*, and manual searches of bibliographies. We contacted authors of trials and pharmaceutical companies.

Selection criteria
Randomised clinical trials comparing colchicine with any kind of control therapy were included irrespective of language, year of publication, and publication status.

Data collection and analysis
The primary outcomes were the number of deaths and the number of death and/or patients who underwent liver transplantation. Dichotomous outcomes were reported as relative risk (RR) with 95% confidence interval (CI). We examined intervention effects by using both a fixed effect model and a random effects model. Heterogeneity was investigated by subgroup analyses and sensitivity analyses.

Main results
Eleven randomised clinical trials involving 716 patients with primary biliary cirrhosis fulfilled the inclusion criteria. No significant differences were detected between colchicine and placebo/no intervention on the number of deaths (RR 1.21, 95% CI 0.71 to 2.06), the number of deaths and/or patients who underwent liver transplantation (RR 1.00, 95% CI 0.67 to 1.49), liver complications, liver biochemical variables, liver histological measurements, and adverse events. Trial methodology was generally low and some trials had high drop-out rate. A best-worst-case-scenario analysis showed no significant effect of colchicine on mortality (RR 0.59, 95%CI 0.30 to 1.15), while a worst-best-case-scenario analysis showed a significant detrimental effect of colchicine on mortality (RR 2.28, 95% CI 1.17 to 4.44). Colchicine significantly decreased the number of patients without improvement of pruritus (RR 0.75, 95% CI 0.65 to 0.87). However, this estimate was based on only 156 patients from three trials. The effect of the combined treatment with ursodeoxycholic acid was not significantly different from that of colchicine alone.

Authors' conclusions
We did not find evidence either to support or refute the use of colchicine for patients with primary biliary cirrhosis. As we are not able to exclude a detrimental effect of colchicine, we suggest that it is only used in randomised clinical trials.

PLAIN LANGUAGE SUMMARY

No convincing evidence either to support or refute the use of colchicine for patients with primary biliary cirrhosis

Primary biliary cirrhosis is a rare, chronic liver disease of unknown etiology. Colchicine, a plant alkaloid, has been used to treat patients with primary biliary cirrhosis and was tested in randomised clinical trials. When all identified trials were combined, colchicine appeared to be not significantly different from placebo/no intervention in respect to mortality, mortality and/or patients who underwent liver transplantation, liver complications, liver biochemistry, liver histology, and the occurrences of adverse events. Colchicine may reduce pruritus, but this finding may be due to bias. The addition of ursodeoxycholic acid did not significantly influence the effect of colchicine.

BACKGROUND

Primary biliary cirrhosis is an uncommon chronic progressive liver disease of unknown etiology. Ninety per cent of patients with primary biliary cirrhosis are females and the majority are diagnosed after the age of 40 years (James 1981). The earlier description was published in 1949 (MacMahon 1949). Later, Ahrens and coworkers comprehensively described primary biliary cirrhosis in 1950 (Ahrens 1950). A progressive granulomatous hepatitis destroys small septal and interlobular bile ducts, eventually leading to cholestasis and biliary cirrhosis. Primary biliary cirrhosis is classically defined on the basis of the triad: antimitochondrial antibodies, which are found in over 95% of patients with primary biliary cirrhosis (Fregeau 1989; Lacerda 1995; Invernizzi 1997; Turchany 1997; Mattalia 1998); abnormal liver function tests that are typically cholestatic (with raised activity of alkaline phosphatases being the most frequently seen abnormality); and characteristic liver histological changes (Scheuer 1967) in the absence of extrahepatic biliary obstruction (Kaplan 1996). Patients may either be diagnosed during a symptomatic phase (the common symptoms being pruritus, fatigue, jaundice, liver enlargement, signs of portal hypertension, sicca complex, and scleroderma-like lesions), in which case survival is significantly decreased, or during an asymptomatic phase of the disease, which has a relatively favourable prognosis (Beswick 1985; Balasubramaniam 1990). However, 40 to 100% of these patients will subsequently develop symptoms of primary biliary cirrhosis (Nyberg 1989; Metcalf 1996; Prince 2000).

Although the etiology remains unknown, primary biliary cirrhosis is in many respects analogous to the graft-versus-host syndrome in which the immune system is sensitised to foreign proteins. Most primary biliary cirrhosis patients have increased class II human leukocyte antigen (HLA) histocompatibility expression on bile duct cells (Ballardini 1984; Van den Oord 1986), and the bile duct epithelium is infiltrated by cytotoxic T-cells (Yamada 1986). Lacrimal and pancreatic glands, for example, with a high concentration of HLA class II antigens on their epithelium, may be involved in the disease process (Epstein 1982).

Patients with primary biliary cirrhosis have been subjected to many drugs. Ursodeoxycholic acid, a bile acid, is the most extensively used drug in these patients (Verma 1999). However, a recent systematic Cochrane Review was unable to demonstrate any significant effect of ursodeoxycholic acid on mortality or liver transplantation (Gluud 2002). Over the years, a number of other drugs have been evaluated for primary biliary cirrhosis. Earlier attempts to treat primary biliary cirrhosis using immunomodulatory and other agents such as azathioprine (Heathcote 1976; Christensen 1985), prednisolone (Mitchison 1992), chlorambucil (Hoofnagle 1986), cyclosporine (Wiesner 1990), D-penicillamine (Epstein 1981; Matloff 1982; Dickson 1985; Neuberger 1985), methotrexate (Kaplan 1991; Lindor 1995), or colchicine have resulted in clinical effects that have not led to widespread acceptance of these drugs for primary biliary cirrhosis patients (Kaplan 1994).

Colchicine is a plant alkaloid. It is effective against gouty arthritis and other forms of rheumatic diseases (rheumatoid arthritis, familial Mediterranean fever, Bechet's disease, etc.) (Ben-Chetrit 1998). The basis for effect of colchicine is inhibition of the migration of granulocytes into inflamed areas and decreased metabolic and phagocytic activity of granulocytes. Further, colchicine is an anti-mitotic (Shi 1998) and anti-fibrotic agent. Colchicine retards the microtubule mediated transport of procollagen (Ehrlich 1972) and enhances collagenase activity (Harris 1971).

Colchicine has been used for primary biliary cirrhosis patients because of its immunomodulatory and antifibrotic potential. Colchicine has been reported to slow the rate of progression of primary biliary cirrhosis (Kaplan 1997) and to produce improvements in liver function tests and immunoglobulin levels (Warnes 1987; Vuoristo 1995; Kaplan 1999). However, colchicine does not affect clinical symptoms or liver histology (Kaplan 1986). The effect of combination therapy with colchicine and ursodeoxycholic acid in patients with primary biliary cirrhosis has been reported, but the results have been conflicting (Shibata 1992; Ikeda 1996; Poupon 1996; Almasio 2000; Battezzati 2001). We have been unable to identify meta-analyses or systematic reviews on the beneficial and harmful effects of colchicine for primary biliary cirrhosis patients.

OBJECTIVES

The objectives were to assess the beneficial and harmful effects of colchicine for patients with primary biliary cirrhosis.

CRITERIA FOR CONSIDERING STUDIES FOR THIS REVIEW

Types of studies

We included randomised clinical trials irrespective of language, year of publication, and publication status. We excluded studies using quasi-randomisation (e.g., allocation by date of birth).

Types of participants

Patients with primary biliary cirrhosis, i.e., patients having at least two of the following: elevated serum activity of alkaline phosphatases (or other markers of intrahepatic cholestasis), and/or a positive result for serum mitochondrial antibody, and/or liver biopsy findings diagnostic for or compatible with primary biliary cirrhosis.

Types of intervention

Administration of any dose of colchicine versus placebo or no intervention or other drugs. Co-interventions were allowed as long as all intervention arms of the randomised clinical trial received similar co-interventions. Therefore, we analysed the following comparisons:
1) Colchicine versus placebo/no intervention (monotherapy).
2) Colchicine plus ursodeoxycholic acid versus placebo/no intervention plus ursodeoxycholic acid (combination therapy).
3) Colchicine versus other drugs.

Types of outcome measures

Primary outcome measures were:

- Number of deaths.
- Number of deaths and/or patients who underwent liver transplantation.

Secondary outcome measures were:

- Number of patients who underwent liver transplantation.
- Pruritus: number of patients without improvement of pruritus and/or pruritus score.
- Fatigue: number of patients without improvement of fatigue and/or fatigue score.
- Incidence of complications: number of patients developing variceal bleeding, ascites, hepatic encephalopathy, jaundice, or hepato-renal syndrome.
- Liver biochemistry: serum (s-)bilirubin; s-alkaline phosphatases; s-gamma-glutamyltransferase; s-aspartate aminotransferase; s-alanine aminotransferase; s-albumin; s-cholesterol (total); plasma immunoglobulin M.
- Liver biopsy findings: deterioration of liver histological stage or score.
- Quality of life: broad nature of a concept that includes physical functioning (ability to carry out activities of daily living such as self-care), psychological functioning (emotional and mental well-being), social functioning (relationships with others and participation in social activities), and perception of health, pain, and overall satisfaction with life.
- Adverse events. The adverse events are defined as any untoward medical occurrences in patients in either of the two arms of the included randomised clinical trials, which did not necessarily have a causal relationship with the treatment, but did, however, result in a dose reduction, discontinuation of treatment, or registration of the event as an adverse event/side effect. The adverse events are subdivided into non-serious and serious, according to the ICH-GCP guidelines (ICH-GCP 1997). A serious adverse event is any event that leads to death, is life-threatening, requires inpatient hospitalisation or prolongation of existing hospitalisation, results in persistent or significant disability or congenital anomaly/birth defect, or any important medical event, which may jeopardize the patient or requires intervention to prevent it.
- Health economics: the estimated costs connected with the interventions are weighed against any possible health gains.

SEARCH METHODS FOR IDENTIFICATION OF STUDIES

See: Cochrane Hepato-Biliary Group methods used in reviews.

Relevant randomised clinical trials were identified by searching *The Cochrane Hepato-Biliary Group Controlled Trials Register* (September 2003), *The Cochrane Central Register of Controlled Trials* on *The Cochrane Library* (Issue 3, 2003), *MEDLINE* (January 1966 to September 2003), and *EMBASE* (January 1980 to September 2003). See 'Table 01' for the search strategies that were applied to the individual electronic databases.

Further trials were identified by reading the reference lists of the identified studies. We wrote to the principal authors of the identified randomised clinical trials and to researchers active in the field to inquire about additional randomised clinical trials they might know of. We also wrote to the pharmaceutical companies that sponsored colchicine in identified trials to obtain unidentified or unpublished randomised clinical trials.

METHODS OF THE REVIEW

The review was performed following the published protocol (Gong 2003) and the recommendations given by the Cochrane Reviewers' Handbook (Clarke 2003).

Trials selection

Identified trials were listed and two contributors (YG and CG) independently evaluated whether the trials fulfilled the inclusion criteria. Excluded trials were listed with the reasons for exclusion. Disagreements were resolved by discussion.

Data extraction

YG extracted the data and CG validated the data extraction. Disagreements were solved by discusiion. YG wrote to the authors of all the included trials on colchicine for primary biliary cirrhosis and asked them to specify data, had they not been reported clearly in the articles.

Assessment of methodological quality of included trials

The methodological quality of the randomised clinical trials was assessed using four components (Schulz 1995; Moher 1998; Kjaergard 2001):

Generation of the allocation sequence

- Adequate, if the allocation sequence was generated by a computer or random number table. Drawing of lots, tossing of a coin, shuffling of cards, or throwing dice will be considered as adequate if a person who was not otherwise involved in the recruitment of participants performed the procedure;
- Unclear, if the trial was described as randomised, but the method used for the allocation sequence generation was not described;
- Inadequate, if a system involving dates, names, or admittance numbers were used for the allocation of patients. These studies are known as quasi-randomised and were excluded from the present review.

Allocation concealment

- Adequate, if the allocation of patients involved a central independent unit, on-site locked computer, identically appearing numbered drug bottles or containers prepared by an independent pharmacist or investigator, or sealed envelopes;
- Unclear, if the trial was described as randomised, but the method used to conceal the allocation was not described;
- Inadequate, if the allocation sequence was known to the investigators who assigned participants or if the study was quasi-randomised. Such studies were excluded from the present review.

Blinding (or masking)

- Adequate, if the trial was described as double blind and the method of blinding involved identical placebo or active drug;
- Unclear, if the trial was described as double blind, but the method of blinding was not described;
- Not performed, if the trial was not double blind.

Follow-up

- Adequate, if the numbers and reasons for dropouts and withdrawals in all intervention groups were described or if it was specified that there were no dropouts or withdrawals;
- Unclear, if the report gave the impression that there had been no dropouts or withdrawals, but this was not specifically stated;
- Inadequate, if the number or reasons for dropouts and withdrawals were not described.

Characteristics of patients

Number of patients randomised; patient inclusion and exclusion criteria; mean (or median) age; sex ratio; number of patients lost to follow-up; drop-outs; withdrawals.

Characteristics of interventions

Type, dose, and form of colchicine intervention; type of intervention in the control group and collateral interventions; duration of treatment, length of follow-up.

Characteristics of outcomes

All outcomes were extracted from each included trial when possible.

We analysed mortality and/or liver transplantation at maximum follow-up. We analysed other outcomes, which were repeatedly observed on patients (like liver biochemistry, clinical symptoms, etc.) at maximum follow-up. However, where possible, we also extracted data on primary outcome measures from the maximal follow-up in each randomised clinical trial, and if available, including data from after the patients were switched from blinded to open label therapy.

Statistical methods

We used RevMan Analyses 1.0.1 provided by The Cochrane Collaboration. Dichotomous data were presented as relative risk (RR) with 95% confidence interval (CI), and continuous outcomes were presented by weighted mean differences (WMD) with 95% CI. The analyses for the primary outcomes were performed according to the intention-to-treat analyses, which means that the participants in the trials were to be analysed in the groups to which they were randomised, regardless of whether they received or adhered to the allocated intervention. We computed a 'reported scenario' analysis. However, we placed most weight on the 'likely scenario' analysis (see Subgroup analyses and sensitivity analyses below).

We examined intervention effects by using both a fixed effect model (DeMets 1987) and a random effects model (DerSimonian 1986) with the level of significance set at $P \leq 0.05$. If the results of the two analyses led to the same conclusion, only the result of the fixed effect model analysis was given in the text. In case of significant discrepancies of the two models, results from both models were reported and discussed. The presence of statistical heterogeneity was explored by the chi-squared test with significance set at $P \leq 0.10$ and measured the quantities of heterogeneity by I^2.

Subgroup analyses and sensitivity analyses

We performed subgroup analyses, in which trials were grouped according to the stage of disease; duration of treatment; adequacy of generation of the allocation sequence; allocation concealment; blinding; and whether the trial reported used intention-to-treat analysis. The cut-off for duration of treatment was determined by comparing the intervention effect of the group of trials lasting for no more than the median treatment duration with that of the group of trials lasting for more than the median duration. The differences between subgroups were estimated according to Altman 2003.

Regarding the binary outcomes, patients with incomplete or missing data were included in sensitivity analyses by imputing them:

- Likely scenario: worst-case scenario for both colchicine and control.
- Best-case scenario: best-case scenario for colchicine and worst-case scenario for control.
- Reported scenario: best-case scenario for both colchicine and control.
- Worst-case scenario: worst-case scenario for colchicine and best-case scenario for control.

For secondary outcomes we adopted 'available case analysis', i.e., include data on only those patients, whose results are known, using for denominator the total number of patients who completed the trial for the particular outcome in question. Thus, in the review, the number of patients as the denominator might change according to the secondary outcomes investigated.

Bias detection

Funnel plot was used to provide a visual assessment of whether treatment estimates are associated with study size. The performance of the available methods of detecting publication bias and other biases (Begg 1994; Egger 1997; Macaskill 2001) vary with the magnitude of the treatment effect, the distribution of study size, and whether a one- or two-tailed test is used (Macaskill 2001). Therefore, we decided to use the most appropriate method having good trade-off in the sensitivity and specificity, based on characteristics of the trials to be included in this review.

DESCRIPTION OF STUDIES

Search results

We identified a total of 559 references through electronic searches of *The Cochrane Hepato-Biliary Group Controlled Trials Register* (n = 29), *The Cochrane Controlled Trials Register* on *The Cochrane Library* (n = 212), *MEDLINE* (n = 148), and *EMBASE* (n = 170). We excluded 465 duplicates and clearly irrelevant references through reading abstracts. Accordingly, 95 references were retrieved for further assessment. Of these, we excluded 57 because they were reviews, meta-analyses, or observational studies. Among the 57 references, the three observational studies were listed under 'Characteristic of excluded studies' with reasons for exclusion. The remaining 38 references referred to 11 randomised trials, which fulfilled our inclusion criteria of this review.

Two of the 11 randomised clinical trials were published as abstracts only (Goddard 1995; Warnes 1996). One trial (Raedsch 1993) was published in symposia proceedings. The remaining eight randomised clinical trials were published in peer-reviewed journals.

Included studies

All the included trials reported random allocation of 716 patients with primary biliary cirrhosis to:

- colchicine versus placebo (Kaplan 1986; Warnes 1987; Bodenheimer 1988; Goddard 1995; Vuoristo 1995; Warnes 1996)
- colchicine plus ursodeoxycholic acid versus placebo/no intervention plus ursodeoxycholic acid (Raedsch 1993; Goddard 1995; Ikeda 1996; Poupon 1996; Almasio 2000)
- colchicine versus ursodeoxycholic acid (Goddard 1995; Vuoristo 1995)
- colchicine versus methotrexate (Kaplan 1999).

Vuoristo 1995 had three intervention arms: colchicine versus ursodeoxycholic acid versus placebo, and Goddard 1995 had four intervention arms: colchicine, ursodeoxycholic acid, colchicine plus ursodeoxycholic acid, and placebo. We were not able to extract data from Goddard 1995 and Warnes 1996 for our meta-analyses because they were published only as abstracts and correspondence with the authors did not lead to additional information. Accordingly, data from nine trials involving 599 patients with primary biliary cirrhosis were pooled in our meta-analyses.

The entry criteria varied across trials, but were generally well-defined, making it highly likely that all or almost all patients did in fact have primary biliary cirrhosis. The dosage of colchicine varied slightly, from 1 mg daily (n = 7) to 1.2 mg daily (n = 3). Only Warnes 1996 did not report the dosage. The duration of colchicine treatment varied from one to two years. Following the stipulated follow-up, two trials (Kaplan 1986; Bodenheimer 1988) continued colchicine-patients on open label colchicine (colchicine → colchicine) and offered open label colchicine to the patients originally receiving placebo (placebo → colchicine). One subsample of Poupon 1996 trial, which was published as an abstract, continued patients in both groups with open label colchicine plus ursodeoxycholic acid. However, we were not able to retrieve additional data after our correspondence with the principal author.

METHODOLOGICAL QUALITY

The methods to generate the allocation sequence were considered adequate in four randomised clinical trials (Kaplan 1986; Warnes 1987; Vuoristo 1995; Almasio 2000) and unclear or inadequate in the remaining seven. The methods to conceal allocation were considered adequate in six (Kaplan 1986; Warnes 1987; Vuoristo 1995; Ikeda 1996; Kaplan 1999; Almasio 2000) and unclear or inadequate in the remaining five. Blinding was adequate in seven trials, unclear in three (Raedsch 1993; Goddard 1995; Warnes 1996) and not performed in Ikeda 1996 trial. The Kaplan 1999 trial, which compared colchicine to methotrexate, employed the double dummy technique to maintain the double-blinding. The description of the placebo was, however, not sufficient - i.e., some of the trials employing placebo only stated that the placebo tablets were identical in appearance or indistinguishable, but did not mention smell and taste. The other randomised clinical trials (Raedsch 1993; Vuoristo 1995; Warnes 1996) did not give any description of the placebo used. There was generally a fair description of follow-up and withdrawal/drop-out, in which eight trials were regarded as adequate and three inadequate. Two trials had high rates of loss of follow-up and withdrawals/drop-outs, 22.8% in Bodenheimer 1988 trial and 15.6% in Warnes 1987 trial.

To note, only two out of the 11 randomised clinical trials (Kaplan 1986; Kaplan 1999) provided pre-trial sample size estimation based on the rates of success defined by the authors. None of the trials used mortality to calculate sample size estimation.

RESULTS

Colchicine versus placebo/no intervention (monotherapy or combination therapy)

Number of deaths

Seven randomised clinical trials involving 398 patients reported data on number of deaths. In the colchicine group 25/200 (12.5%) patients died versus 20/198 (10.1%) patients in the control group. Combining the results of individual trials demonstrated no significant difference in the number of deaths (RR 1.21, 95% CI 0.71 to 2.06) (Comparison 01-01).

We performed sensitivity analyses regarding the number of deaths (Comparison 06-01). Neither the reported-scenario nor the likely-scenario analyses showed any significant difference between colchicine and placebo/no intervention. The best-worst-case-scenario analysis did not show any significant difference either. The worst-best-case-scenario analysis detected a significant detrimental effect of colchicine on mortality.

Including data from the extended follow-up during treatment with colchicine → colchicine versus placebo → colchicine into the analyses demonstrated a RR of 1.15 (95% CI 0.76 to 1.73) (Comparison 02-01).

Number of deaths and/or patients who underwent liver transplantation

Eight randomised clinical trials involving 455 patients reported data on 'number of deaths and/or patients who underwent liver transplantation'. We detected 36/228 (15.8%) deaths and patients who underwent liver transplantation in the colchicine group versus 36/227 (15.9%) in the control group. Combining the results of the eight trials demonstrated no significant difference in this outcome measure (RR 1.00, 95% CI 0.67 to 1.49) (Comparison 01-02).

We performed sensitivity analyses. Neither the reported-scenario nor the likely-scenario analyses showed any significant difference between colchicine and placebo/no intervention. The best-worst-case-scenario analysis showed a significant effect favouring colchicine, while the worst-best-case-scenario analysis showed a significant effect favouring placebo/no intervention.

Including data from the extended follow-up during treatment with colchicine → colchicine versus placebo → colchicine demonstrated a RR of 1.02 (95% CI 0.72 to 1.46) (Comparison 02-02).

Subgroup analyses

The subgroup analyses, taking the dose and duration of colchicine into consideration, did not reveal differing results (Comparison 05-01,05-02). The trials where colchicine was administered with 1 mg/day (RR 1.36, 95% CI 0.73 to 2.52) versus 1.2 mg/day (RR 0.83, 95% CI 0.28 to 2.44) did not reveal any significant influence on the relative risk of mortality. Test of interaction between the two estimates showed no significant difference (P = 0.44). The trials where colchicine was administered for no longer than two years (RR 1.04, 95% CI 0.56 to 1.93) did not differ significantly from the trials where colchicine was administered for longer than two years (RR 1.82, 95% CI 0.62 to 5.39). Test of interaction between the two groups detected no significant difference (P = 0.38).

Subgroup analyses on mortality stratifying the seven trials according to their methodological quality were performed. The adequacy of generation of the allocation sequence, allocation concealment, and blinding did not change this estimate significantly (P = 0.15, 0.15, and 0.26, respectively) (Comparison 05-03 to 05-05). Follow-up was adequate in all the trials, which provided mortality data.

Subgroup analyses stratifying the trials according to monotherapy or combined treatment, i.e., colchicine plus ursodeoxycholic acid, did not change this estimate (Comparison 01-01):

- colchicine versus placebo (RR 0.92, 95% CI 0.48 to 1.75);
- colchicine plus ursodeoxycholic acid versus placebo/no intervention plus ursodeoxycholic acid (RR 2.14, 95% CI 0.78 to 5.87).

Test of interaction between the two groups showed no significant difference (P = 0.17).

Similar findings applied to the risks of mortality or liver transplantation (Comparison 01-02, 05-06 to 05-10).

Pruritus and fatigue

Pooling the data from three trials demonstrated that colchicine significantly decreased the number of patients without improvement of pruritus (RR 0.75, 95% CI 0.65 to 0.87). One trial reported the data of the 'number of patients without improvement of fatigue', and it was not significantly different in the colchicine group and the control group (RR 0.86, 95% CI 0.72 to 1.02).

Liver complications

Overall, no significant difference was detected on liver complications between colchicine and control group (RR 0.37, 95% CI 0.12 to 1.10). Neither the number of patients with development of varices (RR 0.31, 95% CI 0.08 to 1.19), gastrointestinal bleeding (RR 0.50, 95% CI 0.05 to 5.28), nor the number of patients developing hepatic encephalopathy (RR 1.07; 95% CI 0.07 to 16.31) were significantly affected by colchicine. We were not able to extract data on jaundice.

Biochemical variables

Colchicine did not lead to any significant effect on the following biochemical variables (Comparisons 01-07 to 01-15):

- s-bilirubin: WMD (arithmetic mean) -1.35 µmol/L, 95% CI -4.52 to 1.82; WMD (geometric mean) -1.55 µmol/L, 95% CI -2.72 to 1.13;
- s-alkaline phosphatases: WMD (arithmetic mean) -55.35 international units (IU)/L, 95 CI -158.56 to 47.85; WMD (geometric mean) -1.26 IU/L, 95 CI -1.80 to 1.14;
- s-gamma-glutamyltransferase: WMD -25.38 IU/L, 95% CI -73.26 to 22.50;
- s-aspartate aminotransferase: WMD -10.10 IU/L, 95% CI -22.91 to 2.71;
- s-alanine aminotransferase: WMD -2.05 IU/L, 95% CI -8.79 to 4.68;
- s-albumin: WMD 0.09 g/dL, 95% CI -0.03 to 0.21;
- s-total cholesterol: WMD (arithmetic mean) 0.10 mmol/L, 95% CI -0.88 to 1.08; WMD (geometric mean) -1.02 mmol/L, 95% CI -1.20 to 1.15;
- plasma immunoglobulin M: WMD -0.49 g/L, 95% CI -1.03 to 0.06;
- prothrombin time: WMD -0.03 seconds, 95% CI -0.75 to 0.69.

The Kaplan 1986 trial reported bilirubin, cholesterol, and alkaline phosphatases using geometric mean (Comparison 05-13, 05-14, 05-15), and we reported them as log transformed geometric mean for the sake of comparison.

Liver histology

There was no significant influence of colchicine on the number of patients experiencing worsening of histological stage (RR 0.85, 95% CI 0.41 to 1.75), fibrosis (RR 0.60, 95% CI 0.24 to 1.49), piecemeal necrosis (RR 0.58, 95% CI 0.23 to 1.44), parenchymal inflammation (RR 0.69, 95% CI 0.28 to 1.72), or parenchymal necrosis (RR 1.00, 95% CI 0.31 to 3.18). In addition, the Warnes 1987 trial reported no significant effect of colchicine on the number of patients who underwent worsening of cholestasis and granulomas. Poupon 1996 demonstrated no significant effects of colchicine on the number of patients with worsening of ductular proliferation and cholangitis. However, Poupon 1996 observed a significant lower incidence of patients with worsening of lobular inflammation in the colchicine group (RR 0.16, 95% CI 0.03 to 0.80) (Comparison 01-17). No significant effects on histological score were observed (WMD 0.56, 95% CI -0.24 to 1.36) in colchicine patients when compared to the patients in the control group (Comparison 01-18).

Quality of life

None of the trials examined specific quality-of-life scales or health economics.

Adverse events

In the colchicine group, 39/228 (17.1%) patients had adverse events (mostly transient diarrhoea) versus 26/227 (11.5%) patients in the control group (Comparison 01-19). This was not significantly different (RR 1.45, 95% CI 0.94 to 2.25). Also, no significantly different occurrences of serious adverse events were observed (RR 1.17, 95% CI = 0.50 to 2.75) (Comparison 01- 20).

Colchicine versus ursodeoxycholic acid

Vuoristo 1995 compared colchicine versus ursodeoxycholic acid. They observed that 5/29 patients died in the colchicine group versus 0/30 patients in the ursodeoxycholic acid group (RR 11.37, 95% CI 0.66 to 196.74) and no one underwent liver transplantation. The number of patients without improvement of fatigue was significantly less in the colchicine group than in the ursodeoxycholic acid group (RR 0.83, 95% CI 0.70 to 0.98). Regarding liver biochemical outcomes, only the levels of s-alkaline phosphatases and gamma-glutamyltransferase were significantly higher in the colchicine group than in the ursodeoxycholic acid group (WMD 378.00 IU/L, 95% CI 116.91 to 639.09; WMD 459.00 IU/L, 95% CI 157.57 to 760.43, respectively). For other outcomes (i.e., number of patients without improvement of pruritus, number of patients developing liver complications, number of patients with adverse events), no significant differences were detected.

Colchicine versus methotrexate

Kaplan 1999 compared colchicine versus methotrexate. This study observed that 9/43 patients died or underwent liver transplantation in the colchicine group versus 11/42 patients in the methotrexate group (RR 0.80, 95% CI 0.37 to 1.73). The pruritus score was significantly higher in patients receiving colchicine than methotrexate (WMD 0.68, 95% CI 0.25 to 1.11). Regarding liver biochemical outcomes, only the levels of s-alkaline phos-

phatases and plasma immunoglobulin M were significantly higher in the colchicine group than in the methotrexate group (WMD 0.41, 95% CI 0.12 to 0.70; WMD 0.47, 95% CI 0.20 to 0.74), respectively. For other outcomes (i.e., fatigue score, liver histology, number of patients with adverse events) no significant differences were detected.

Bias detectionn

We did not perform funnel plot analysis and did not apply the three statistical methods to detect publication bias and other biases because the power of those would have been low and inconsistent because of the small number of included trials.

DISCUSSION

We found no significant difference on mortality or mortality and liver transplantation between colchicine and placebo/no intervention for patients with primary biliary cirrhosis. These observations were robust to subgroup analyses taking methodological quality of trials, dose, and treatment duration into consideration. It has been reported that trials with inadequate methodological quality do significantly overestimate the effect of interventions (Schulz 1995; Moher 1998; Kjaergard 2001). However, we found that our results are not sensitive to the adequacy of generation of the allocation sequence, allocation concealment, blinding status, and the use of intention-to-treat analysis, probably due to the relatively small sample size and low number of trials included.

Our systematic review may have a number of limitations. Firstly, our systematic review regarding the comparison of colchicine versus placebo/no intervention on mortality analysed only seven trials involving 398 patients. This is a low number of patients (Ioannidis 2001). Additionally, compared to the natural history of primary biliary cirrhosis, most of the trials had relatively short period of medication and follow-up. Thus, the risk of type 2 error (the risk of overlooking an effect if it really exists) is present and a potential beneficial effect of colchicine on survival cannot be reliably excluded. Secondly, the present meta-analyses on mortality and mortality or liver transplantation were based on number of events per randomised patients from the individual trial, not on individual patient data analysis based on time-to-event data. An individual patient data analysis takes time and censored data into consideration and may offer potential advantages. However, the use of meta-analysis based on aggregate data extracted from published and unpublished reports can be considered a useful approach and seems to reach similar conclusions (Liberati 1996). Thirdly, since we could not stratify summary data of included trials according to the patients' baseline stage of primary biliary cirrhosis, we do not know whether the effect of colchicine was associated with the severity of primary biliary cirrhosis. Fourthly, we performed a high number of statistical tests, which increases the risk of 'mass significance' (i.e., spurious significant findings due to repetitive testing). Therefore, significant findings ought to be conservatively interpreted. Fifthly, although we employed considerable search strategies and applied no publication status or language limitations, we are concerned about the existence of publication bias and other biases, which leads us to identify 'positive' studies more easily than 'negative' ones (Gluud 1998).

Our findings regarding primary outcome measures did not seem to be sensitive to missing data. Neither the reported scenario nor the likely scenario analyses, which are attempts to fill in missing data in a realistic manner, showed any significant difference between colchicine and placebo/no intervention. The best-case scenario did not show any significant difference. The worst-case scenario detected a significant detrimental effect of colchicine. Although the best-worst-case- and worst-best-case-scenario analyses are extreme and unlikely, it is more probable that the treatment effect did not favour colchicine but placebo/no intervention. Additionally, we found that the effect of colchicine on mortality and liver transplantation (favouring colchicine in the best-worst-case-scenario analyses and favouring placeb/no intervention in the worst-best-case-scennario analysis) to be heavily depended on the Bodenheimer 1988 trial, in which the rate of loss of follow-up in the colchicine group was 28.5%. After excluding this trial, we got a non-significant difference (Comparison 06-03). In addition, when we included data from 114 patients from two trials switched from blinded to open label colchicine therapy, these differences were not significant either on mortality or on mortality or liver transplantation.

In order to examine the effects of colchicine in a broader context, we expanded our analyses by including trials on colchicine versus placebo/no intervention for alcoholic and non-alcoholic liver fibrosis and cirrhosis (Rambaldi 2003). The pooled results showed no significant difference on mortality. In the colchicine group 109/786 (13.9%) patients died versus 106/762 (13.9%) patients in the control group (see Figure 01).

The Goddard et al. trial was a 2 multiplied by 2 factorial designed trial, which could have investigated the possible interaction between colchicine and UDCA. However, the trial was only published as an abstract and the author did not reply to our request for further information. A synergistic effect was claimed based on a non-randomised study (Shibata 1992). However, our subgroup analyses, stratifying the included trials into monotherapy (i.e., colchicine versus placebo/no intervention) and combination therapy (i.e., colchicine plus UDCA versus placebo/no intervention plus UDCA) did not suggest additional effect of colchicine introduced by the combination with UDCA in the identified trials.

We found that colchicine had a significant beneficial effect on pruritus. This finding was from three trials involving only 156 patients. A number of arguments may contradict this observation. First, lack of efficient blinding of trials (Kjaergard 2001) and the subjective nature of pruritus assessment could have biased the estimate. Second, pruritus usually reflects indices of cholestasis

(e.g., serum alkaline phosphatases) and a correlation between the severity of pruritus and the presence of florid bile duct lesions in the liver has been reported (Poupon 1999). Our analyses did not show any significant effect of colchicine on any plasma indices of cholestasis or on liver histology. Furthermore, due to the large number of statistical comparisons having been performed some of the comparisons might have come out with a significant difference simply due to 'mass significance'. Therefore, we are not convinced that the improvement of pruritus was due to colchicine. The potential beneficial effect of colchicine on pruritus might be worth exploring in future high-quality randomised trials.

We did not find any significant difference on liver biochemical parameters between colchicine treatment and placebo/no intervention. It appeared that the use of colchicine was associated with improvement in hepatic biochemistries in three early randomised clinical trials (Kaplan 1986; Warnes 1987; Bodenheimer 1988). In those three trials, however, the protocol violations regarding per cent of randomised patients who were: (i) lost to follow-up; (ii) refused liver biopsy; (iii) were noncompliant, and (iv) were withdrawn due to adverse events or disease progression - were: 33%, 13%, and 38%, respectively. Only one trial (Warnes 1987) stated having employed the intention-to-treat principle.

Primary biliary cirrhosis is a pathological process starting with portal inflammation, which progresses towards three irreversible stages: a stage of compensated cirrhosis, a stage of decompensated cirrhosis (defined by high bilirubin levels (greater than 100 µmol/L), ascites, and variceal bleeding), and a terminal stage, in which death occurs unless liver transplantation is performed (Gluud 2002). The purpose of the randomised clinical trials assessing colchicine for primary biliary cirrhosis has not been to evaluate whether colchicine could reverse the decompensated stage or the terminal stage of the disease, but rather, if colchicine could slow the progression towards the cirrhotic stage and the more advanced stages. It is, therefore, interesting to study the effect of colchicine on liver histology. In this review, we were not able to identify any significant effect of colchicine on a number of histological variables. The Almasio 2000 trial reported a significant reduction in histological grading score in patients administrating colchicine plus ursodeoxycholic acid; however, the proportion of patients having liver biopsy was very low (15 patients out of 90). Thus, its significance could be biased by impact of missing data on liver histology.

Vuoristo et al. performed comparison of colchicine versus ursodeoxycholic acid, the most widely used drug in the treatment of primary biliary cirrhosis. No significant difference was detected regarding mortality (Vuoristo 1995). Colchicine appeared to relieve fatigue, but the effect size was small. For the liver biochemical outcomes, the significant difference detected on s-alkaline phosphatases and gamma-glutamyltransferase was suggestive of a favourable effect of ursodeoxycholic acid. This is in accordance with trials comparing ursodeoxycholic acid with placebo/no intervention (Gluud 2002). Overall, we were not able to suggest beneficial effect of colchicine compared to ursodeoxycholic acid.

Compared to methotrexate, a folic acid antagonist that blocks nucleic acid synthesis, colchicine seemed to be less effective against methotrexate regarding severity of pruritus and level of s-alkaline phosphases and plasma immunoglobulin M. The data we extracted were from a two-year interim analysis of the ten-year Kaplan 1999 trial. The trial is finished, but the published data are not available presently (October 2003).

Regarding the safety issue of colchicine treatment in primary biliary cirrhosis, this systematic review could not demonstrate that colchicine was associated with an increase or decrease of non-serious adverse events (mainly transient diarrhoea, usually resolved by lowering the dose of colchicine) or serious adverse events. We were not able to identify data on the effects of colchicine concerning quality of life and health economics.

AUTHORS' CONCLUSIONS

Implications for practice

We did not find convincing evidence showing that colchicine had significant beneficial effects on patients with primary biliary cirrhosis when compared to placebo or no intervention. The combination of colchicine and UDCA did not significantly change the effects of colchicine. We are not able to exclude the possibility that colchicine may reduce mortality by 70%. On the other hand, it may increase mortality by 344%. We therefore cannot recommend the use of colchicine outside randomised clinical trials.

Implications for research

If researchers have an interest to investigate colchicine for primary biliary cirrhosis, they may consider the following:

- due to the chronic progression of primary biliary cirrhosis and thanks to the low toxicity of colchicine, long-term follow-up is needed and seems feasible;
- to have an independent data monitoring and safety committee, which can follow the data and stop the trial should it start to demonstrate harmful effects of colchicine
- to study in detail the potential effect of colchicine on pruritus;
- to ensure that enough patients with primary biliary cirrhosis are kept followed to undergo liver biopsy in order to obtain more data on liver histology;
- to include quality-of-life and health economics analyses;
- to adhere to the Consort Statement (www.consort-statement.org).

FEEDBACK

Colchicine for primary biliary cirrhosis

Summary

Date of Submission: 13-Jun-2006
Name: Roger Pepin
Email Address: r.pepin@elsevier.com
Personal Description: Occupation EBM Editor

Feedback: Could someone clarify the relationship of this review and the review published in American Journal of Gastroenterology, 2005 Aug;100(8):1876-85. In particular, could they comment on the differing numbers of trials included in each review, and the observations made of trial quality in each abstract.

Submitter agrees with default conflict of interest statement:
I certify that I have no affiliations with or involvement in any organization or entity with a financial interest in the subject matter of my feedback.

Author's reply

We published the systematic review in The Cochrane Library 2004, Issue 2, based on which we published a modified version in The American Journal of Gastroenterology in 2005. The Cochrane Hepato-Biliary Group has endorsed a co-publication agreement with The American Journal of Gastroenterology.

The trials included in the review published in The American Journal of Gastroenterology compare colchicine versus placebo/no intervention. The review published in The Cochrane Library is broader since it also includes trials comparing colchicine versus ursodeoxycholic acid and colchicine versus methotrexate.

We stated in the The American Journal of Gastroenterology the following:
'This review is published as a Cochrane Review in The Cochrane Library 2004, Issue 2. Cochrane Reviews are regularly updated as new evidence emerges and in response to comments and criticisms. The Cochrane Library should be consulted for the most recent version of the Review.'

Contributors

Yan Gong, primary author
23 August 2006
Copenhagen, Denmark

Christian Gluud, author and Criticism Editor
23 August 2006
Copenhagen, Denmark

POTENTIAL CONFLICT OF INTEREST

None known.

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Gong 2005
Gong Y, Gluud C. Colchicine for primary biliary cirrhosis: a Cochrane Hepato-Biliary Group Systematic Review of randomized

clinical trials. *American Journal of Gastroenterology* 2005;**100**:1876–85.

* *Indicates the major publication for the study*

TABLES

Characteristics of included studies

Study	**Almasio 2000**
Methods	Generation of the allocation sequence: adequate, computer-generated list. Allocation concealment: adequate, central unit. Blinding: adequate, double-blinding, indistinguishable placebo. Follow-up: adequate, 6/90 patients dropped out: 2 on UDCA plus placebo, 4 on UDCA plus colchicine.
Participants	Country: Italy. 90 patients (9 males and 81 females, being 55.5 ± 10.9 years in the UDCA/P group and 53.3 ± 10.2 years in UDCA/C group). Inclusion criteria: 1. An established diagnosis of primary biliary cirrhosis according to Taal et al. 2. Pruritus. 3. Serum bilirubin exceeding 2 mg/dL. 4. Histological diagnosis of cirrhosis. Exclusion criteria: 1. Ascites. 2. Gastrointestinal bleeding or encephalopathy. 3. Serum bilirubin levels exceeding 10 mg/dL. 4. Evidence of malignant conditions or of other major diseases unrelated to PBC. 5. Alcohol abuse. 6. Previous treatment with colchicine or immunosuppressant agents. 7. Low compliance.
Interventions	a) Colchicine plus UDCA: 1 mg/day colchicine plus 250 mg UDCA twice daily. b) Placebo plus UDCA: placebo plus 250 mg UDCA twice daily. Duration of medication: 3 years.
Outcomes	1. Biochemical variables. 2. Ig M. 3. Mayo score. 4. Major clinical events: death, liver transplantation, decompensation of liver disease, doubling of bilirubin. 5. Liver biopsy findings.
Notes	1. It was a multicenter-study (six centres). 2. Sent letter (4 Nov. 2002). P. L. Almasio responded and provided additional data on liver biochemical variables. 3. This trial included the 44 patients described by Battezzati 2001, which followed patients for up to 10 years of treatment: 6/44 patients dropped out: 4 in UDCA+placebo, 2 in UDCA+colchicine.
Allocation concealment	A – Adequate

Characteristics of included studies (*Continued*)

Study	**Bodenheimer 1988**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: unclear. Blinding: adequate, double-blinding, identically appearing placebo. Follow-up: adequate, 14/57 withdrew and lost to follow-up during the blind period of the trial (6 in placebo, 8 in colchicine). In addition, one patient in the control group lost to follow-up in the opened label period. Sample size estimation: no.
Participants	Country: USA. 57 patients (5 males and 52 females; mean age: 53 years in colchicine group and 51 years in placebo group). Inclusion criteria: 1. History of chronic cholestatic liver disease. 2. Liver biopsy results compatible with PBC.
Interventions	a) Colchicine: 0.6 mg, twice daily. b) Placebo: Identically appearing placebo. Duration of medication: 4 years.
Outcomes	1. Biochemical variables. 2. Immunological variables. 3. Histologic parameters proven by liver biopsy. 4. Number of death and number of patients undergoing liver transplantation. 5. Adverse events: diarrhoea, etc.
Notes	1. Patients assigned to placebo at entry were crossed to opened label colchicine for 4 additional years after the first 4-year double blind interventions. The results of this trial were published by Zifroni 1991. 2. Sent letter (4 Nov. 2002), but no response received.
Allocation concealment	B – Unclear

Study	**Goddard 1995**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: unclear. Blinding: unclear. Follow-up: inadequate.
Participants	Country: UK Sample size: 57. Inclusion criteria: unclear.
Interventions	a) Colchicine: 1mg/day colchicine. b) UDCA: 10mg/kg/day UDCA. c) Colchicine plus UDCA. d) Placebo. Duration of treatment: 30 months.
Outcomes	Biochemical variables.
Notes	1. Published as an abstract. 2. Sent letter (4 Nov. 2002), but no response received.
Allocation concealment	B – Unclear

Characteristics of included studies (*Continued*)

Study	**Ikeda 1996**
Methods	Generation of the allocation sequence: inadequate, a consecutive case number. Allocation concealment: adequate, sealed envelope. Blinding: not performed. Follow-up: adequate, no patients withdrew/lost to follow-up/drop-out.
Participants	Country: Japan. 22 patients (3 males and 19 females; being 59.5 ± 3 years in UDCA/C group and 66.5 ± 3 years in UDCA group). Inclusion criteria: 1. Elevation of alkaline phosphatase over the upper limit of normal. 2. AMA. 3. Compatible histological appearance of liver biopsy specimens. 4. Radiological or ultrasonographic evidence that the bile ducts were patent.
Interventions	a) Colchicine plus UDCA: 1 mg/day colchicine plus 600 mg/day UDCA. b) UDCA alone: 600 mg/day UDCA. Duration of combined medication: 2 years.
Outcomes	1. Biochemical variables. 2. Adverse events: diarrhoea. 3. Clinical findings: pruritus, oesophageal varices. 4. Major clinical events: death, liver transplantation.
Notes	1. Before randomisation, all the patients were treated 600 mg/day UDCA for 30 months. 2. Sent letter (4 Nov. 2002). T. Ikeda responded and provided the information on trial design, clinical findings, adverse events, and liver biochemical variables.
Allocation concealment	A – Adequate

Study	**Kaplan 1986**
Methods	Generation of the allocation sequence: adequate, randomisation scheme. Allocation concealment: adequate, a single study monitor. Blinding: adequate, double-blinding identically appearing placebo. Follow-up: adequate, 8/60 patients were classified as drop-outs: five in placebo group, three in colchicine group.
Participants	Country: USA. 60 patients (3 males and 57 females; mean age was not given). Inclusion criteria: 1. A positive test for antimitochondrial antibody. 2. Liver-biopsy proven PBC. 3. Radiologic or ultrasonographic evidence that bile ducts were patent.
Interventions	a) Colchicine: 0.6 mg colchicine twice daily. b) Placebo: Identically appearing placebo. Duration of blinded medication: two years. Duration of open label medication: the following two years.
Outcomes	1. Biochemical variables. 2. Clinical findings. 3. Liver histology score. 4. Cumulative mortality.

Characteristics of included studies (*Continued*)

	5. Adverse events: diarrhoea.
Notes	1. At the end of the two-year double-blind period, each patient was placed in an open-lable trial of colchicine, 0.6 mg twice daily, for additional two years. 2. Sent letter (4 Nov. 2002), but no response received.
Allocation concealment	A – Adequate

Study	**Kaplan 1999**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: adequate, a single study monitor. Blinding: adequate, double-blinding and double-dummy. Follow-up: inadequate.
Participants	Country: USA. 85 patients (3 males and 82 females; being 51 ± 1.4 years in colchicine group and 51 ± 1.5 years in methotrexate group). Inclusion criteria: 1. Serum ALP level of at least 2 times greater than the upper limit of normal. 2. Serum bilirubin level not greater than 10 mg/dL. 3. Liver biopsy performed consistent with PBC. 4. Radiological or ultrasonic evidence.
Interventions	a) Colchicine: 0.6 mg colchicine twice daily. b) Methotrexate: 15 mg/week, 5 mg every 12 hours 3 times. Duration of medication: 10 years.
Outcomes	1. Biochemical variables. 2. IgM. 3. Pruritus and fatigue. 4. Liver histological evidence.
Notes	1. It is an interim analysis of a ten-year trial. 2. 2/87 withdrew from the trial immediately after randomisation before they received any drugs, did not return for follow-up testings, and were not included in the analyses. Ten patients dropped out of the trial. The reasons were specified, but the number in each group was not given. 3. Sent letter (4 Nov. 2002). M. Kaplan responded, but did not provide additional information. The final results of this ten-year trial are waiting publication.
Allocation concealment	A – Adequate

Study	**Poupon 1996**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: unclear. Blinding: adequate, double-blinding, identically appearing placebo. Follow-up: adequate, 2 patients dropped out: 2 in UDCA + colchicine.
Participants	Countries: France and Canada. 74 patients (11 males and 63 females; being 55 ± 2 years in UDCA/C group and 52 ± 2 years in UDCA/P group). Inclusion criteria: 1. Biopsy-proven PBC. 2. No less than eight months previous treatment with UDCA(13-15 mg/kg/day). 3. ALP activity more than 1.5 times the upper limit of normal.

Characteristics of included studies (*Continued*)

	Exclusion criteria: 1. Drug therapy (except UDCA) for PBC during the 6 months (colchicine, azathioprine, chlorambucil, corticosteroids, D-penicillamine, and cyclosporine). 2. Serum bilirubin concentration greater than 100umol/L. 3. A serum albumin concentration less than 25 g/L. 4. Past or active bleeding form oesophageal varices. 5. Ascites. 6. Other identified cause of liver of biliary diseases. 7. Excessive alcohol consumption (greater than 50 g/day). 8. Severe intercurrent disease. 9. Age older than 75 years.
Interventions	a) Colchicine plus UDCA: 1 mg/day colchicine, 5 days/week plus UDCA (13 to 15 mg/kg/day). b) Placebo plus UDCA: identically appearing placebo plus UDCA (13 to 15 mg/kg/day). Duration of intervention and follow-up: 2 years.
Outcomes	1. Clinical findings. 2. Laboratory findings, including bilirubin level. 3. Serum markers of liver fibrosis. 4. Histologic parameters, including the degree of fibrosis. 5. Sulphobromophthalein pharmakinetics. 6. Clinical complications. 7. Adverse events: peripheral polyneuropathy.
Notes	1. This was a multicenter trial (10 study centres) and it included a subsample (22/74 patients) trial designed by Huet 1996 (only published as an abstract) in which all patients were given colchicine plus UDCA for additional 2 years at the end of the two-year double-blind period. 2. Sent letter (4 Nov. 2002), but no response received.
Allocation concealment	B – Unclear

Study	**Raedsch 1993**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: unclear. Blinding: unclear. Follow-up: adequate, 2/28 patients dropped out: 2 in UDCA plus colchicine.
Participants	Country: Germany. All 28 patients were females with a mean age of 54 years. Inclusion criteria: 1. Blood biochemistry. 2. Specific AMA. 3. Compatible liver histology.
Interventions	a) Colchicine plus UDCA: 1 mg/day colchicine plus 10 to 12 mg/kg/day UDCA. b) Placebo plus UDCA: placebo plus 10 to 12mg/kg/day UDCA . Duration of medication: 3 years.
Outcomes	1. Biochemical variables. 2. Immunological variables. 3. Clinical symptoms.

Characteristics of included studies (*Continued*)

	4. Histological parameters.
Notes	1. All patients were pretreated with UDCA 10 to 12 mg/kg/day for 12 months. 2. Sent letter (4 Nov. 2002), but no response received.
Allocation concealment	B – Unclear

Study	**Vuoristo 1995**
Methods	Generation of allocation sequence: adequate, computerized randomisation number. Allocation concealment: adequate, sealed envelopes. Blinding: adequate, double-blinding, placebo with identical looking and film-coated. Follow-up: adequate, 6/90 drop-outs: 3 in the placebo group, 3 in the colchicine group.
Participants	Country: Finland. 90 patients (16 males, 74 females; mean age: 57, 56 and 52 years in placebo, colchicine and UDCA group, respectively). PBC defined as: elevated alkaline phosphatases, liver biopsy compatible with PBC, and positive AMA. End-stage PBC and patients treated with drugs that might affect prognosis were excluded.
Interventions	a) Colchicine: 1 mg/day colchicine. b) UDCA: 12 to 15 mg/kg/day UDCA. c) Placebo. Duration of medication: two years.
Outcomes	1. Major clinical events: death, liver transplantation, etc.. 2. Clinical findings. 3. Liver biochemistry. 4. Liver histology.
Notes	1. Sent letter (4 Nov. 2002). Vuoristo responded and provided additional information on trial design, clinical findings and liver biochemical variables.
Allocation concealment	A – Adequate

Study	**Warnes 1987**
Methods	Generation of the allocation sequence: adequate, random tables. Allocation of concealment: adequate, staff pharmacist. Blinding: adequate, double-blinding, identical placebo. Follow-up: adequate, 10/64 patients withdrew: 8 on colchicine, 2 on placebo.
Participants	Country: UK. Sample size: 89. Inclusion criteria: 1. A raised serum ALP. 2. A positive AMA test. 3. Liver histology compatible with, or diagnostic of PBC.
Interventions	a) Colchicine: 500ug, twice daily. b) Placebo: Identical placebo. Duration of medication is 12 months. Median duration of follow-up at the time of analysis was 23 months in the colchicine group and 15 months in the placebo group.
Outcomes	1. Biochemical findings.

	2. Immunological findings. 3. Liver histological findings. 4. Survival data. 5. Adverse events: diarrhoea, upper gastrointestinal symptoms, peripheral neuropathy, proteinuria, etc.
Notes	1. Pair-matched study, patients being matched on the basis of age and serum bilirubin. 2. Biochemical, immunological and histological findings at 12 months were compared, whilst survival data were compared up to 18 months. 3. Patients' age and sex ratio were not described. 4. Sent letter (4 Nov. 2002), but no response received.
Allocation concealment	A – Adequate

Study	**Warnes 1996**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: unclear. Blinding: unclear. Follow-up: inadequate.
Participants	Country: UK Sample size: 89. Inclusion criteria: unclear.
Interventions	a) Colchicine b) Placebo.
Outcomes	Biochemical findings: serum bilirubin, galactose elimination capacity and serum albumin.
Notes	1. Published as an abstract. 2. Sent letter (4 Nov. 2002), but no response received.
Allocation concealment	B – Unclear

Ig: immunoglobulin
UDCA: ursodeoxycholic acid
PBC: primary biliary cirrhosis
AMA: antimitochondrial antibody
ALP: alkaline phosphatases

Characteristics of excluded studies

Study	**Reason for exclusion**
Klion 1990	An observational study. It compared the risk score (R) using the Mayo model for a group of patients treated with colchicine using their pre-treatment period as control.
Koldinger 1980	A case series of five patients with PBC for periods ranging from 12 to 40 months.
Shibata 1992	A non-randomised trial. They divided twelve patients with PBC into two groups, one with UDCA and one with colchicine for three months. After three months both groups received combination therapy.

ADDITIONAL TABLES

Table 01. Search strategy for identification of studies

Database	Period	Search Strategy Used
The Controlled Trial Register of The Cochrane Hepato-Biliary Group	September 2003	#1 = 'RCT' and ' PRIMARY BILIARY CIRRHOSIS' and ' COLCHICINE'
The Cochrane Library (CENTRAL)	2003 Issue 3	#1 = LIVER-CIRRHOSIS-BILIARY*: MESH #2 = (PRIMARY and BILIARY and CIRRHOSIS) or PBC #3 = COLCHICINE: MESH #4 = IMMUNOSUPPRES* : MESH #5 = URSODEOXYCHOLIC-ACID: MESH #6 = COLCHICINE or IMMUNOSUPPRES* or (URSODEOXYCHOLIC and ACID) #7 = #3 or #4 or #5 or #6 #8 = (#1 and #7) #9 = (#2 and #7) #10 = (#8 or #9)
MEDLINE	January 1966 to September 2003	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 =(PRIMARY and BILIARY and CIRRHOSIS) or PBC #3 = "PRIMARY BILIARY CIRRHOSIS" or PBC #4 = #2 or #3 #5 = COLCHICINE #6 = IMMUNOSUPPRES* #7 = URSODEOXYCHOLIC #8 = ACID #9 = #5 or #6 or (#7 and #8) #10 = COLCHICINE: MESH #11 = IMMUNOSUPPRESS*: MESH #12 = URSODEOXYCHOLIC-ACID: MESH #13 = #9 or #10 or #11 or #12 #14 = #1 and #13 #15 = #4 and #13 #16 = #14 or #15 #17 = random* #18 = placebo* #19 = blind* #20 = meta-analysis #21 = #17 or #18 or #19 or #20 #22 = #16 and # 21
EMBASE	January 1980 to September 2003	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 = (PRIMARY and BILIARY and CIRRHOSIS) or PBC #3 = "PRIMARY BILIARY CIRRHOSIS" or PBC #4 = #2 or #3 #5 = COLCHICINE

Table 01. Search strategy for identification of studies (*Continued*)

Database	Period	Search Strategy Used
		#6 = IMMUNOSUPPRES* #7 = URSODEOXYCHOLIC #8 = ACID #9 = #5 or #6 or (#7 and #8) #10 = COLCHICINE: MESH #11 = IMMUNOSUPPRESS*: MESH #12 = URSODEOXYCHOLIC-ACID: MESH #13 = #9 or #10 or #11 or #12 #14 = #1 and #13 #15 = #4 and #13 #16 = #14 or #15 #17 = random* #18 = placebo* #19 = blind* #20 = meta-analysis #21 = #17 or #18 or #19 or #20 #22 = #16 and # 21

ANALYSES

Comparison 01. Colchicine versus placebo/no intervention

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Number of deaths	7	398	Relative Risk (Fixed) 95% CI	1.21 [0.71, 2.06]
02 Number of deaths and/or patients who underwent liver transplantation	8	455	Relative Risk (Fixed) 95% CI	1.00 [0.67, 1.49]
03 Number of patients who underwent liver transplantation	5	274	Relative Risk (Fixed) 95% CI	0.34 [0.06, 2.10]
04 Number of patients without improvement of pruritus	3	156	Relative Risk (Fixed) 95% CI	0.75 [0.65, 0.87]
05 Number of patients without improvement of fatigue	1	60	Relative Risk (Fixed) 95% CI	0.86 [0.72, 1.02]
06 Number of patients developing liver complications	3	156	Relative Risk (Fixed) 95% CI	0.37 [0.12, 1.10]
07 Appearence of liver complications			Relative Risk (Fixed) 95% CI	Subtotals only
08 S-bilirubin (µmol/L)	4	202	Weighted Mean Difference (Fixed) 95% CI	-1.35 [-4.52, 1.82]
09 S-alkaline phosphatases (ALP)(IU/L)	4	200	Weighted Mean Difference (Fixed) 95% CI	-55.35 [-158.56, 47.85]
10 S-gamma-glutamyltransferase (GGT)(IU/L)	4	200	Weighted Mean Difference (Fixed) 95% CI	-25.38 [-73.26, 22.50]
11 S-aspartate aminotransferase (AST)(IU/L)	2	82	Weighted Mean Difference (Fixed) 95% CI	-10.10 [-22.91, 2.71]
12 S-alanine aminotransferase (ALT)(IU/L)	4	201	Weighted Mean Difference (Fixed) 95% CI	-2.05 [-8.79, 4.68]

13 S-albumin (g/dL)	4	235	Weighted Mean Difference (Fixed) 95% CI	0.09 [-0.03, 0.21]
14 S-cholesterol (total) (mmol/L)	1	60	Weighted Mean Difference (Fixed) 95% CI	0.10 [-0.88, 1.08]
15 Plasma immunoglobulin M (g/L)	4	198	Weighted Mean Difference (Fixed) 95% CI	-0.49 [-1.03, 0.06]
16 Prothrombin time (second)	1	57	Weighted Mean Difference (Fixed) 95% CI	-0.03 [-0.75, 0.69]
17 Liver biopsy findings - dichotomous variables			Odds Ratio (Fixed) 95% CI	Subtotals only
18 Liver biopsy findings - histological score	1	50	Weighted Mean Difference (Fixed) 95% CI	0.56 [-0.24, 1.36]
19 Number of patients with adverse events	8	455	Relative Risk (Fixed) 95% CI	1.45 [0.94, 2.25]
20 Number of patients with serious adverse events	8	455	Relative Risk (Fixed) 95% CI	1.17 [0.50, 2.75]

Comparison 02. Colchcine - colchicine versus placebo - colchicine (including open label period)

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Number of deaths	2	117	Relative Risk (Fixed) 95% CI	1.15 [0.76, 1.73]
02 Number of deaths and/or patients who underwent liver transplantation	2	117	Relative Risk (Fixed) 95% CI	1.02 [0.72, 1.46]

Comparison 03. Colchicine versus ursodeoxycholic acid

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Number of deaths	1	59	Relative Risk (Fixed) 95% CI	11.37 [0.66, 196.74]
02 Number of deaths and/or patients who underwent liver transplantation	1	59	Relative Risk (Fixed) 95% CI	11.37 [0.66, 196.74]
03 Number of patients who underwent liver transplantation	1	59	Relative Risk (Fixed) 95% CI	3.10 [0.13, 73.14]
04 Number of patients without improvement of pruritus	1	59	Relative Risk (Fixed) 95% CI	0.78 [0.55, 1.09]
05 Number of patients without improvement of fatigue	1	59	Relative Risk (Fixed) 95% CI	0.83 [0.70, 0.98]
06 Appearance of liver complications			Relative Risk (Fixed) 95% CI	Subtotals only
07 S-bilirubin (µmol/L)	1	59	Weighted Mean Difference (Fixed) 95% CI	3.40 [-13.26, 20.06]
08 S-alkaline phosphatases (ALP)(IU/L)	1	59	Weighted Mean Difference (Fixed) 95% CI	378.00 [116.92, 639.08]
09 S-gamma-glutamyltransferase (GGT)(IU/L)	1	59	Weighted Mean Difference (Fixed) 95% CI	459.00 [157.57, 760.43]
10 S-aspartate aminotransferase (AST)(IU/L)	1	59	Weighted Mean Difference (Fixed) 95% CI	19.00 [-8.86, 46.86]
11 S-alanine aminotransferase (ALT)(IU/L)	1	59	Weighted Mean Difference (Fixed) 95% CI	24.00 [-8.62, 56.62]
12 S-albumin (g/dL)	1	59	Weighted Mean Difference (Fixed) 95% CI	0.04 [-0.14, 0.22]
13 S-cholesterol (total) (mmol/L)	1	59	Weighted Mean Difference (Fixed) 95% CI	0.00 [-1.14, 1.14]
14 Plasma immunoglobulin M (g/L)	1	59	Weighted Mean Difference (Fixed) 95% CI	0.70 [-0.99, 2.39]

15 Number of patients with adverse events		Relative Risk (Fixed) 95% CI	Subtotals only

Comparison 04. Colchicine versus methotrexate

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Number of deaths and/or patients who underwent liver transplantation	1	85	Relative Risk (Fixed) 95% CI	0.80 [0.37, 1.73]
02 Pruritus score	1	85	Weighted Mean Difference (Fixed) 95% CI	0.68 [0.25, 1.11]
03 Fatigue score	1	85	Weighted Mean Difference (Fixed) 95% CI	-0.11 [-0.54, 0.32]
04 S-bilirubin (µmol/L) (presented as logtransformed geometric mean)	1	85	Weighted Mean Difference (Fixed) 95% CI	-0.07 [-0.37, 0.23]
05 S-alkaline phosphatases (ALP)(IU/L) (presented as logtransformed geometric mean)	1	85	Weighted Mean Difference (Fixed) 95% CI	0.41 [0.12, 0.70]
06 S-aspartate aminotransferase (AST)(IU/L) (presented as logtransformed geometric mean)	1	85	Weighted Mean Difference (Fixed) 95% CI	0.06 [-0.18, 0.30]
07 S-alanine aminotransferase (ALT)(IU/L) (presented as logtransformed geometric mean)	1	85	Weighted Mean Difference (Fixed) 95% CI	0.00 [-0.26, 0.26]
08 S-albumin (g/dL)	1	85	Weighted Mean Difference (Fixed) 95% CI	0.17 [-0.01, 0.35]
09 S-cholesterol (total) (mmol/L) (presented as logtransformed geometric mean)	1	85	Weighted Mean Difference (Fixed) 95% CI	0.09 [-0.03, 0.21]
10 Plasma immunoglobulin M (g/L) (presented as logtransformed geometric mean)	1	83	Weighted Mean Difference (Fixed) 95% CI	0.47 [0.20, 0.74]
11 Prothrombin time (second)	1	85	Weighted Mean Difference (Fixed) 95% CI	-0.14 [-0.54, 0.26]
12 Liver biopsy findings - histological stage	1	55	Weighted Mean Difference (Fixed) 95% CI	0.10 [-0.60, 0.80]
13 Liver biopsy findings - histological score	1	55	Weighted Mean Difference (Fixed) 95% CI	1.40 [-1.53, 4.33]
14 Number of patients with adverse events	1	85	Relative Risk (Fixed) 95% CI	1.47 [0.45, 4.82]

Comparison 05. Subgroup analyses - colchicine versus placebo/no intervention

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Number of deaths - dose variation	7	398	Relative Risk (Fixed) 95% CI	1.21 [0.71, 2.06]
02 Number of deaths - treatment duration	7	398	Relative Risk (Fixed) 95% CI	1.21 [0.71, 2.06]
03 Number of deaths - generation of the allocation sequence	7	398	Relative Risk (Fixed) 95% CI	1.21 [0.71, 2.06]

04 Number of deaths - allocation concealment	7	398	Relative Risk (Fixed) 95% CI	1.21 [0.71, 2.06]
05 Number of deaths - blinding	7	398	Relative Risk (Fixed) 95% CI	1.21 [0.71, 2.06]
06 Number of deaths and/or patients who underwent liver transplantation - dose variation	8	455	Relative Risk (Fixed) 95% CI	1.00 [0.67, 1.49]
07 Number of deaths and/or patients who underwent liver transplantation - treatment duration	8	455	Relative Risk (Fixed) 95% CI	1.00 [0.67, 1.49]
08 Number of deaths and/or patients who underwent liver transplantation - generation of the allocation sequen	8	455	Relative Risk (Fixed) 95% CI	1.00 [0.67, 1.49]
09 Number of deaths and/or patients who underwent liver transplantation - allocation concealment	8	455	Relative Risk (Fixed) 95% CI	1.00 [0.67, 1.49]
10 Number of deaths and/or patients who underwent liver transplantation - blinding	8	455	Relative Risk (Fixed) 95% CI	1.00 [0.67, 1.49]
11 S-bilirubin (µmol/L) - reported as arithmetic mean or geometric mean			Weighted Mean Difference (Fixed) 95% CI	Subtotals only
12 S-cholesterol (total) (mmol/L) - reported as arithmetic mean or geometric mean			Weighted Mean Difference (Fixed) 95% CI	Subtotals only
13 S-alkaline phosphatase (ALP) (IU/L) - reported as arithmetic mean or geometric mean			Weighted Mean Difference (Fixed) 95% CI	Subtotals only

Comparison 06. Sensitivity analyses - colchicine versus placebo/no intervention

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Number of deaths			Relative Risk (Fixed) 95% CI	Subtotals only
02 Number of deaths and/or patients who underwent liver transplantation			Relative Risk (Fixed) 95% CI	Subtotals only
03 Number of deaths and/or patients who underwent liver transplantation (excluding Bodenheimer 1988)			Relative Risk (Fixed) 95% CI	Subtotals only

INDEX TERMS

Medical Subject Headings (MeSH)

Cholagogues and Choleretics [*therapeutic use]; Colchicine [*therapeutic use]; Liver Cirrhosis, Biliary [*drug therapy; mortality]; Liver Transplantation; Methotrexate [therapeutic use]; Randomized Controlled Trials; Ursodeoxycholic Acid [therapeutic use]

MeSH check words

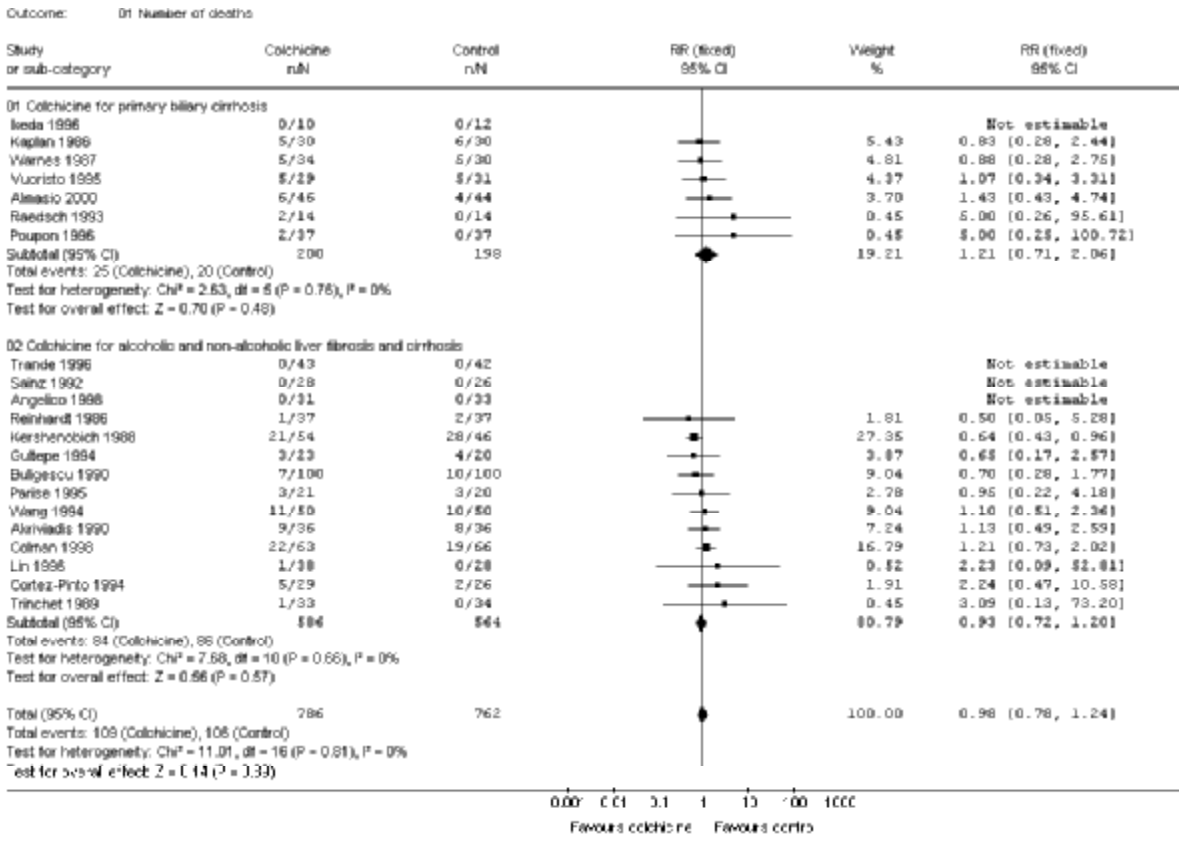
Humans

COVER SHEET

Title	Colchicine for primary biliary cirrhosis
Authors	Gong Y, Gluud C
Contribution of author(s)	YG performed the searches, selected trials for inclusion, wrote to authors and pharmaceutical companies, performed data extraction and data analyses, and drafted the protocol and the systematic review. CG formulated the idea of this review and revised the protocol, selected trials for inclusion, validated, solved discrepancy of data extraction, and revised the review.
Issue protocol first published	2003/4
Review first published	2004/2
Date of most recent amendment	23 August 2006
Date of most recent SUBSTANTIVE amendment	24 February 2004
What's New	Serum immunoglobulins generally reveal an elevated immunoglobulin M (IgM) value. Because of this, we decided to replace the biochemical outcome specified in the protocol as immunoglobulins with measurement of plasma immunoglobulin M.
Date new studies sought but none found	05 October 2003
Date new studies found but not yet included/excluded	Information not supplied by author
Date new studies found and included/excluded	Information not supplied by author
Date authors' conclusions section amended	Information not supplied by author
Contact address	Dr Yan Gong Copenhagen Trial Unit Centre for Clinical Intervention Research, Copenhagen University Hospital Dept. 7102, Blegdamsvej 9 H:S Rigshospitalet Copenhagen DK-2100 DENMARK E-mail: ygong@ctu.rh.dk Tel: +45 3545 7161 Fax: +45 3545 7101
DOI	10.1002/14651858.CD004481.pub2
Cochrane Library number	CD004481
Editorial group	Cochrane Hepato-Biliary Group
Editorial group code	HM-LIVER

GRAPHS AND OTHER TABLES

Figure 01. Relative risk of mortality in patients with primary biliary cirrhosis, alcoholic, and non-alcoholic liver fibrosis and cirrhosis randomised to colchicine versus placebo/no intervention

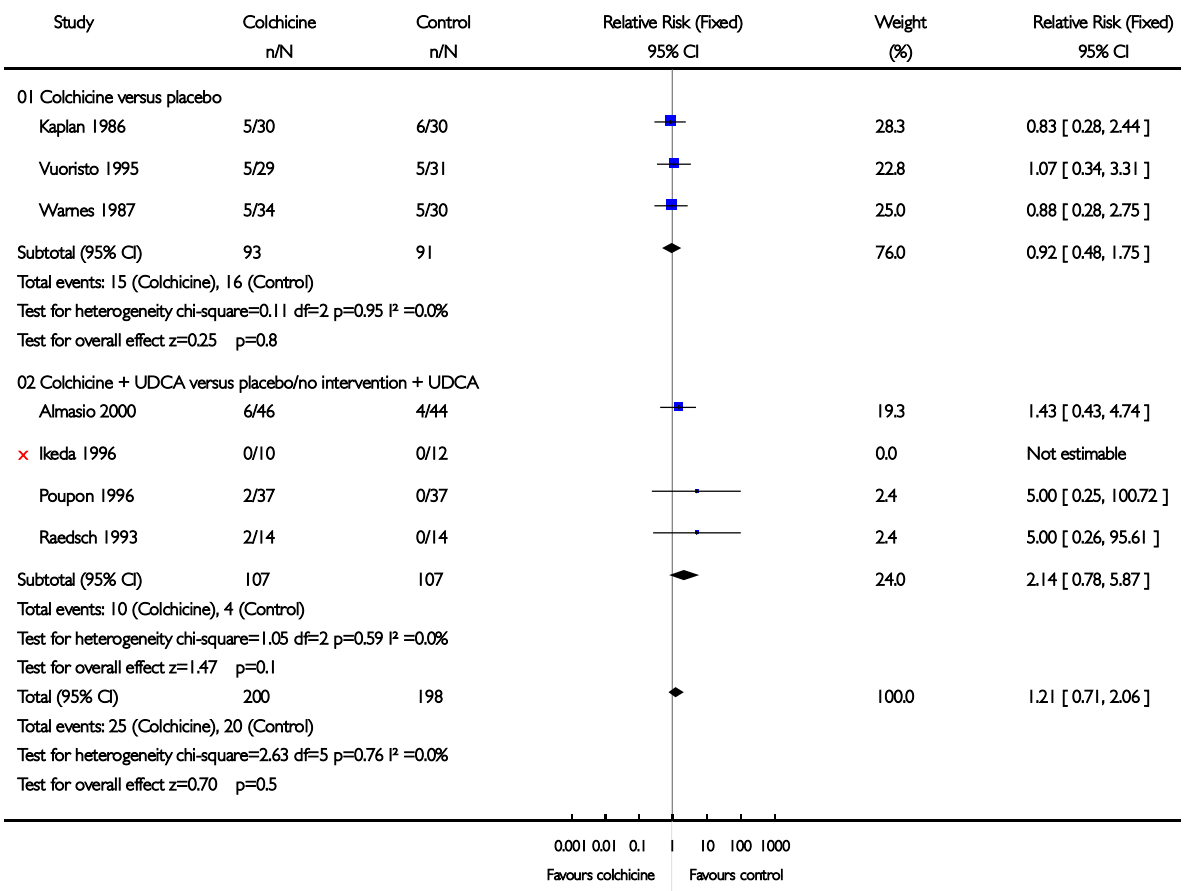


Analysis 01.01. Comparison 01 Colchicine versus placebo/no intervention, Outcome 01 Number of deaths

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 01 Number of deaths

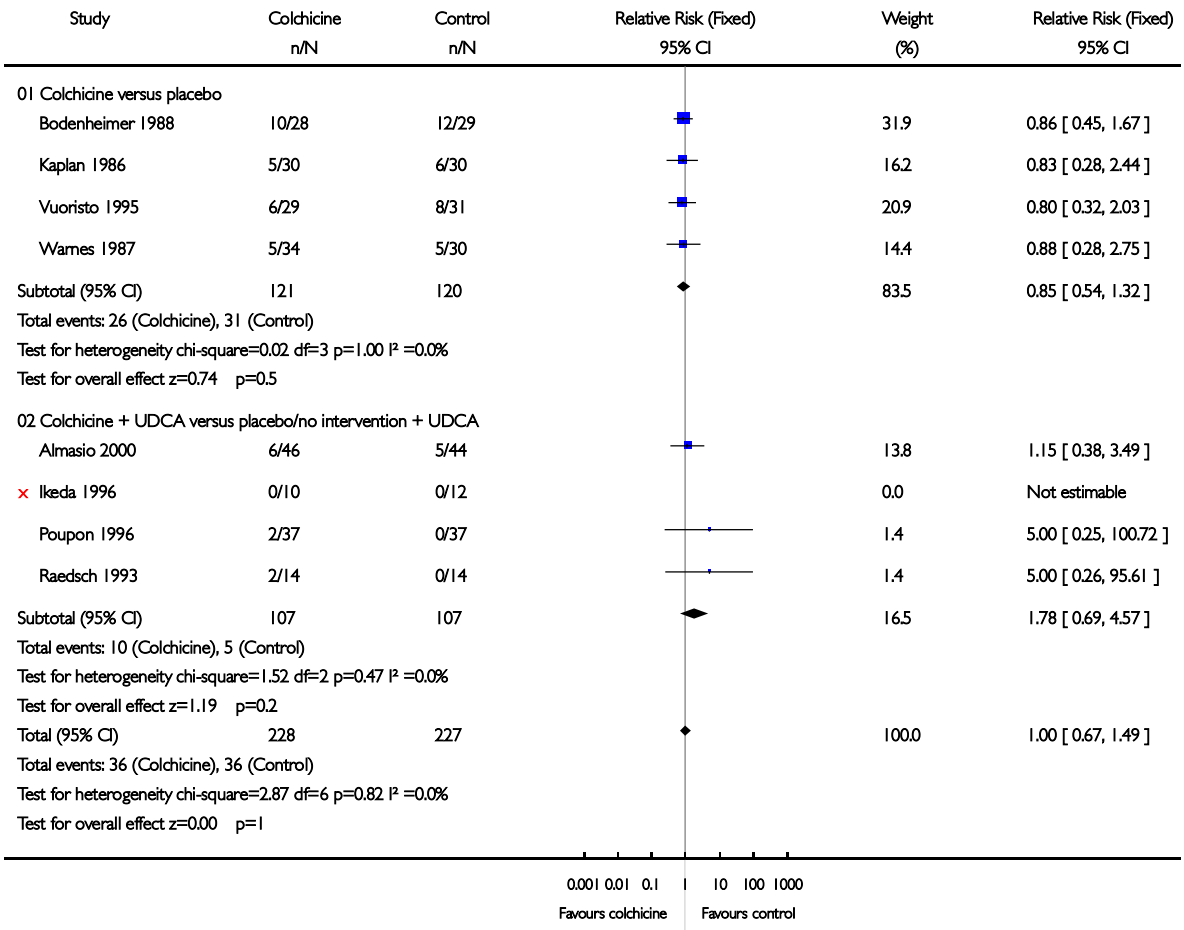


Analysis 01.02. Comparison 01 Colchicine versus placebo/no intervention, Outcome 02 Number of deaths and/or patients who underwent liver transplantation

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 02 Number of deaths and/or patients who underwent liver transplantation



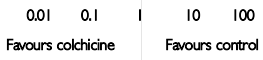
Analysis 01.03. Comparison 01 Colchicine versus placebo/no intervention, Outcome 03 Number of patients who underwent liver transplantation

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 03 Number of patients who underwent liver transplantation

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Colchicine versus placebo					
Vuoristo 1995	1/29	3/31		65.4	0.36 [0.04, 3.23]
Subtotal (95% CI)	29	31		65.4	0.36 [0.04, 3.23]
Total events: 1 (Colchicine), 3 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=0.92 p=0.4					
02 Colchicine + UDCA versus placebo/no intervention + UDCA					
Almasio 2000	0/46	1/44		34.6	0.32 [0.01, 7.63]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
× Poupon 1996	0/37	0/37		0.0	Not estimable
× Raedsch 1993	0/14	0/14		0.0	Not estimable
Subtotal (95% CI)	107	107		34.6	0.32 [0.01, 7.63]
Total events: 0 (Colchicine), 1 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=0.71 p=0.5					
Total (95% CI)	136	138		100.0	0.34 [0.06, 2.10]
Total events: 1 (Colchicine), 4 (Control)					
Test for heterogeneity chi-square=0.00 df=1 p=0.96 I^2 =0.0%					
Test for overall effect z=1.16 p=0.2					



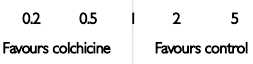
Analysis 01.04. Comparison 01 Colchicine versus placebo/no intervention, Outcome 04 Number of patients without improvement of pruritus

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 04 Number of patients without improvement of pruritus

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Colchicine versus placebo					
Vuoristo 1995	18/29	31/31		40.0	0.62 [0.47, 0.82]
Subtotal (95% CI)	29	31		40.0	0.62 [0.47, 0.82]
Total events: 18 (Colchicine), 31 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=3.29 p=0.001					
02 Colchicine + UDCA versus placebo/no intervention + UDCA					
Ikeda 1996	8/10	12/12		14.6	0.80 [0.59, 1.09]
Poupon 1996	29/37	34/37		45.4	0.85 [0.70, 1.04]
Subtotal (95% CI)	47	49		60.0	0.84 [0.71, 0.99]
Total events: 37 (Colchicine), 46 (Control)					
Test for heterogeneity chi-square=0.12 df=1 p=0.73 I² =0.0%					
Test for overall effect z=2.06 p=0.04					
Total (95% CI)	76	80		100.0	0.75 [0.65, 0.87]
Total events: 55 (Colchicine), 77 (Control)					
Test for heterogeneity chi-square=3.51 df=2 p=0.17 I² =43.0%					
Test for overall effect z=3.79 p=0.0002					



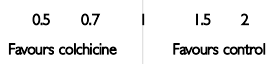
Analysis 01.05. Comparison 01 Colchicine versus placebo/no intervention, Outcome 05 Number of patients without improvement of fatigue

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 05 Number of patients without improvement of fatigue

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Colchicine versus placebo					
Vuoristo 1995	24/29	30/31		100.0	0.86 [0.72, 1.02]
Subtotal (95% CI)	29	31		100.0	0.86 [0.72, 1.02]
Total events: 24 (Colchicine), 30 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.72 p=0.09					
02 Colchicine + UDCA versus placebo/no intervention + UDCA					
Subtotal (95% CI)	0	0		0.0	Not estimable
Total events: 0 (Colchicine), 0 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect: not applicable					
Total (95% CI)	29	31		100.0	0.86 [0.72, 1.02]
Total events: 24 (Colchicine), 30 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.72 p=0.09					

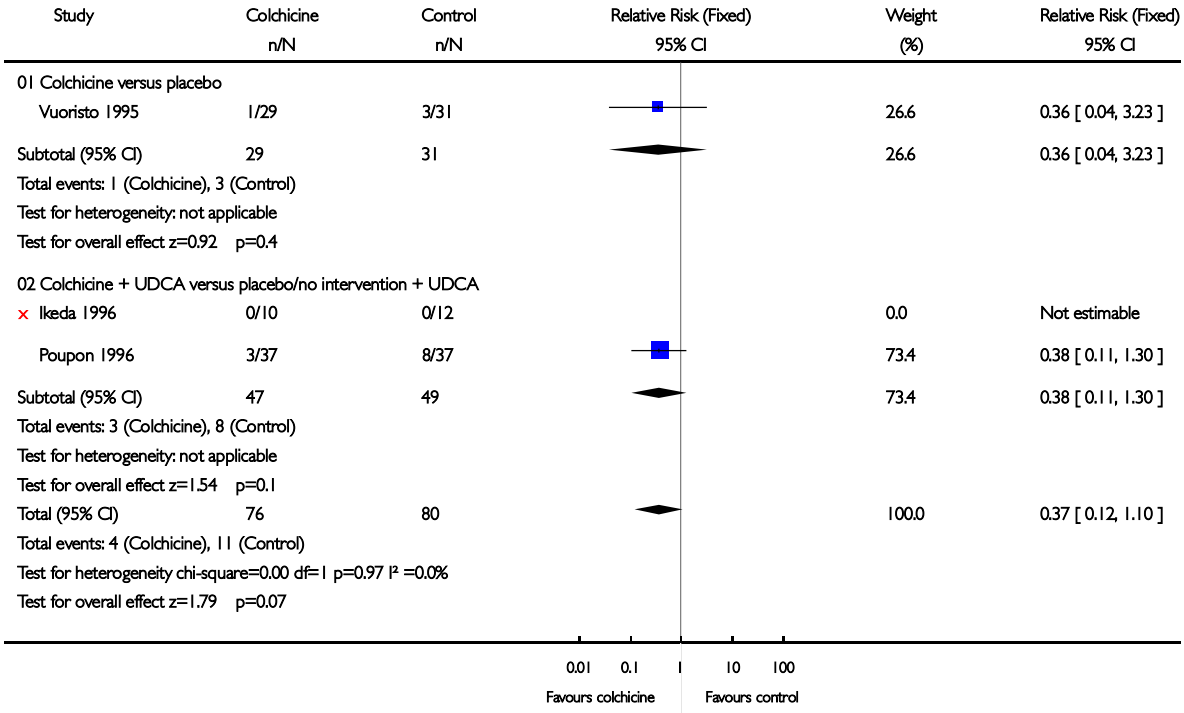


Analysis 01.06. Comparison 01 Colchicine versus placebo/no intervention, Outcome 06 Number of patients developing liver complications

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 06 Number of patients developing liver complications



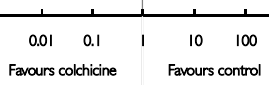
Analysis 01.07. Comparison 01 Colchicine versus placebo/no intervention, Outcome 07 Appearence of liver complications

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 07 Appearence of liver complications

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Development of varices					
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Poupon 1996	2/35	6/36		71.0	0.34 [0.07, 1.59]
Vuoristo 1995	0/29	2/31		29.0	0.21 [0.01, 4.26]
Subtotal (95% CI)	74	79		100.0	0.31 [0.08, 1.19]
Total events: 2 (Colchicine), 8 (Control)					
Test for heterogeneity chi-square=0.08 df=1 p=0.78 I^2 =0.0%					
Test for overall effect z=1.71 p=0.09					
02 Gastrointestinal bleeding					
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Poupon 1996	1/37	2/37		100.0	0.50 [0.05, 5.28]
Subtotal (95% CI)	47	49		100.0	0.50 [0.05, 5.28]
Total events: 1 (Colchicine), 2 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=0.58 p=0.6					
03 Development of ascites					
× Ikeda 1996	0/10	0/12		0.0	Not estimable
× Vuoristo 1995	0/29	0/31		0.0	Not estimable
Subtotal (95% CI)	39	43		0.0	Not estimable
Total events: 0 (Colchicine), 0 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect: not applicable					
04 Hepatic encephalopathy					
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Vuoristo 1995	1/29	1/31		100.0	1.07 [0.07, 16.31]
Subtotal (95% CI)	39	43		100.0	1.07 [0.07, 16.31]
Total events: 1 (Colchicine), 1 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=0.05 p=1					

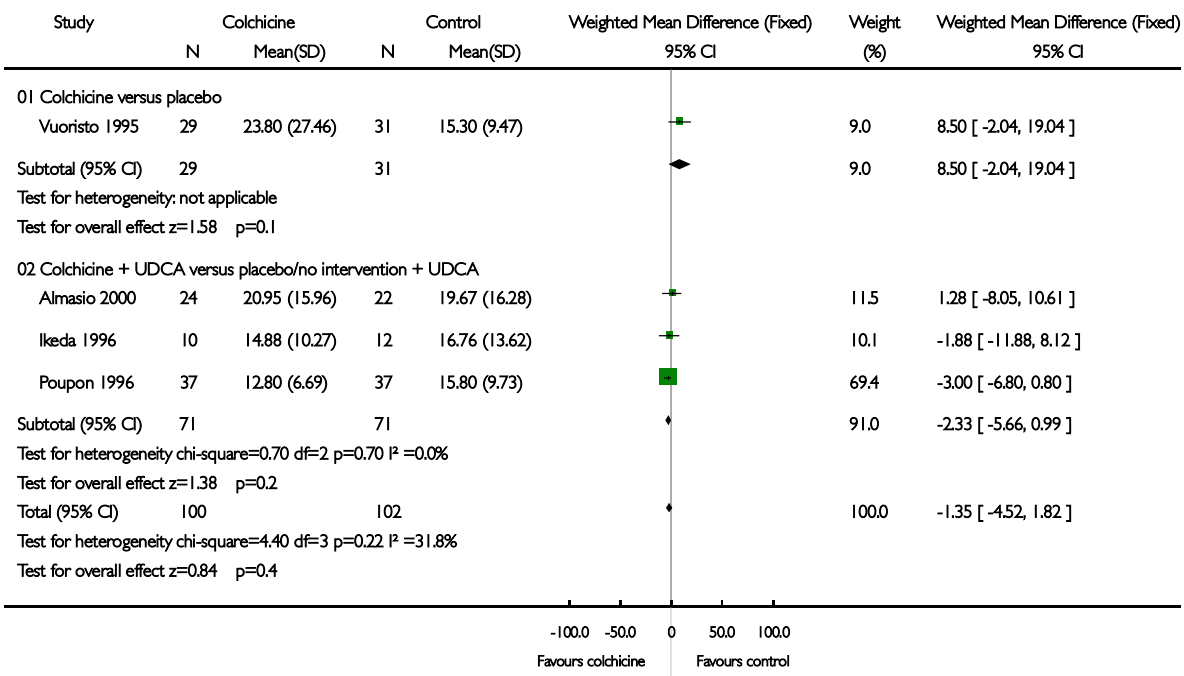


Analysis 01.08. Comparison 01 Colchicine versus placebo/no intervention, Outcome 08 S-bilirubin (ømol/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 08 S-bilirubin (µmol/L)

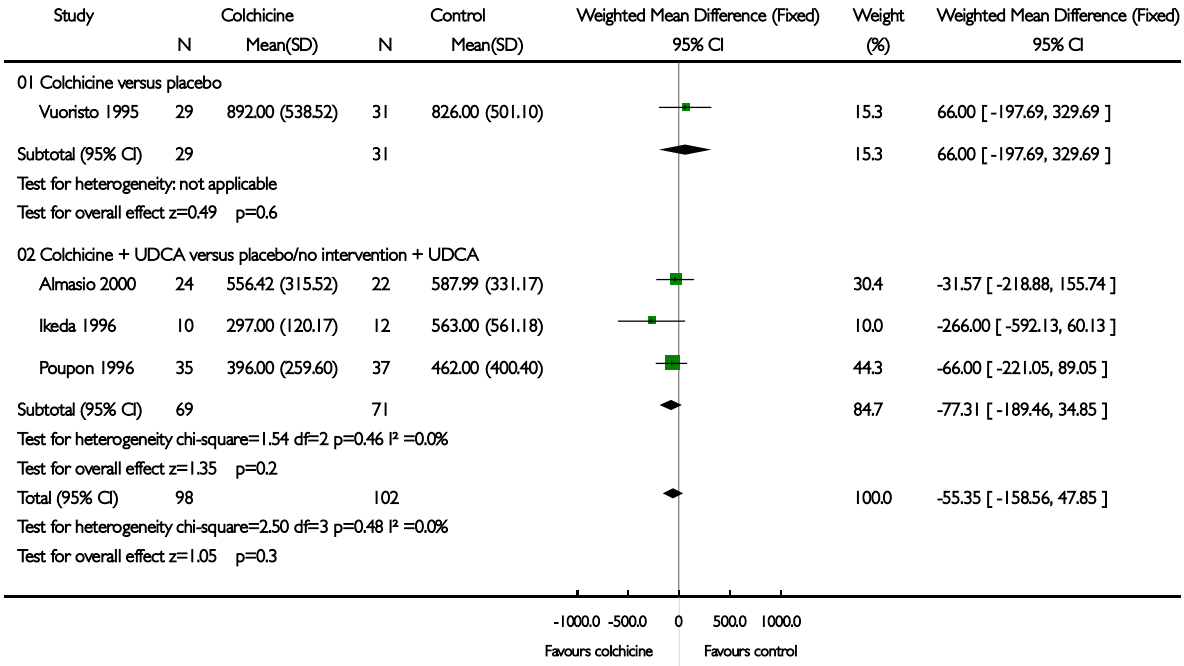


Analysis 01.09. Comparison 01 Colchicine versus placebo/no intervention, Outcome 09 S-alkaline phosphatases (ALP)(IU/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 09 S-alkaline phosphatases (ALP)(IU/L)



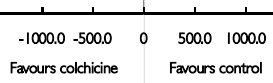
Analysis 01.10. Comparison 01 Colchicine versus placebo/no intervention, Outcome 10 S-gamma-glutamyltransferase (GGT)(IU/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 10 S-gamma-glutamyltransferase (GGT)(IU/L)

Study	Colchicine N	Colchicine Mean(SD)	Control N	Control Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Colchicine versus placebo							
Vuoristo 1995	29	649.00 (775.46)	31	428.00 (579.05)		1.9	221.00 [-127.15, 569.15]
Subtotal (95% CI)	29		31			1.9	221.00 [-127.15, 569.15]
Test for heterogeneity: not applicable							
Test for overall effect z=1.24 p=0.2							
02 Colchicine + UDCA versus placebo/no intervention + UDCA							
Almasio 2000	25	193.55 (235.20)	21	141.75 (98.70)		22.3	51.80 [-49.60, 153.20]
Ikeda 1996	10	135.00 (88.54)	12	228.00 (173.21)		18.2	-93.00 [-205.32, 19.32]
Poupon 1996	35	128.10 (134.75)	37	170.10 (138.25)		57.6	-42.00 [-105.07, 21.07]
Subtotal (95% CI)	70		70			98.1	-30.13 [-78.47, 18.21]
Test for heterogeneity chi-square=3.85 df=2 p=0.15 I^2 =48.0%							
Test for overall effect z=1.22 p=0.2							
Total (95% CI)	99		101			100.0	-25.38 [-73.26, 22.50]
Test for heterogeneity chi-square=5.81 df=3 p=0.12 I^2 =48.4%							
Test for overall effect z=1.04 p=0.3							



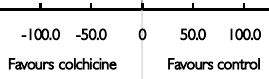
Analysis 01.11. Comparison 01 Colchicine versus placebo/no intervention, Outcome 11 S-aspartate aminotransferase (AST)(IU/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 11 S-aspartate aminotransferase (AST)(IU/L)

Study	Colchicine N	Colchicine Mean(SD)	Control N	Control Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Colchicine versus placebo							
Vuoristo 1995	29	89.00 (48.47)	31	91.00 (50.11)		26.4	-2.00 [-26.95, 22.95]
Subtotal (95% CI)	29		31			26.4	-2.00 [-26.95, 22.95]
Test for heterogeneity: not applicable							
Test for overall effect z=0.16 p=0.9							
02 Colchicine + UDCA versus placebo/no intervention + UDCA							
Ikeda 1996	10	37.00 (9.49)	12	50.00 (24.25)		73.6	-13.00 [-27.93, 1.93]
Subtotal (95% CI)	10		12			73.6	-13.00 [-27.93, 1.93]
Test for heterogeneity: not applicable							
Test for overall effect z=1.71 p=0.09							
Total (95% CI)	39		43			100.0	-10.10 [-22.91, 2.71]
Test for heterogeneity chi-square=0.55 df=1 p=0.46 I^2 =0.0%							
Test for overall effect z=1.55 p=0.1							



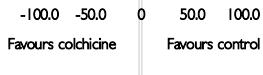
Analysis 01.12. Comparison 01 Colchicine versus placebo/no intervention, Outcome 12 S-alanine aminotransferase (ALT)(IU/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 12 S-alanine aminotransferase (ALT)(IU/L)

Study	Colchicine N	Colchicine Mean(SD)	Control N	Control Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Colchicine versus placebo							
Vuoristo 1995	29	87.00 (48.47)	31	92.00 (61.25)		5.9	-5.00 [-32.86, 22.86]
Subtotal (95% CI)	29		31			5.9	-5.00 [-32.86, 22.86]
Test for heterogeneity: not applicable							
Test for overall effect z=0.35 p=0.7							
02 Colchicine + UDCA versus placebo/no intervention + UDCA							
Almasio 2000	25	34.56 (18.00)	22	31.20 (16.80)		45.8	3.36 [-6.59, 13.31]
Ikeda 1996	10	37.00 (9.48)	12	57.00 (31.18)		13.1	-20.00 [-38.59, -1.41]
Poupon 1996	35	25.20 (29.76)	37	27.12 (17.52)		35.2	-1.92 [-13.28, 9.44]



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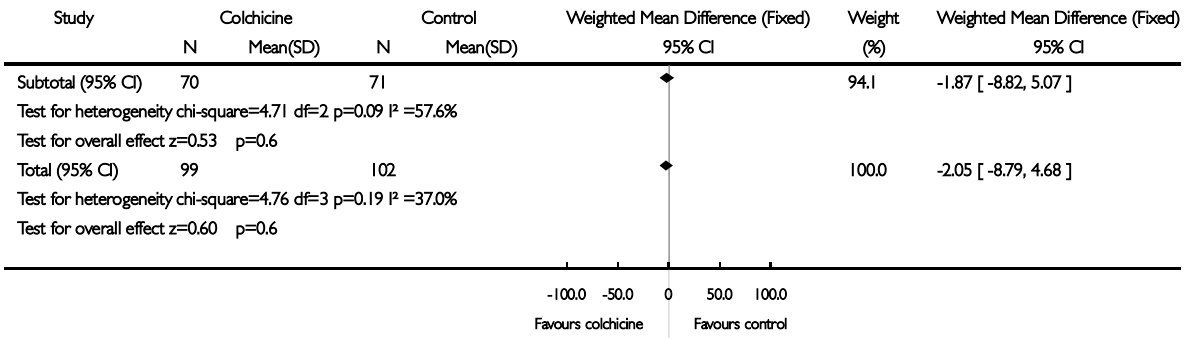
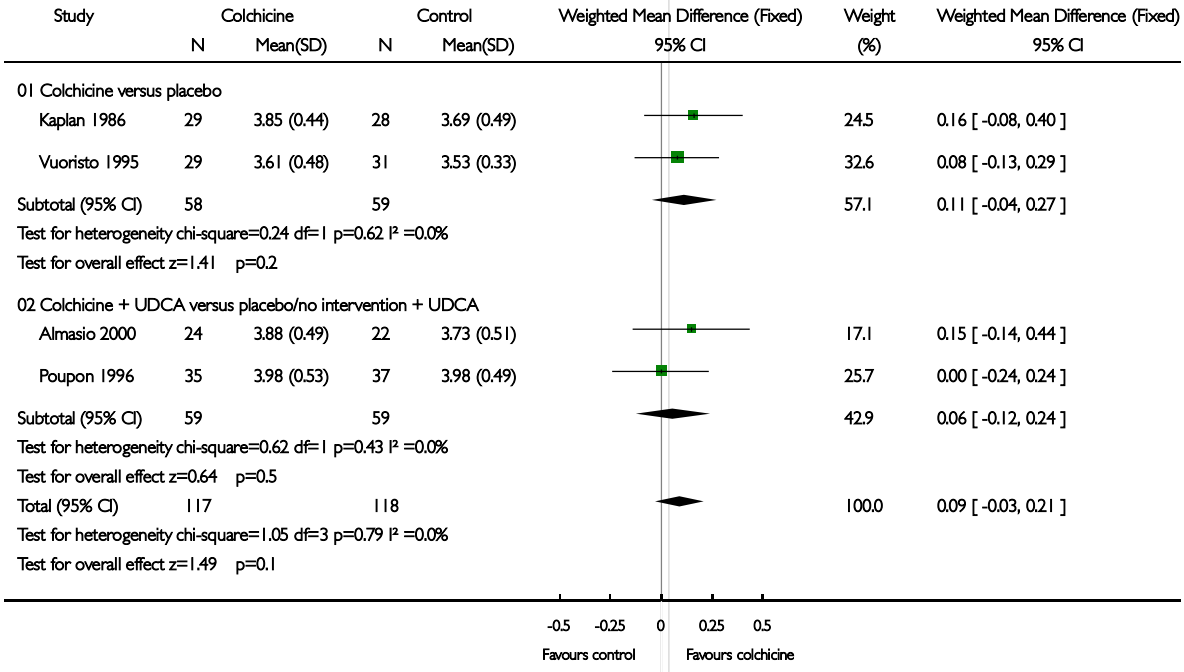


Analysis 01.13. Comparison 01 Colchicine versus placebo/no intervention, Outcome 13 S-albumin (g/dL)

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 13 S-albumin (g/dL)



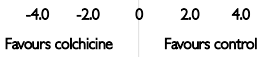
Analysis 01.14. Comparison 01 Colchicine versus placebo/no intervention, Outcome 14 S-cholesterol (total) (mmol/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 14 S-cholesterol (total) (mmol/L)

Study	Colchicine N	Colchicine Mean(SD)	Control N	Control Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Colchicine versus placebo							
Vuoristo 1995	29	6.10 (1.62)	31	6.00 (2.23)		100.0	0.10 [-0.88, 1.08]
Subtotal (95% CI)	29		31			100.0	0.10 [-0.88, 1.08]
Test for heterogeneity: not applicable							
Test for overall effect z=0.20 p=0.8							
02 Colchicine + UDCA versus placebo/no intervention + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							
Total (95% CI)	29		31			100.0	0.10 [-0.88, 1.08]
Test for heterogeneity: not applicable							
Test for overall effect z=0.20 p=0.8							



Analysis 01.15. Comparison 01 Colchicine versus placebo/no intervention, Outcome 15 Plasma immunoglobulin M (g/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 15 Plasma immunoglobulin M (g/L)

Study	Colchicine N	Colchicine Mean(SD)	Control N	Control Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Colchicine versus placebo							
Vuoristo 1995	29	5.30 (3.77)	31	6.80 (4.45)		6.9	-1.50 [-3.58, 0.58]
Subtotal (95% CI)	29		31			6.9	-1.50 [-3.58, 0.58]
Test for heterogeneity: not applicable							
Test for overall effect z=1.41 p=0.2							
02 Colchicine + UDCA versus placebo/no intervention + UDCA							
Almasio 2000	22	3.72 (2.27)	22	3.23 (2.27)		16.6	0.49 [-0.85, 1.83]
Ikeda 1996	10	2.23 (0.66)	12	3.15 (1.18)		48.8	-0.92 [-1.70, -0.14]
Poupon 1996	35	3.70 (2.09)	37	3.76 (2.40)		27.7	-0.06 [-1.10, 0.98]
Subtotal (95% CI)	67		71			93.1	-0.41 [-0.98, 0.15]

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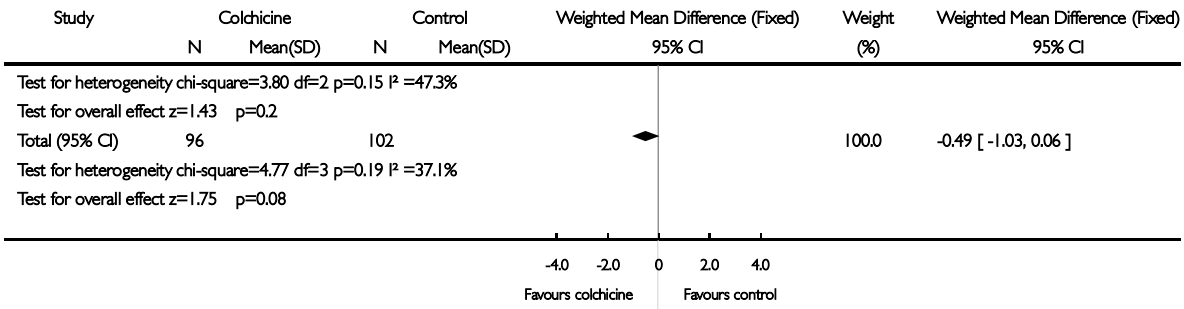
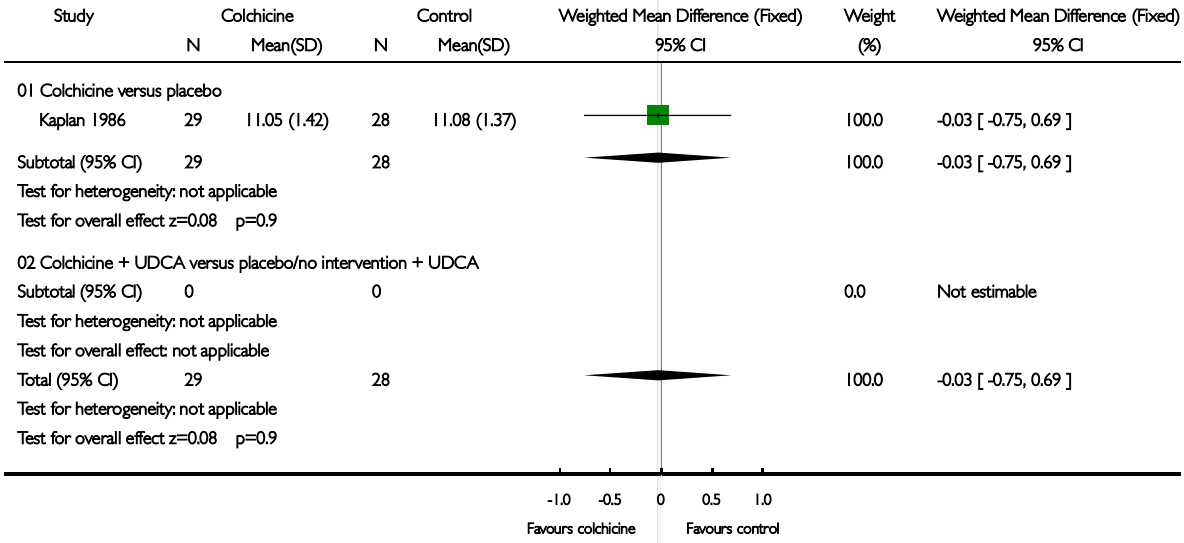


Analysis 01.16. Comparison 01 Colchicine versus placebo/no intervention, Outcome 16 Prothrombin time (second)

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 16 Prothrombin time (second)

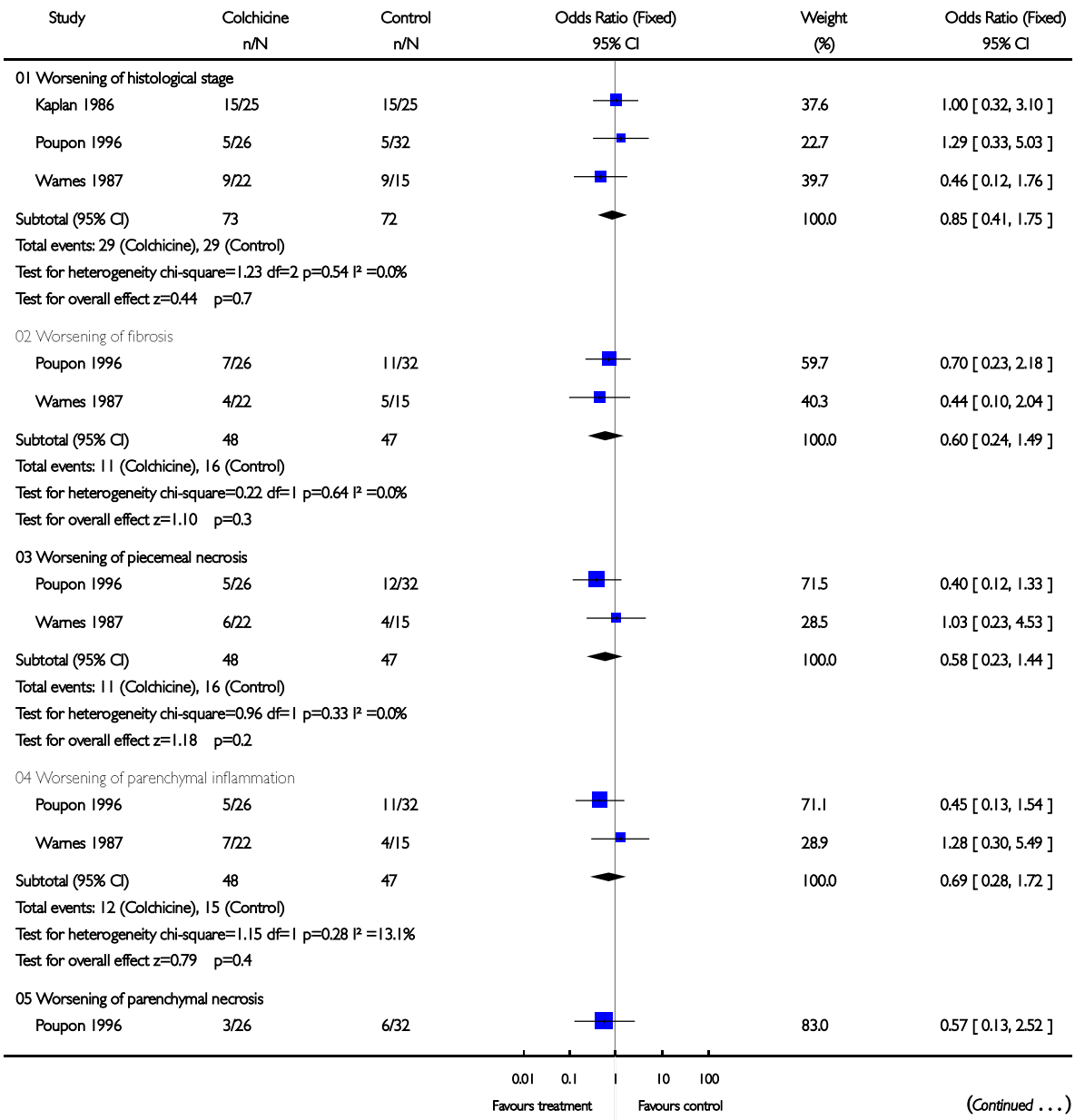


Analysis 01.17. Comparison 01 Colchicine versus placebo/no intervention, Outcome 17 Liver biopsy findings - dichotomous variables

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 17 Liver biopsy findings - dichotomous variables



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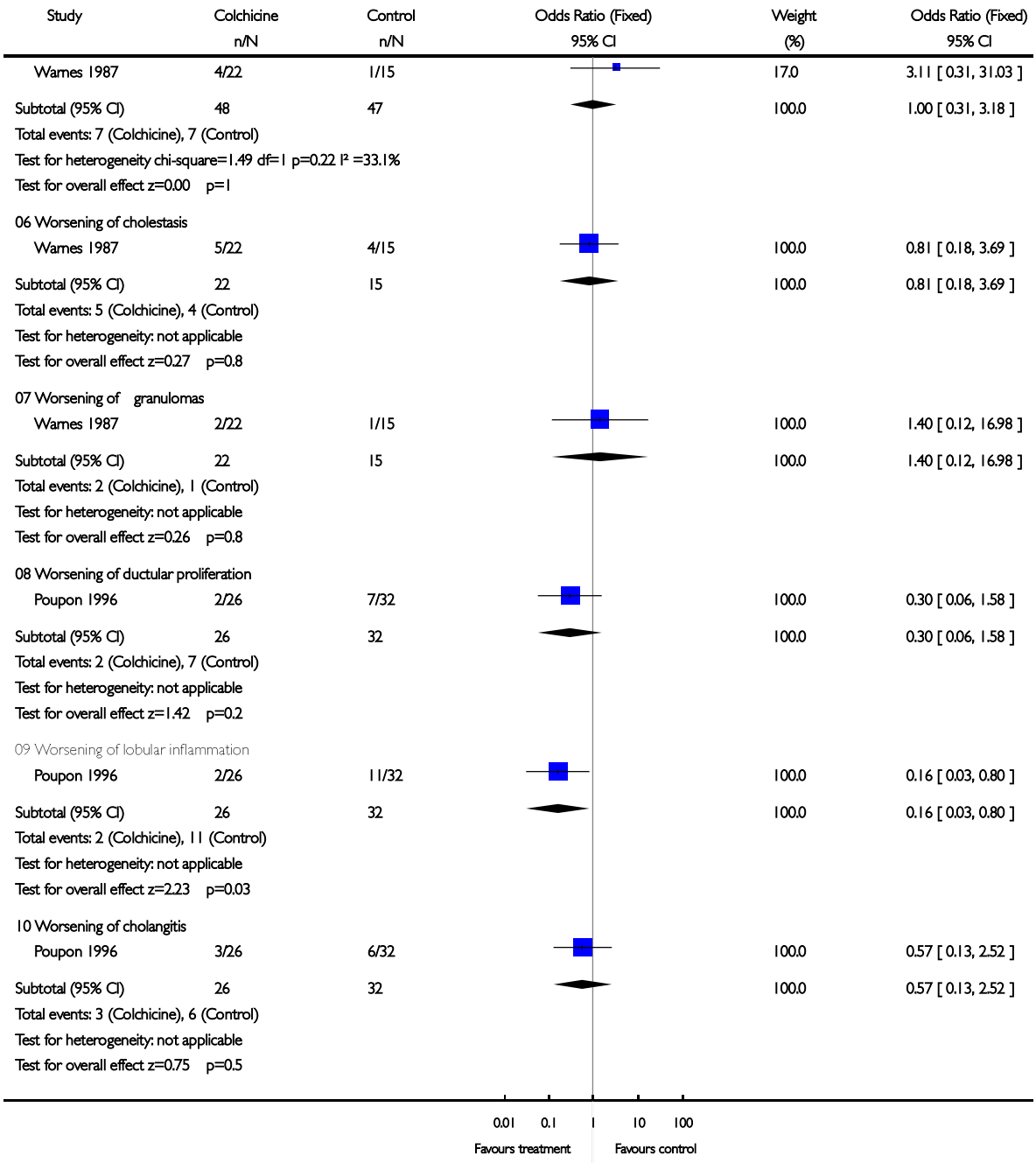


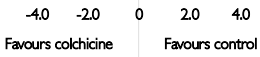
Analysis 01.18. Comparison 01 Colchicine versus placebo/no intervention, Outcome 18 Liver biopsy findings - histological score

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 18 Liver biopsy findings - histological score

Study	Colchicine N	Colchicine Mean(SD)	Control N	Control Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Colchicine versus placebo							
Kaplan 1986	25	0.76 (1.42)	25	0.20 (1.48)		100.0	0.56 [-0.24, 1.36]
Subtotal (95% CI)	25		25			100.0	0.56 [-0.24, 1.36]
Test for heterogeneity: not applicable							
Test for overall effect z=1.37 p=0.2							
02 Colchicine + UDCA versus placebo/no intervention + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							
Total (95% CI)	25		25			100.0	0.56 [-0.24, 1.36]
Test for heterogeneity: not applicable							
Test for overall effect z=1.37 p=0.2							



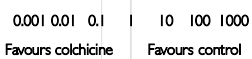
Analysis 01.19. Comparison 01 Colchicine versus placebo/no intervention, Outcome 19 Number of patients with adverse events

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 19 Number of patients with adverse events

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Colchicine versus placebo					
Bodenheimer 1988	8/28	6/29		21.6	1.38 [0.55, 3.47]
Kaplan 1986	5/30	6/30		22.0	0.83 [0.28, 2.44]
Vuoristo 1995	4/29	3/31		10.6	1.43 [0.35, 5.83]
Warnes 1987	13/34	7/30		27.3	1.64 [0.75, 3.56]
Subtotal (95% CI)	121	120		81.6	1.33 [0.82, 2.15]
Total events: 30 (Colchicine), 22 (Control)					
Test for heterogeneity chi-square=1.02 df=3 p=0.80 I^2 =0.0%					
Test for overall effect z=1.14 p=0.3					
02 Colchicine + UDCA versus placebo/no intervention + UDCA					



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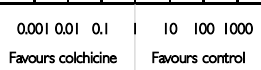
Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Almasio 2000	4/46	2/44		7.5	1.91 [0.37, 9.92]
Ikeda 1996	2/10	0/12		1.7	5.91 [0.32, 110.47]
Poupon 1996	2/37	2/37		7.3	1.00 [0.15, 6.73]
Raedsch 1993	1/14	0/14		1.8	3.00 [0.13, 67.91]
Subtotal (95% CI)	107	107		18.4	2.02 [0.72, 5.69]
Total events: 9 (Colchicine), 4 (Control)					
Test for heterogeneity chi-square=1.11 df=3 p=0.78 I^2 =0.0%					
Test for overall effect z=1.34 p=0.2					
Total (95% CI)	228	227		100.0	1.45 [0.94, 2.25]
Total events: 39 (Colchicine), 26 (Control)					
Test for heterogeneity chi-square=2.48 df=7 p=0.93 I^2 =0.0%					
Test for overall effect z=1.67 p=0.1					



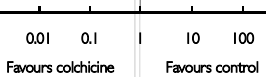
Analysis 01.20. Comparison 01 Colchicine versus placebo/no intervention, Outcome 20 Number of patients with serious adverse events

Review: Colchicine for primary biliary cirrhosis

Comparison: 01 Colchicine versus placebo/no intervention

Outcome: 20 Number of patients with serious adverse events

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Colchicine versus placebo					
× Bodenheimer 1988	0/28	0/29		0.0	Not estimable
Kaplan 1986	2/30	1/30		11.4	2.00 [0.19, 20.90]
Vuoristo 1995	2/29	2/31		22.1	1.07 [0.16, 7.10]
Warnes 1987	5/34	5/30		60.7	0.88 [0.28, 2.75]
Subtotal (95% CI)	121	120		94.3	1.06 [0.43, 2.59]
Total events: 9 (Colchicine), 8 (Control)					
Test for heterogeneity chi-square=0.38 df=2 p=0.83 I^2 =0.0%					
Test for overall effect z=0.13 p=0.9					
02 Colchicine + UDCA versus placebo/no intervention + UDCA					
× Almasio 2000	0/46	0/44		0.0	Not estimable
× Ikeda 1996	0/10	0/12		0.0	Not estimable



(*Continued* . . .)

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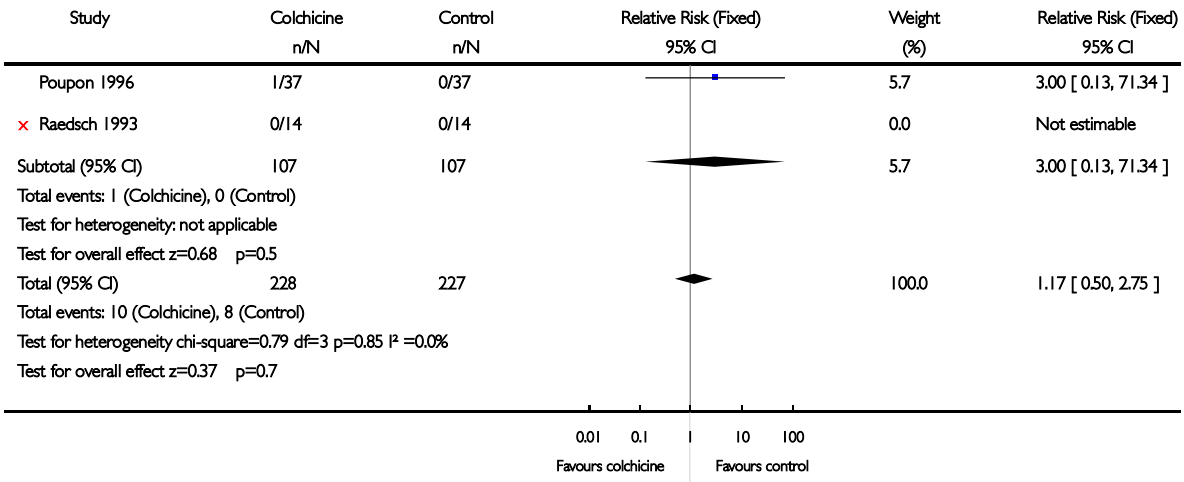
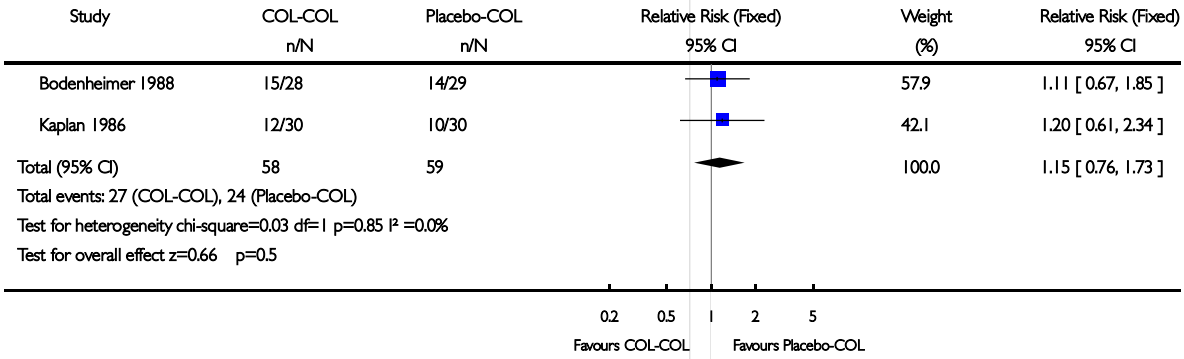


Analysis 02.01. Comparison 02 Colchcine - colchicine versus placebo - colchicine (including open label period), Outcome 01 Number of deaths

Review: Colchicine for primary biliary cirrhosis

Comparison: 02 Colchcine - colchicine versus placebo - colchicine (including open label period)

Outcome: 01 Number of deaths

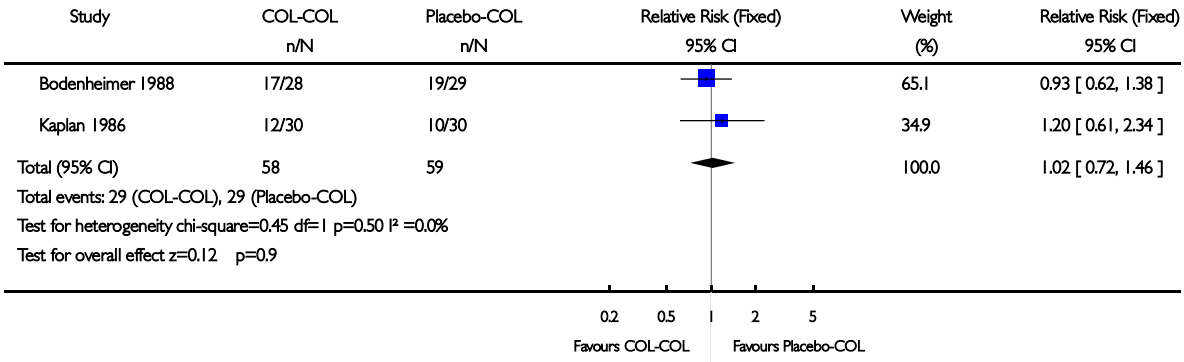


Analysis 02.02. Comparison 02 Colchcine - colchicine versus placebo - colchicine (including open label period), Outcome 02 Number of deaths and/or patients who underwent liver transplantation

Review: Colchicine for primary biliary cirrhosis

Comparison: 02 Colchcine - colchicine versus placebo - colchicine (including open label period)

Outcome: 02 Number of deaths and/or patients who underwent liver transplantation

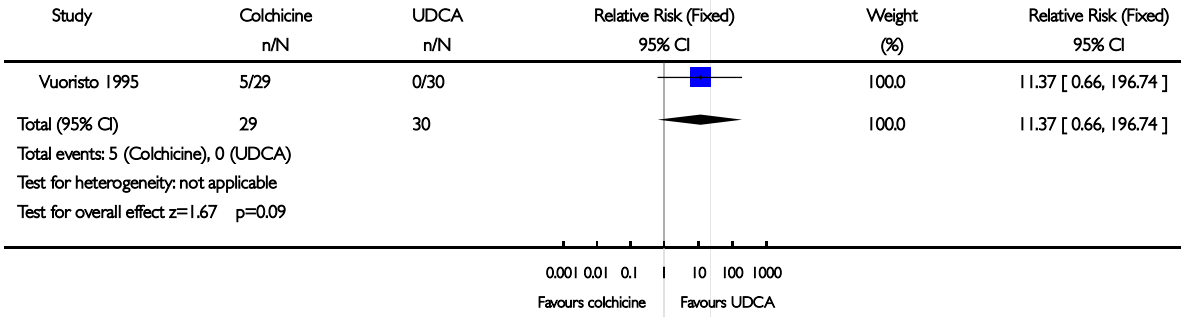


Analysis 03.01. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 01 Number of deaths

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 01 Number of deaths

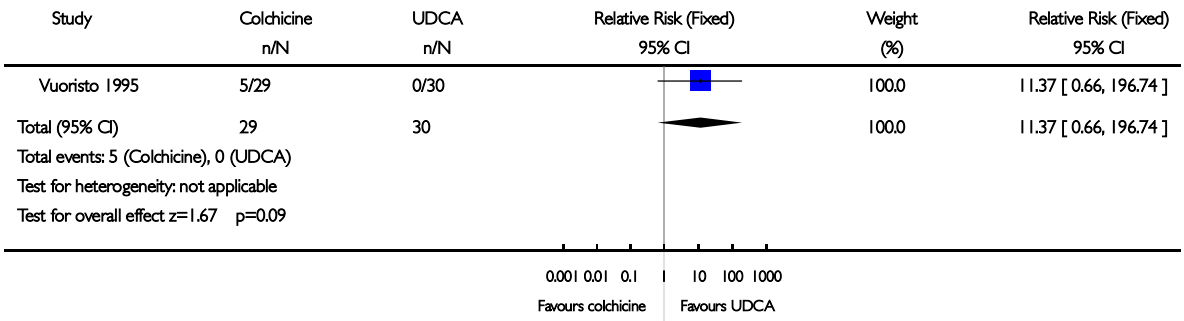


Analysis 03.02. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 02 Number of deaths and/or patients who underwent liver transplantation

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 02 Number of deaths and/or patients who underwent liver transplantation

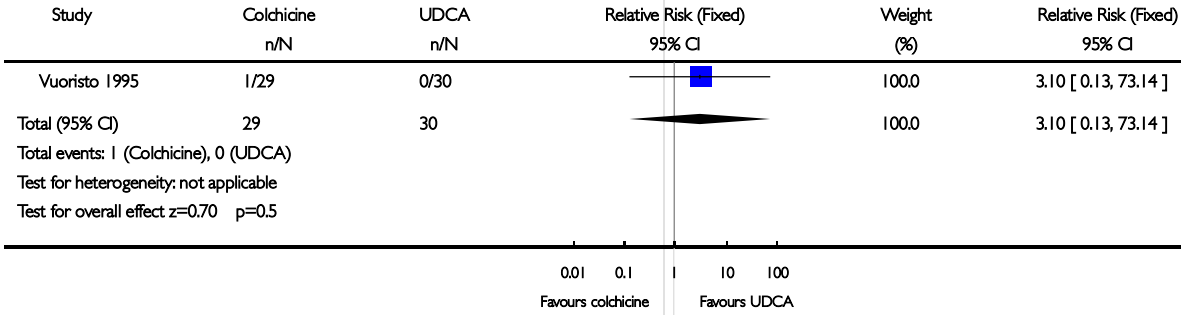


Analysis 03.03. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 03 Number of patients who underwent liver transplantation

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 03 Number of patients who underwent liver transplantation



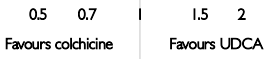
Analysis 03.04. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 04 Number of patients without improvement of pruritus

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 04 Number of patients without improvement of pruritus

Study	Colchicine n/N	UDCA n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Vuoristo 1995	18/29	24/30		100.0	0.78 [0.55, 1.09]
Total (95% CI)	29	30		100.0	0.78 [0.55, 1.09]
Total events: 18 (Colchicine), 24 (UDCA)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.48 p=0.1					



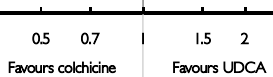
Analysis 03.05. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 05 Number of patients without improvement of fatigue

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 05 Number of patients without improvement of fatigue

Study	Colchicine n/N	UDCA n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Vuoristo 1995	24/29	30/30		100.0	0.83 [0.70, 0.98]
Total (95% CI)	29	30		100.0	0.83 [0.70, 0.98]
Total events: 24 (Colchicine), 30 (UDCA)					
Test for heterogeneity: not applicable					
Test for overall effect z=2.23 p=0.03					

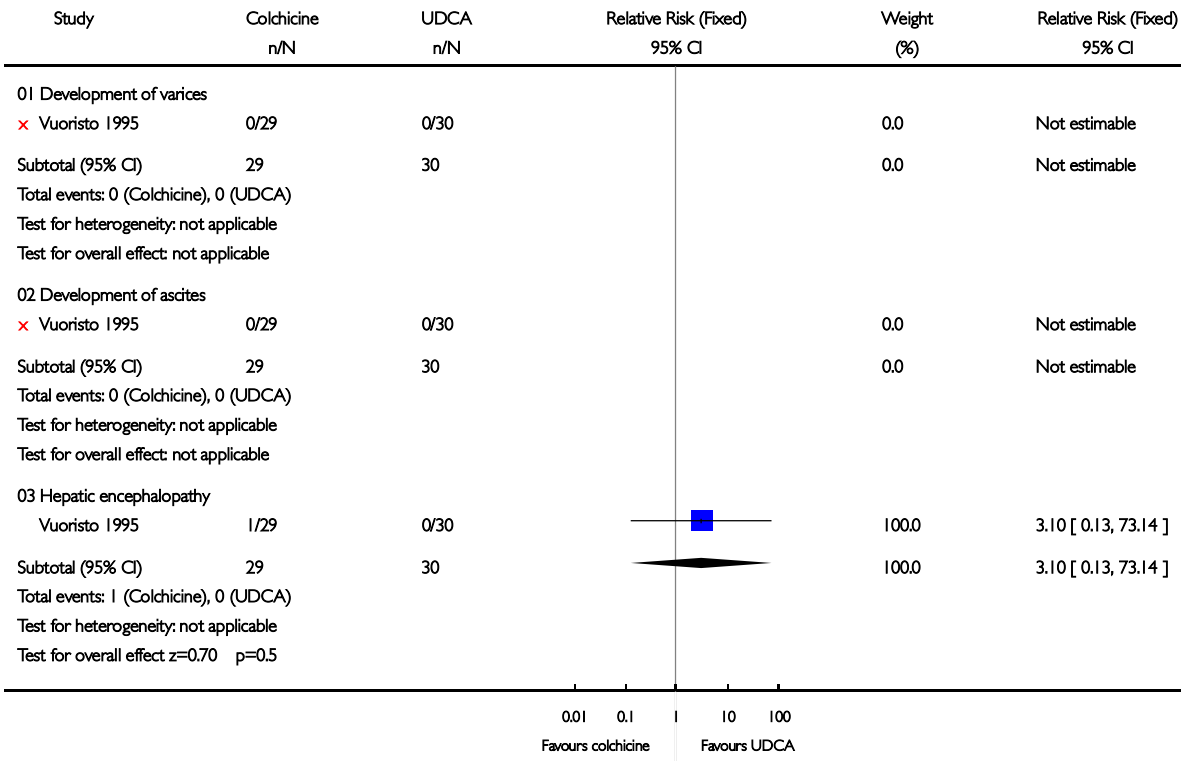


Analysis 03.06. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 06 Appearance of liver complications

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 06 Appearance of liver complications

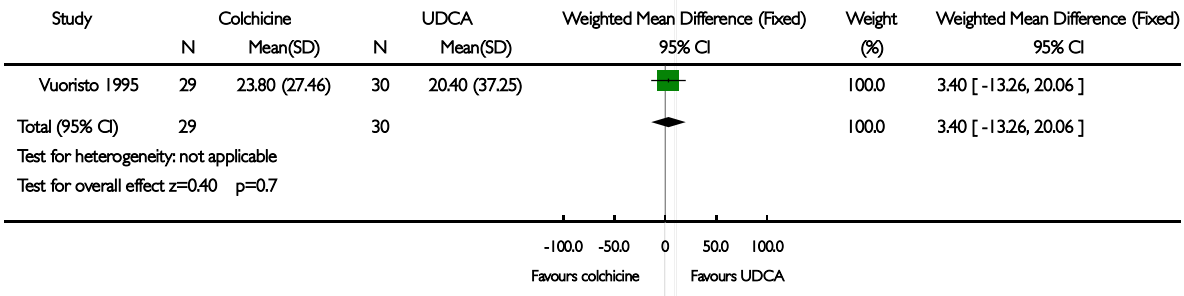


Analysis 03.07. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 07 S-bilirubin (ømol/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 07 S-bilirubin (µmol/L)

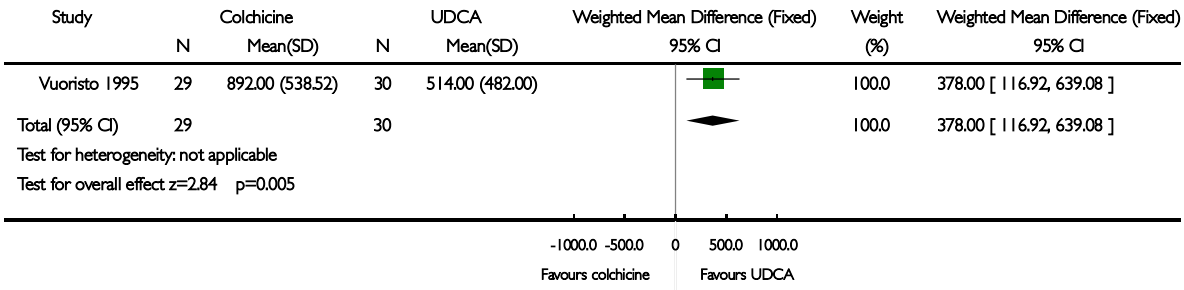


Analysis 03.08. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 08 S-alkaline phosphatases (ALP)(IU/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 08 S-alkaline phosphatases (ALP)(IU/L)

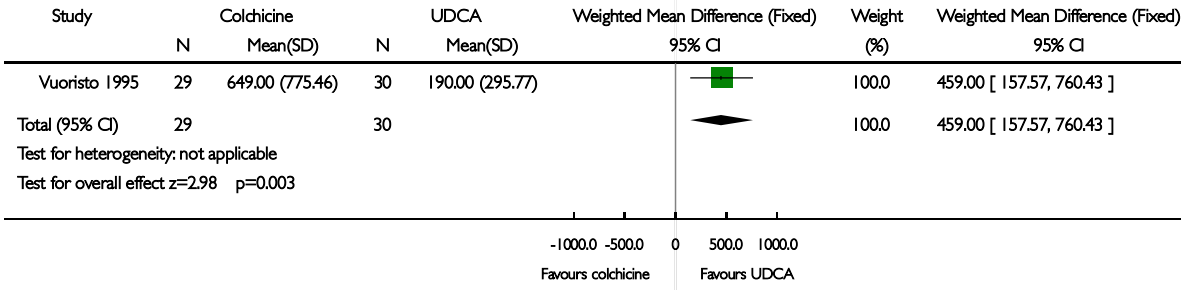


Analysis 03.09. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 09 S-gamma-glutamyltransferase (GGT)(IU/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 09 S-gamma-glutamyltransferase (GGT)(IU/L)

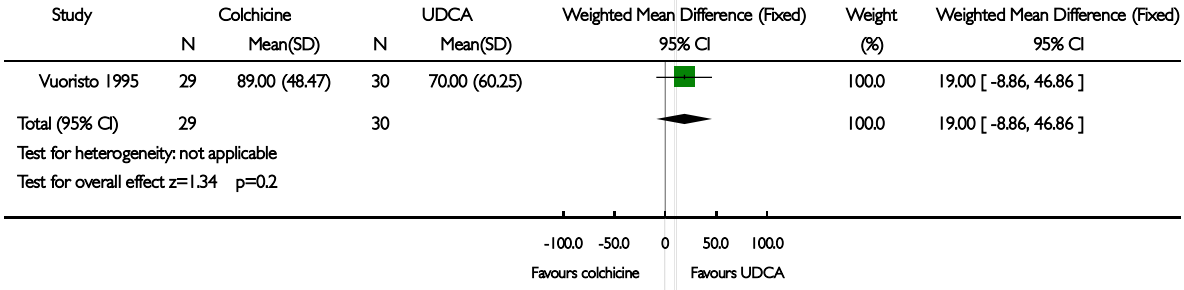


Analysis 03.10. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 10 S-aspartate aminotransferase (AST)(IU/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 10 S-aspartate aminotransferase (AST)(IU/L)

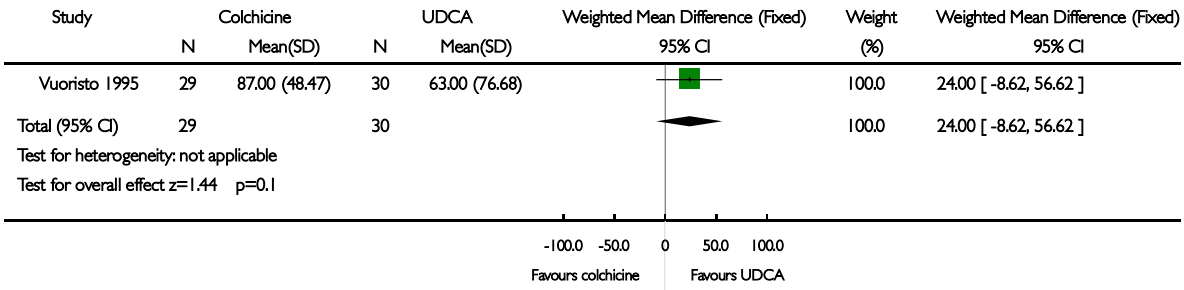


Analysis 03.11. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 11 S-alanine aminotransferase (ALT)(IU/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 11 S-alanine aminotransferase (ALT)(IU/L)

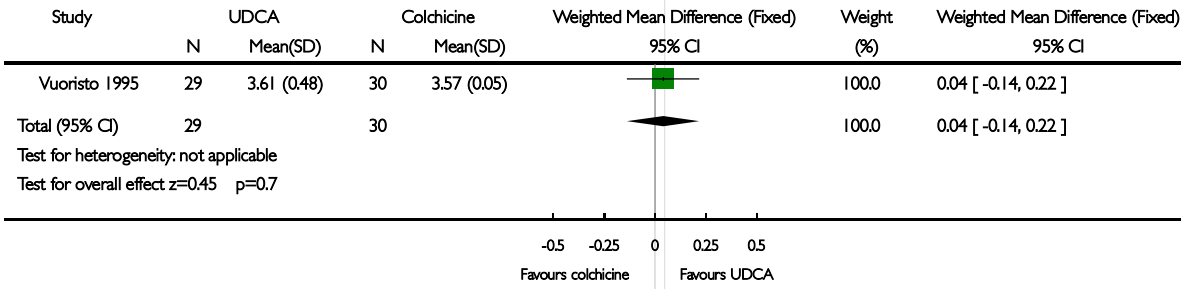


Analysis 03.12. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 12 S-albumin (g/dL)

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 12 S-albumin (g/dL)

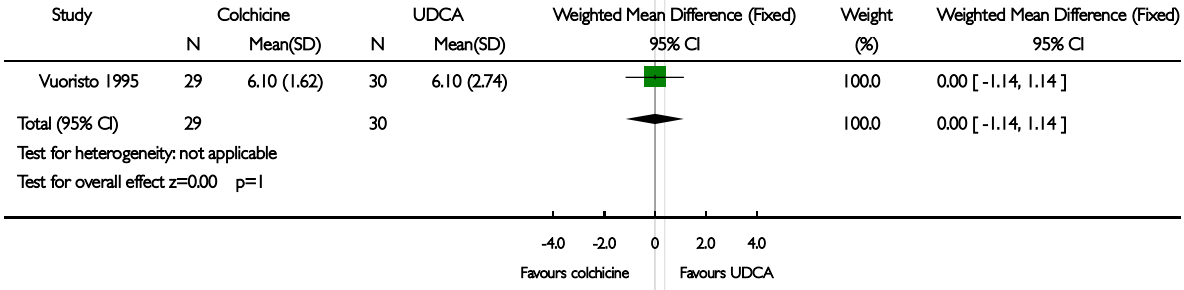


Analysis 03.13. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 13 S-cholesterol (total) (mmol/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 13 S-cholesterol (total) (mmol/L)

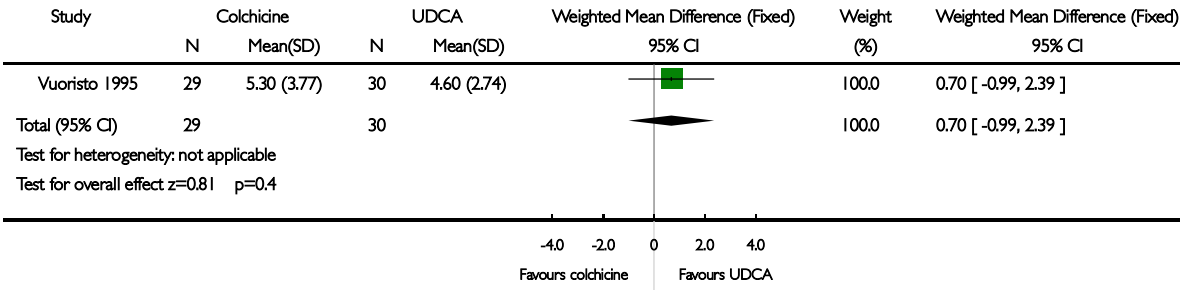


Analysis 03.14. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 14 Plasma immunoglobulin M (g/L)

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 14 Plasma immunoglobulin M (g/L)

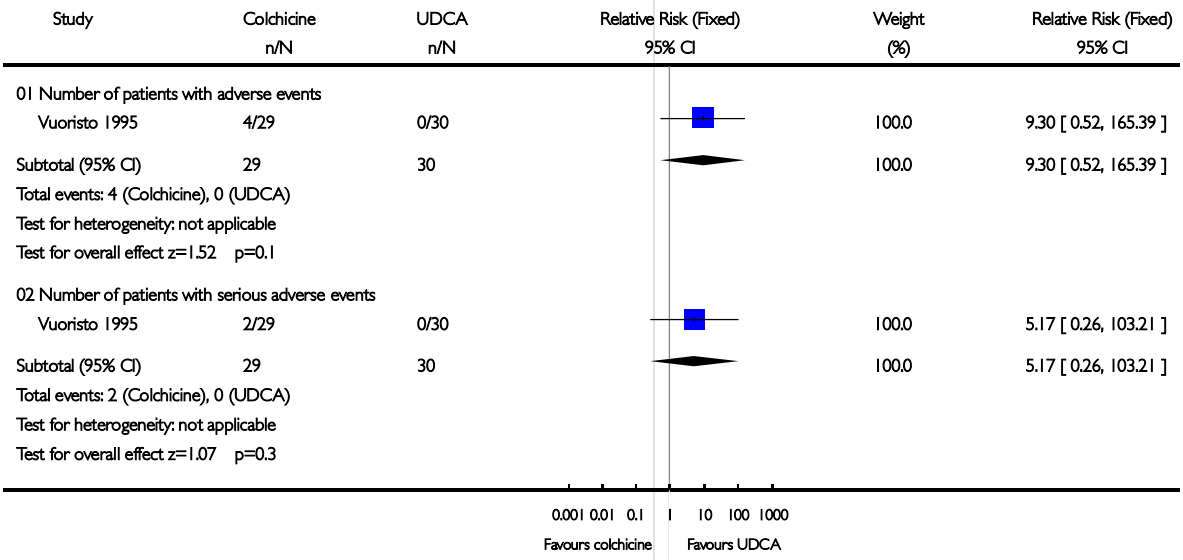


Analysis 03.15. Comparison 03 Colchicine versus ursodeoxycholic acid, Outcome 15 Number of patients with adverse events

Review: Colchicine for primary biliary cirrhosis

Comparison: 03 Colchicine versus ursodeoxycholic acid

Outcome: 15 Number of patients with adverse events

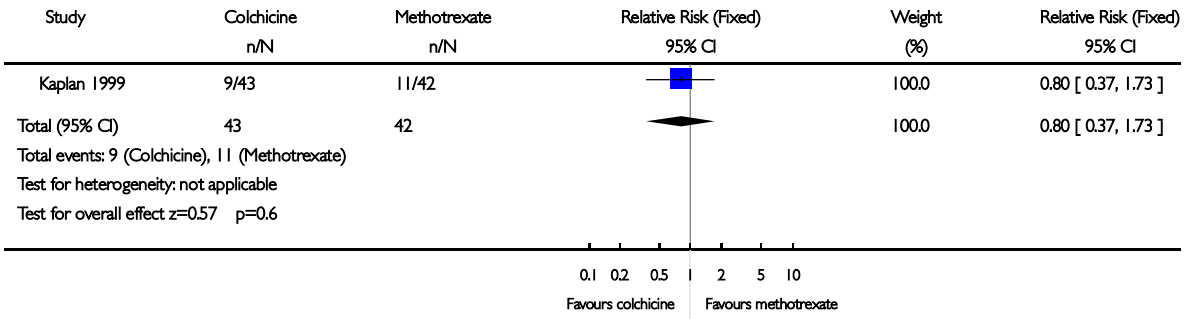


Analysis 04.01. Comparison 04 Colchicine versus methotrexate, Outcome 01 Number of deaths and/or patients who underwent liver transplantation

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 01 Number of deaths and/or patients who underwent liver transplantation

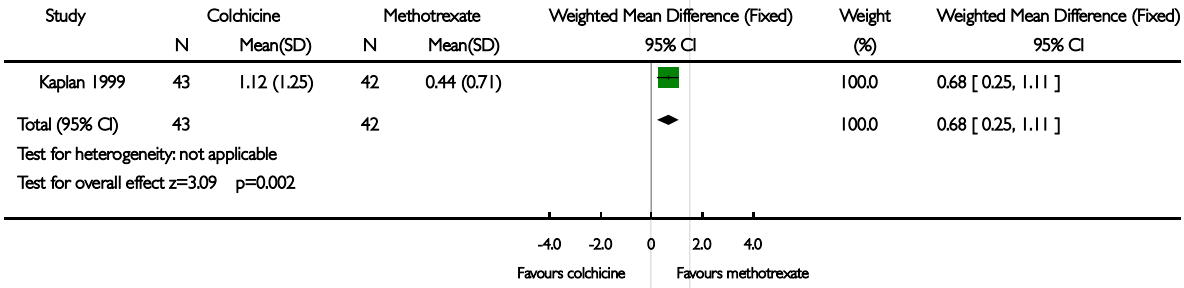


Analysis 04.02. Comparison 04 Colchicine versus methotrexate, Outcome 02 Pruritus score

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 02 Pruritus score

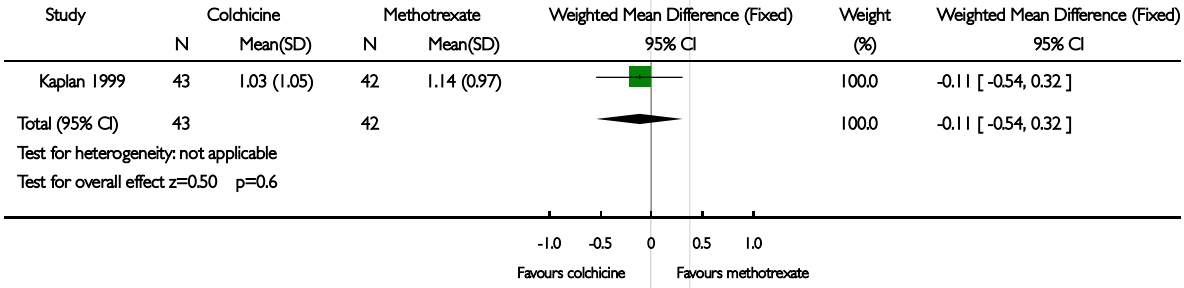


Analysis 04.03. Comparison 04 Colchicine versus methotrexate, Outcome 03 Fatigue score

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 03 Fatigue score

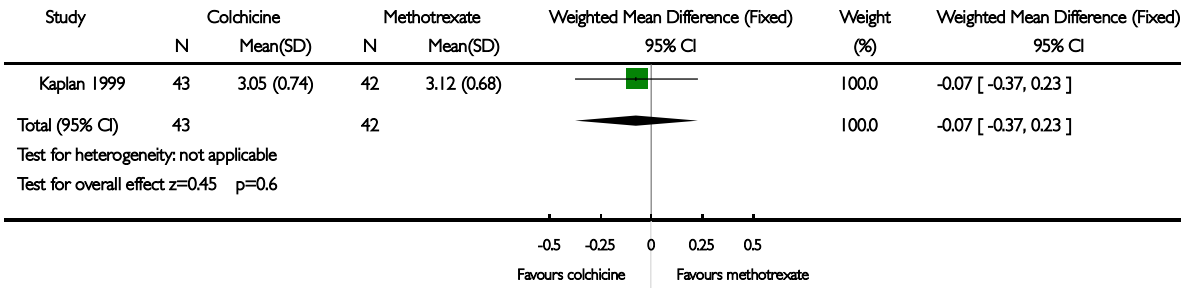


Analysis 04.04. Comparison 04 Colchicine versus methotrexate, Outcome 04 S-bilirubin (ømol/L) (presented as logtransformed geometric mean)

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 04 S-bilirubin (µmol/L) (presented as logtransformed geometric mean)

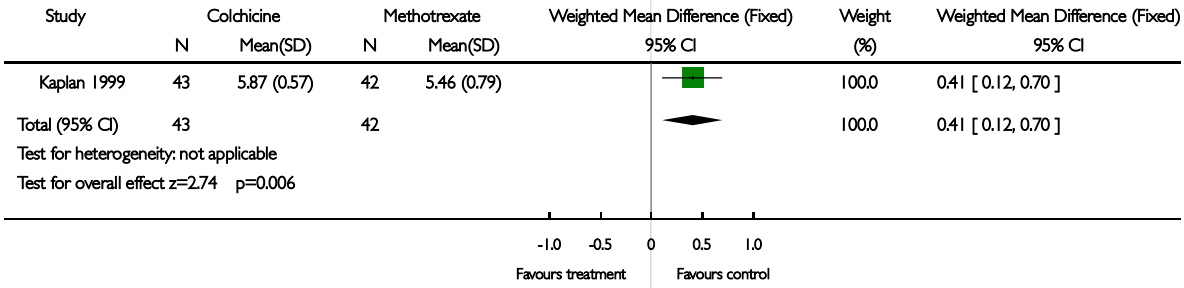


Analysis 04.05. Comparison 04 Colchicine versus methotrexate, Outcome 05 S-alkaline phosphatases (ALP)(IU/L) (presented as logtransformed geometric mean)

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 05 S-alkaline phosphatases (ALP)(IU/L) (presented as logtransformed geometric mean)

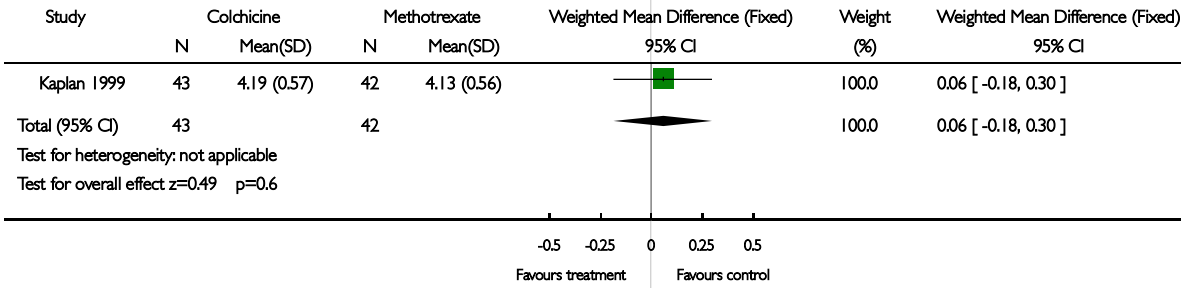


Analysis 04.06. Comparison 04 Colchicine versus methotrexate, Outcome 06 S-aspartate aminotransferase (AST)(IU/L) (presented as logtransformed geometric mean)

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 06 S-aspartate aminotransferase (AST)(IU/L) (presented as logtransformed geometric mean)

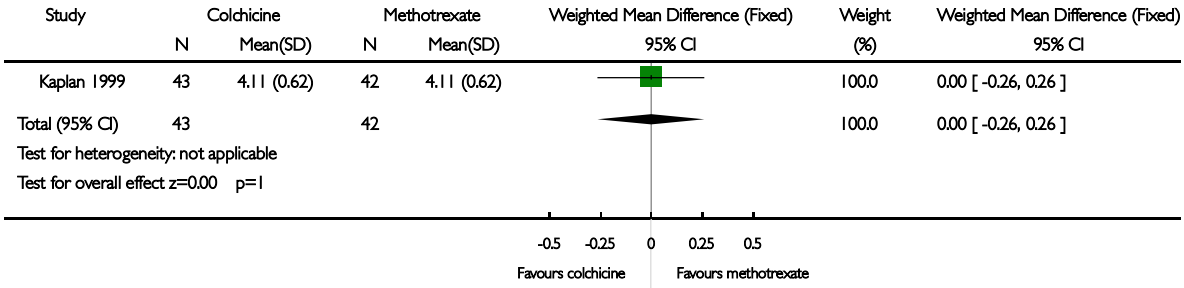


Analysis 04.07. Comparison 04 Colchicine versus methotrexate, Outcome 07 S-alanine aminotransferase (ALT)(IU/L) (presented as logtransformed geometric mean)

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 07 S-alanine aminotransferase (ALT)(IU/L) (presented as logtransformed geometric mean)

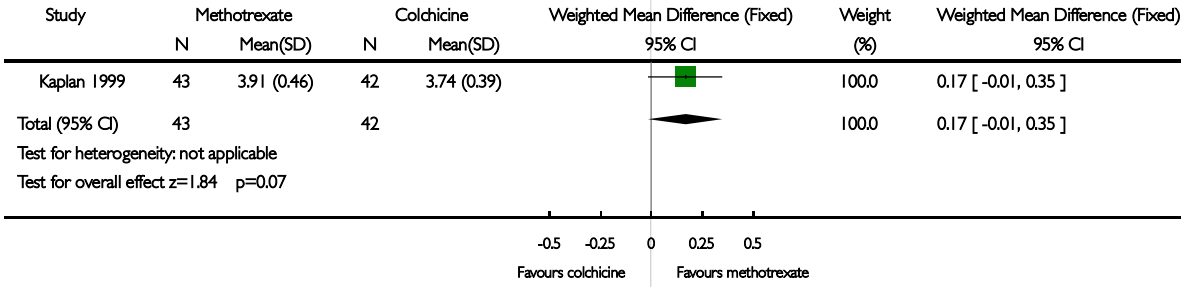


Analysis 04.08. Comparison 04 Colchicine versus methotrexate, Outcome 08 S-albumin (g/dL)

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 08 S-albumin (g/dL)

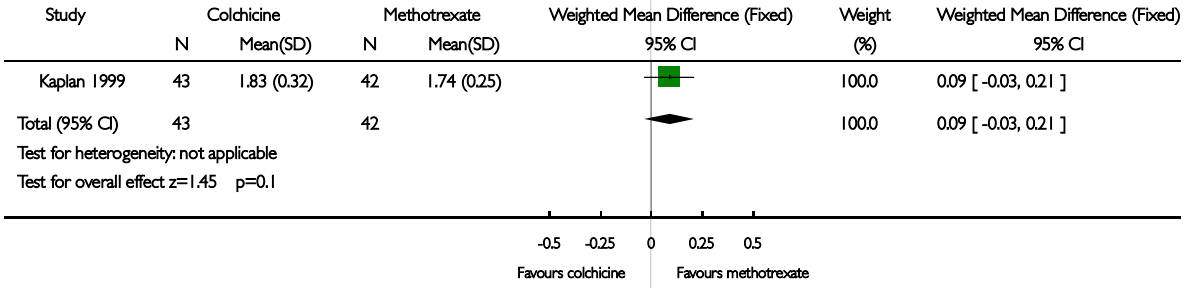


Analysis 04.09. Comparison 04 Colchicine versus methotrexate, Outcome 09 S-cholesterol (total) (mmol/L) (presented as logtransformed geometric mean)

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 09 S-cholesterol (total) (mmol/L) (presented as logtransformed geometric mean)

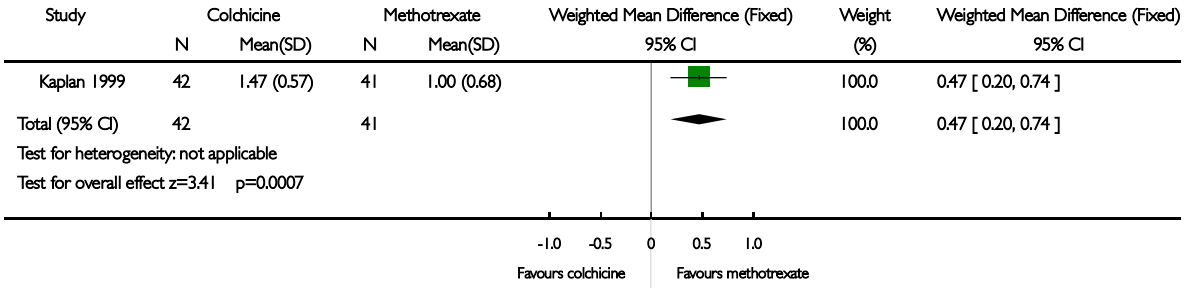


Analysis 04.10. Comparison 04 Colchicine versus methotrexate, Outcome 10 Plasma immunoglobulin M (g/L) (presented as logtransformed geometric mean)

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 10 Plasma immunoglobulin M (g/L) (presented as logtransformed geometric mean)

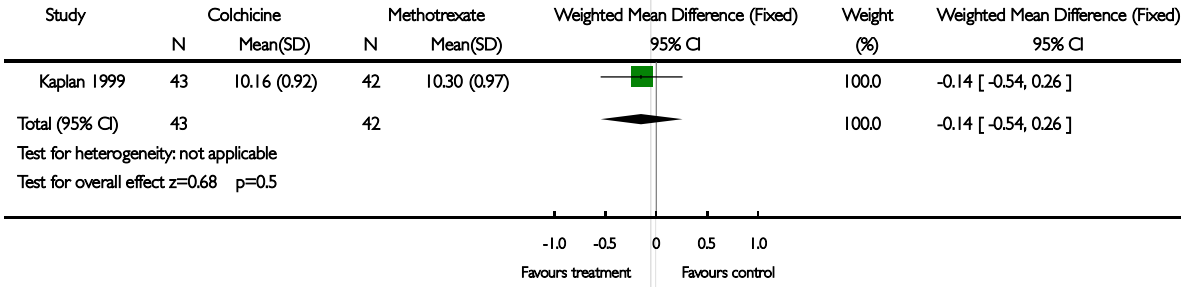


Analysis 04.11. Comparison 04 Colchicine versus methotrexate, Outcome 11 Prothrombin time (second)

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 11 Prothrombin time (second)

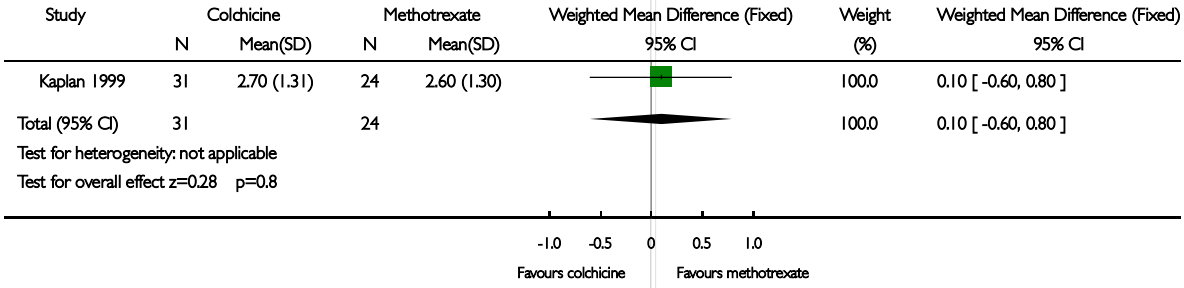


Analysis 04.12. Comparison 04 Colchicine versus methotrexate, Outcome 12 Liver biopsy findings - histological stage

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 12 Liver biopsy findings - histological stage

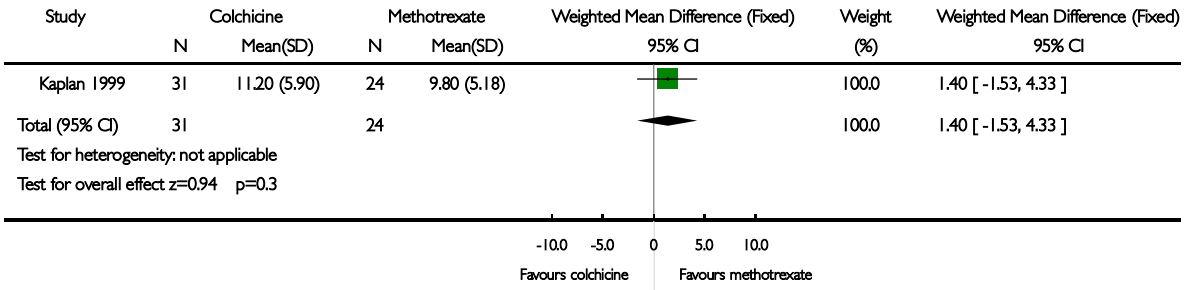


Analysis 04.13. Comparison 04 Colchicine versus methotrexate, Outcome 13 Liver biopsy findings - histological score

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 13 Liver biopsy findings - histological score

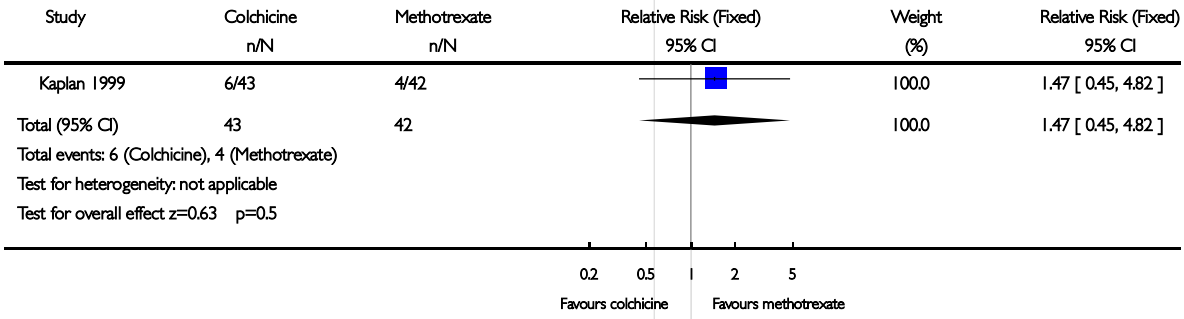


Analysis 04.14. Comparison 04 Colchicine versus methotrexate, Outcome 14 Number of patients with adverse events

Review: Colchicine for primary biliary cirrhosis

Comparison: 04 Colchicine versus methotrexate

Outcome: 14 Number of patients with adverse events



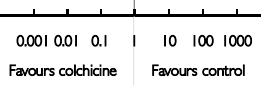
Analysis 05.01. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 01 Number of deaths - dose variation

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 01 Number of deaths - dose variation

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Colchicine - 1mg/day					
Almasio 2000	6/46	4/44		19.3	1.43 [0.43, 4.74]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Poupon 1996	2/37	0/37		2.4	5.00 [0.25, 100.72]
Raedsch 1993	2/14	0/14		2.4	5.00 [0.26, 95.61]
Vuoristo 1995	5/29	5/31		22.8	1.07 [0.34, 3.31]
Warnes 1987	5/34	5/30		25.0	0.88 [0.28, 2.75]
Subtotal (95% CI)	170	168		71.7	1.36 [0.73, 2.52]
Total events: 20 (Colchicine), 14 (Control)					
Test for heterogeneity chi-square=2.21 df=4 p=0.70 I² =0.0%					
Test for overall effect z=0.97 p=0.3					
02 Colchicine - 1.2mg/day					
Kaplan 1986	5/30	6/30		28.3	0.83 [0.28, 2.44]
Subtotal (95% CI)	30	30		28.3	0.83 [0.28, 2.44]
Total events: 5 (Colchicine), 6 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=0.33 p=0.7					
Total (95% CI)	200	198		100.0	1.21 [0.71, 2.06]
Total events: 25 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=2.63 df=5 p=0.76 I² =0.0%					
Test for overall effect z=0.70 p=0.5					



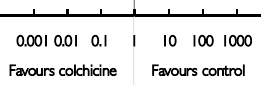
Analysis 05.02. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 02 Number of deaths - treatment duration

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 02 Number of deaths - treatment duration

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Colchicine for <=2 years duration					
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	5/30	6/30		28.3	0.83 [0.28, 2.44]
Poupon 1996	2/37	0/37		2.4	5.00 [0.25, 100.72]
Vuoristo 1995	5/29	5/31		22.8	1.07 [0.34, 3.31]
Warnes 1987	5/34	5/30		25.0	0.88 [0.28, 2.75]
Subtotal (95% CI)	140	140		78.4	1.04 [0.56, 1.93]
Total events: 17 (Colchicine), 16 (Control)					
Test for heterogeneity chi-square=1.30 df=3 p=0.73 I² =0.0%					
Test for overall effect z=0.13 p=0.9					
02 Colchicine for > 2 years duration					
Almasio 2000	6/46	4/44		19.3	1.43 [0.43, 4.74]
Raedsch 1993	2/14	0/14		2.4	5.00 [0.26, 95.61]
Subtotal (95% CI)	60	58		21.6	1.82 [0.62, 5.39]
Total events: 8 (Colchicine), 4 (Control)					
Test for heterogeneity chi-square=0.60 df=1 p=0.44 I² =0.0%					
Test for overall effect z=1.09 p=0.3					
Total (95% CI)	200	198		100.0	1.21 [0.71, 2.06]
Total events: 25 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=2.63 df=5 p=0.76 I² =0.0%					
Test for overall effect z=0.70 p=0.5					

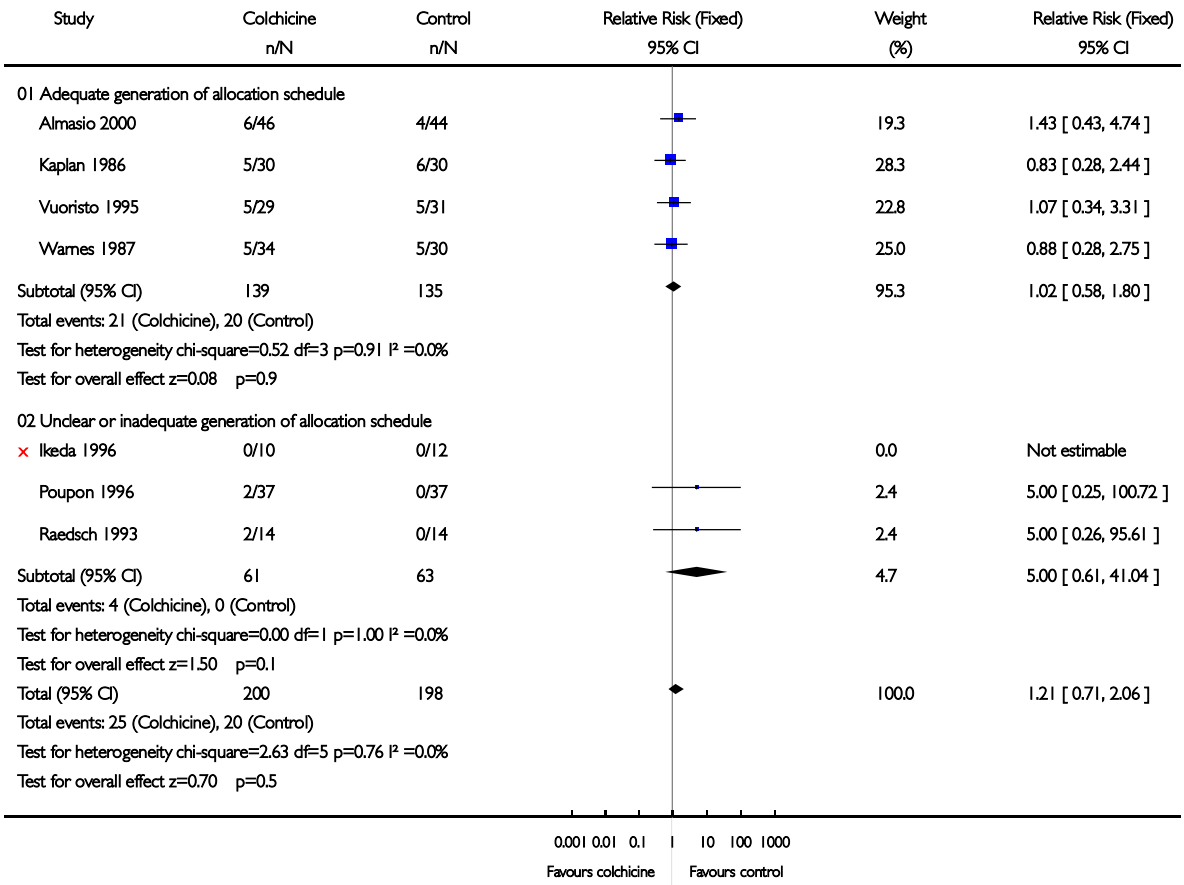


Analysis 05.03. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 03 Number of deaths - generation of the allocation sequence

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 03 Number of deaths - generation of the allocation sequence



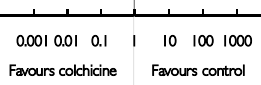
Analysis 05.04. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 04 Number of deaths - allocation concealment

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 04 Number of deaths - allocation concealment

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Adequate allocation concealment					
Almasio 2000	6/46	4/44		19.3	1.43 [0.43, 4.74]
x Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	5/30	6/30		28.3	0.83 [0.28, 2.44]
Vuoristo 1995	5/29	5/31		22.8	1.07 [0.34, 3.31]
Warnes 1987	5/34	5/30		25.0	0.88 [0.28, 2.75]
Subtotal (95% CI)	149	147		95.3	1.02 [0.58, 1.80]
Total events: 21 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=0.52 df=3 p=0.91 I² =0.0%					
Test for overall effect z=0.08 p=0.9					
02 Unclear or inadequate allocation concealment					
Poupon 1996	2/37	0/37		2.4	5.00 [0.25, 100.72]
Raedsch 1993	2/14	0/14		2.4	5.00 [0.26, 95.61]
Subtotal (95% CI)	51	51		4.7	5.00 [0.61, 41.04]
Total events: 4 (Colchicine), 0 (Control)					
Test for heterogeneity chi-square=0.00 df=1 p=1.00 I² =0.0%					
Test for overall effect z=1.50 p=0.1					
Total (95% CI)	200	198		100.0	1.21 [0.71, 2.06]
Total events: 25 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=2.63 df=5 p=0.76 I² =0.0%					
Test for overall effect z=0.70 p=0.5					



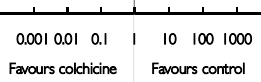
Analysis 05.05. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 05 Number of deaths - blinding

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 05 Number of deaths - blinding

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Adequate					
Almasio 2000	6/46	4/44		19.3	1.43 [0.43, 4.74]
Kaplan 1986	5/30	6/30		28.3	0.83 [0.28, 2.44]
Poupon 1996	2/37	0/37		2.4	5.00 [0.25, 100.72]
Vuoristo 1995	5/29	5/31		22.8	1.07 [0.34, 3.31]
Warnes 1987	5/34	5/30		25.0	0.88 [0.28, 2.75]
Subtotal (95% CI)	176	172		97.6	1.12 [0.65, 1.94]
Total events: 23 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=1.58 df=4 p=0.81 I² =0.0%					
Test for overall effect z=0.41 p=0.7					
02 Unclear or not perfomed					
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Raedsch 1993	2/14	0/14		2.4	5.00 [0.26, 95.61]
Subtotal (95% CI)	24	26		2.4	5.00 [0.26, 95.61]
Total events: 2 (Colchicine), 0 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.07 p=0.3					
Total (95% CI)	200	198		100.0	1.21 [0.71, 2.06]
Total events: 25 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=2.63 df=5 p=0.76 I² =0.0%					
Test for overall effect z=0.70 p=0.5					



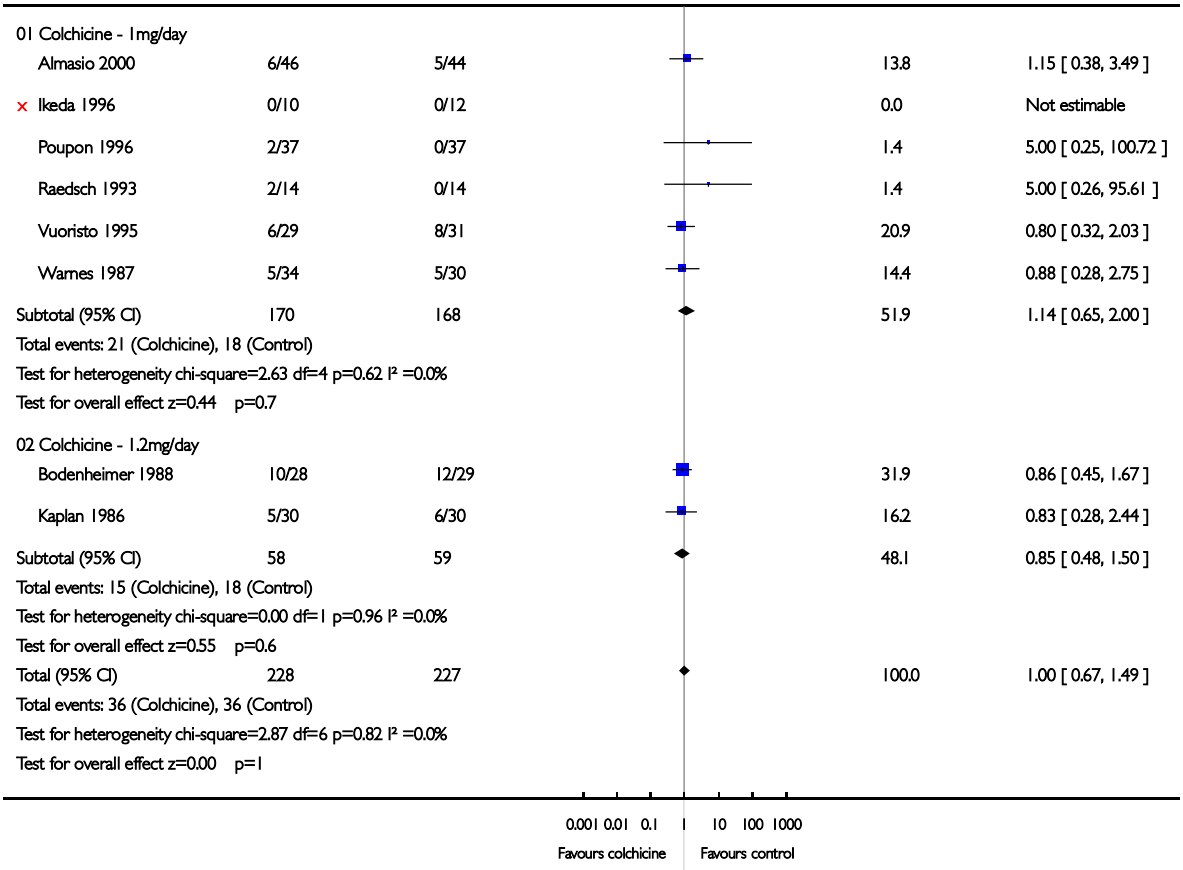
Analysis 05.06. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 06 Number of deaths and/or patients who underwent liver transplantation - dose variation

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 06 Number of deaths and/or patients who underwent liver transplantation - dose variation

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Colchicine - 1mg/day					
Almasio 2000	6/46	5/44		13.8	1.15 [0.38, 3.49]
x Ikeda 1996	0/10	0/12		0.0	Not estimable
Poupon 1996	2/37	0/37		1.4	5.00 [0.25, 100.72]
Raedsch 1993	2/14	0/14		1.4	5.00 [0.26, 95.61]
Vuoristo 1995	6/29	8/31		20.9	0.80 [0.32, 2.03]
Warnes 1987	5/34	5/30		14.4	0.88 [0.28, 2.75]
Subtotal (95% CI)	170	168		51.9	1.14 [0.65, 2.00]
Total events: 21 (Colchicine), 18 (Control)					
Test for heterogeneity chi-square=2.63 df=4 p=0.62 I² =0.0%					
Test for overall effect z=0.44 p=0.7					
02 Colchicine - 1.2mg/day					
Bodenheimer 1988	10/28	12/29		31.9	0.86 [0.45, 1.67]
Kaplan 1986	5/30	6/30		16.2	0.83 [0.28, 2.44]
Subtotal (95% CI)	58	59		48.1	0.85 [0.48, 1.50]
Total events: 15 (Colchicine), 18 (Control)					
Test for heterogeneity chi-square=0.00 df=1 p=0.96 I² =0.0%					
Test for overall effect z=0.55 p=0.6					
Total (95% CI)	228	227		100.0	1.00 [0.67, 1.49]
Total events: 36 (Colchicine), 36 (Control)					
Test for heterogeneity chi-square=2.87 df=6 p=0.82 I² =0.0%					
Test for overall effect z=0.00 p=1					

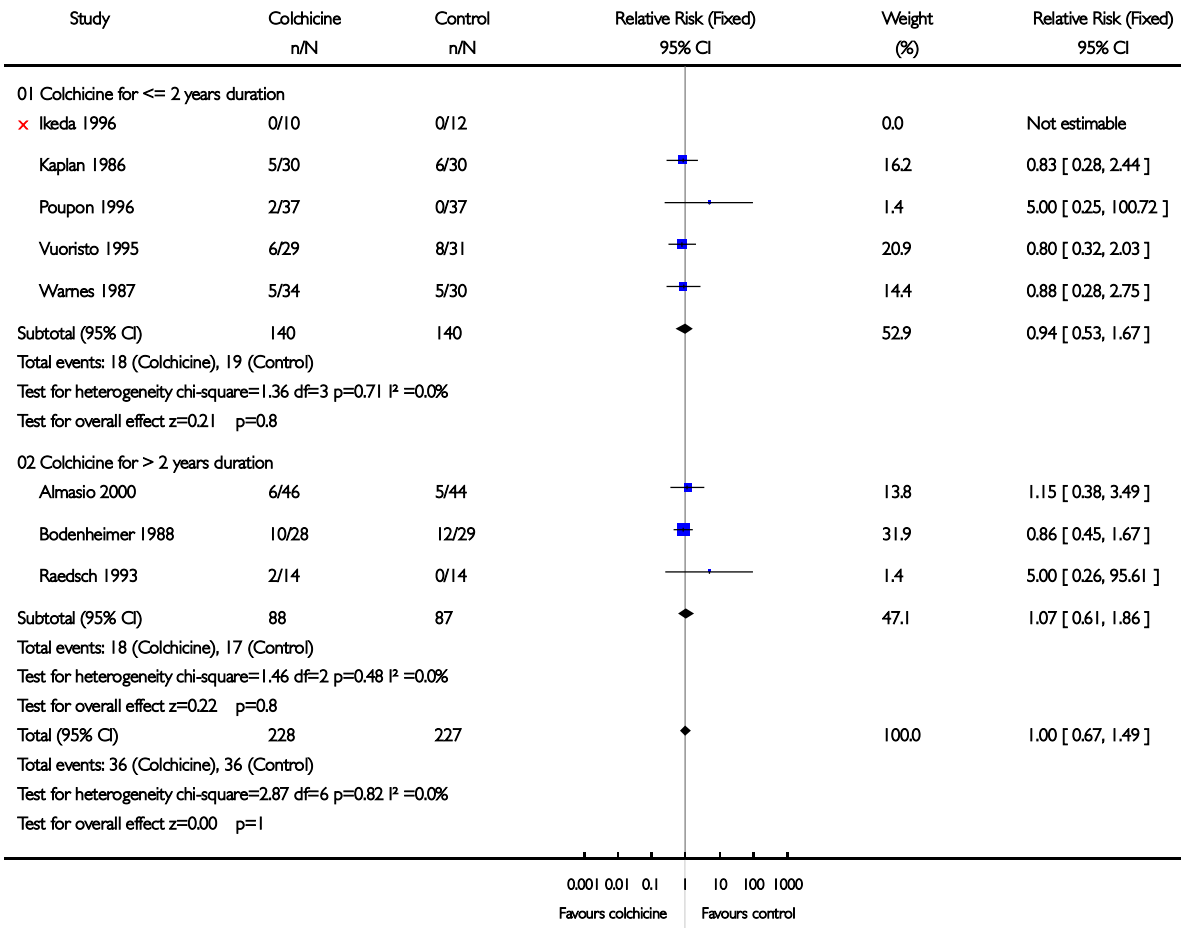


Analysis 05.07. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 07 Number of deaths and/or patients who underwent liver transplantation - treatment duration

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 07 Number of deaths and/or patients who underwent liver transplantation - treatment duration

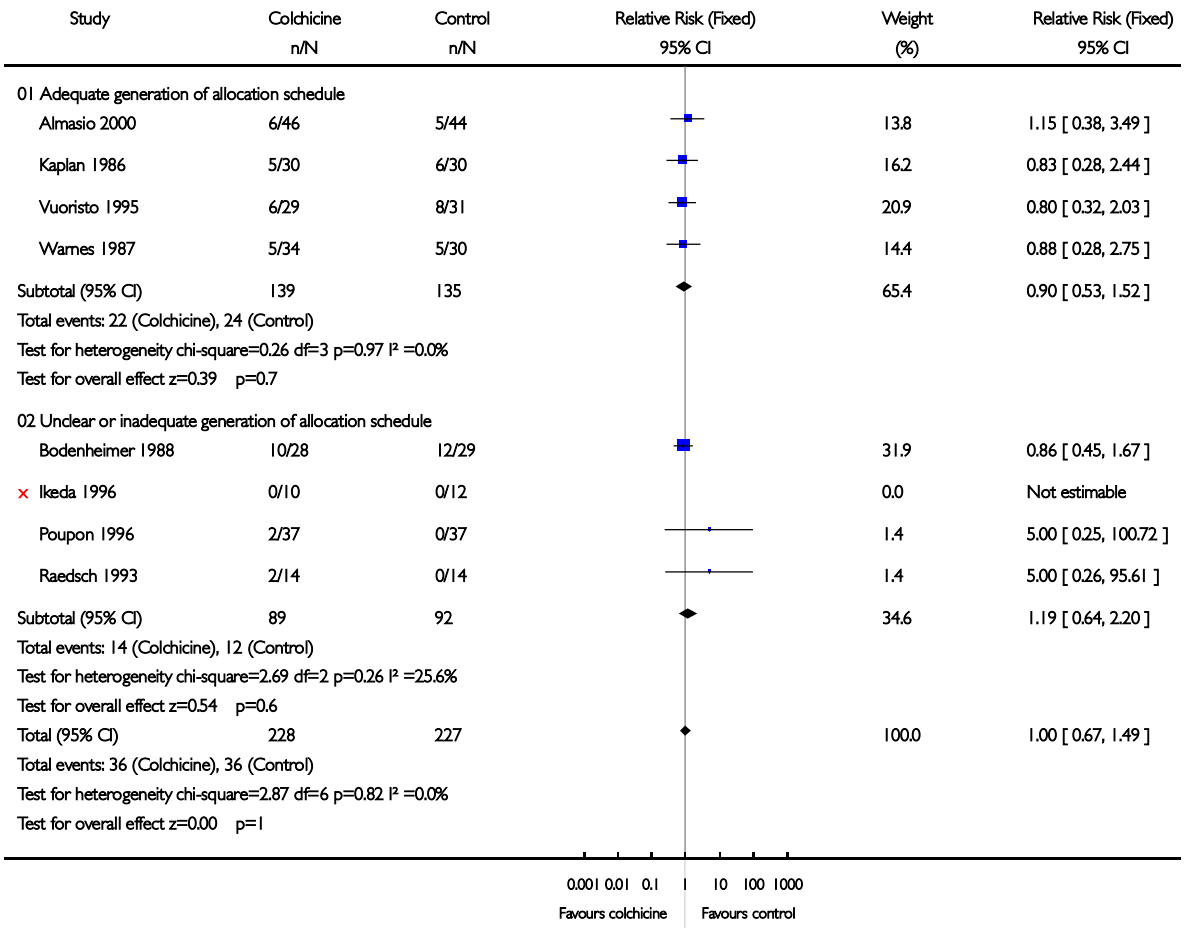


Analysis 05.08. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 08 Number of deaths and/or patients who underwent liver transplantation - generation of the allocation sequen

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 08 Number of deaths and/or patients who underwent liver transplantation - generation of the allocation sequen

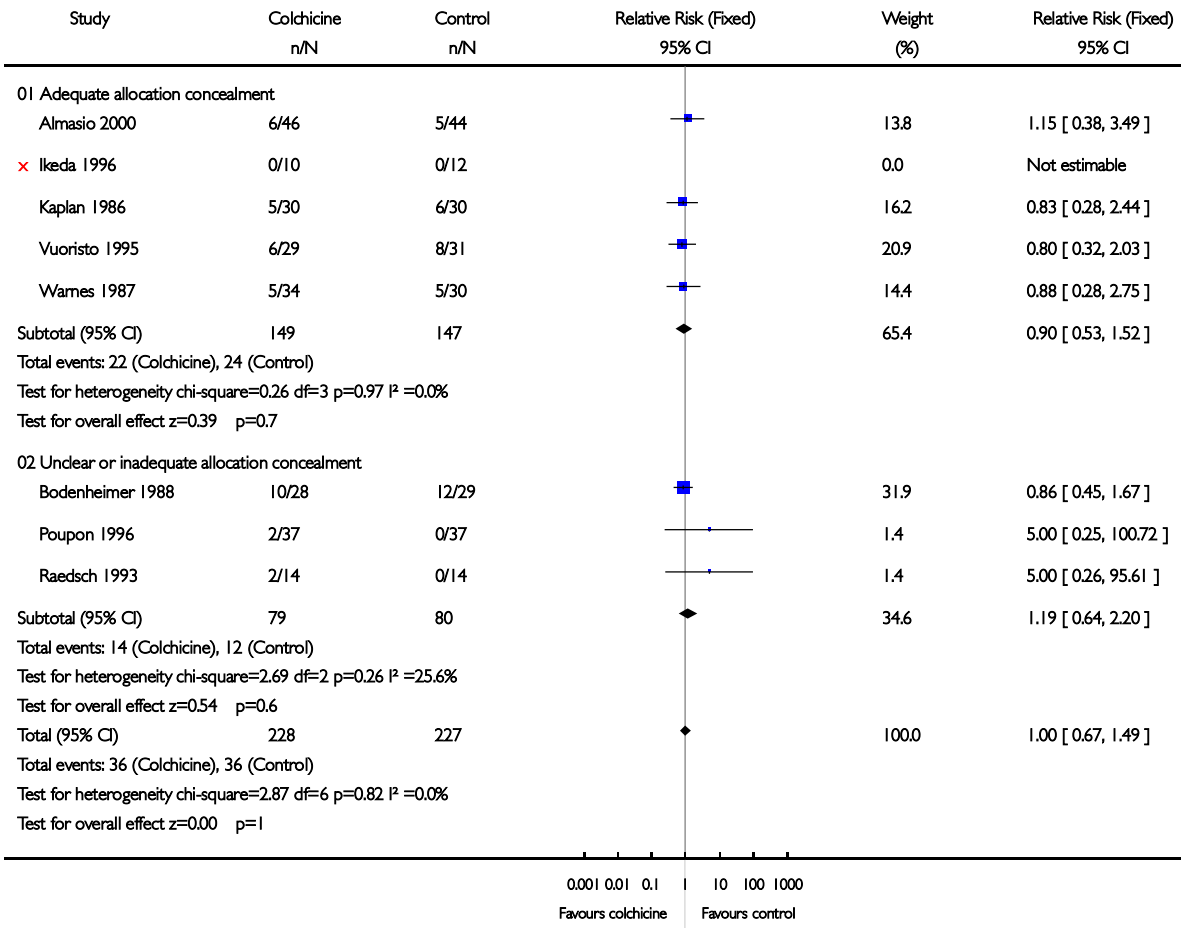


Analysis 05.09. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 09 Number of deaths and/or patients who underwent liver transplantation - allocation concealment

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 09 Number of deaths and/or patients who underwent liver transplantation - allocation concealment



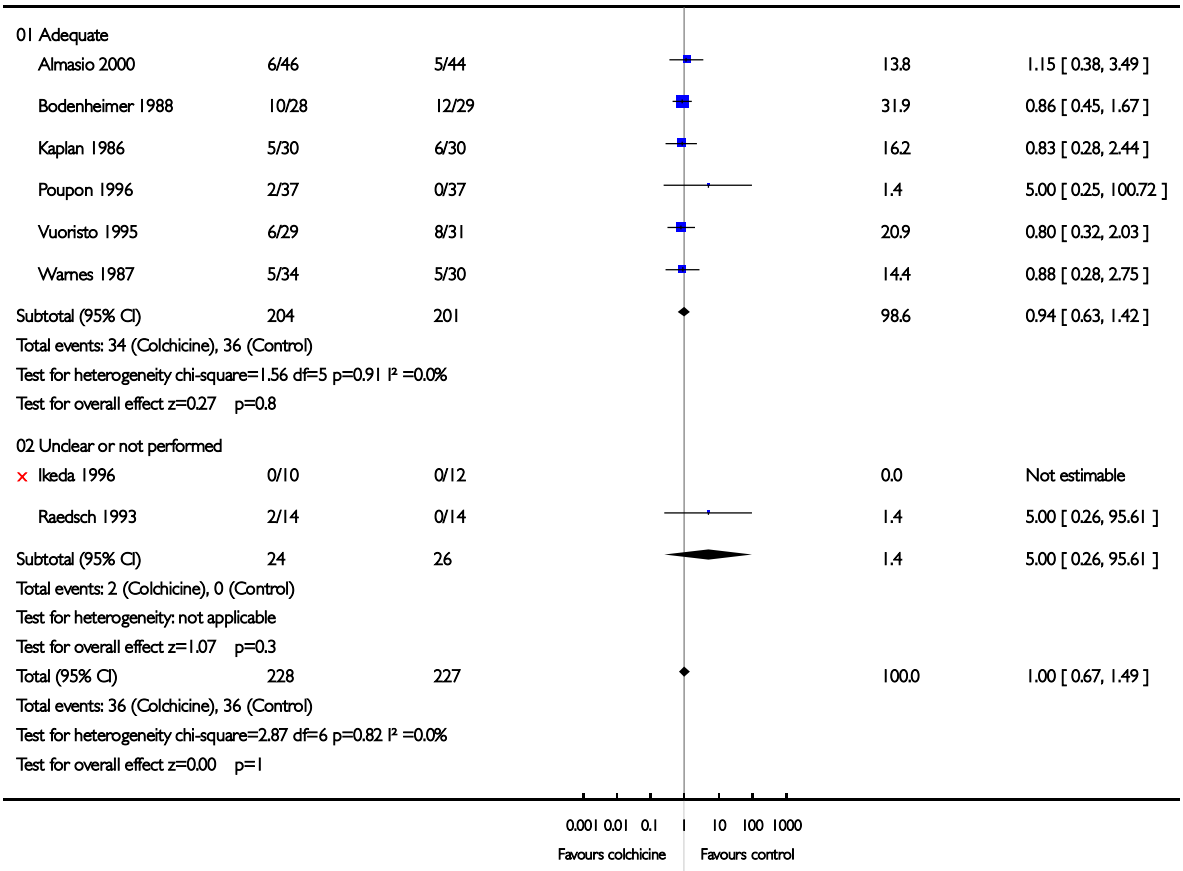
Analysis 05.10. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 10 Number of deaths and/or patients who underwent liver transplantation - blinding

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 10 Number of deaths and/or patients who underwent liver transplantation - blinding

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Adequate					
Almasio 2000	6/46	5/44		13.8	1.15 [0.38, 3.49]
Bodenheimer 1988	10/28	12/29		31.9	0.86 [0.45, 1.67]
Kaplan 1986	5/30	6/30		16.2	0.83 [0.28, 2.44]
Poupon 1996	2/37	0/37		1.4	5.00 [0.25, 100.72]
Vuoristo 1995	6/29	8/31		20.9	0.80 [0.32, 2.03]
Warnes 1987	5/34	5/30		14.4	0.88 [0.28, 2.75]
Subtotal (95% CI)	204	201		98.6	0.94 [0.63, 1.42]
Total events: 34 (Colchicine), 36 (Control)					
Test for heterogeneity chi-square=1.56 df=5 p=0.91 I² =0.0%					
Test for overall effect z=0.27 p=0.8					
02 Unclear or not performed					
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Raedsch 1993	2/14	0/14		1.4	5.00 [0.26, 95.61]
Subtotal (95% CI)	24	26		1.4	5.00 [0.26, 95.61]
Total events: 2 (Colchicine), 0 (Control)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.07 p=0.3					
Total (95% CI)	228	227		100.0	1.00 [0.67, 1.49]
Total events: 36 (Colchicine), 36 (Control)					
Test for heterogeneity chi-square=2.87 df=6 p=0.82 I² =0.0%					
Test for overall effect z=0.00 p=1					



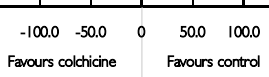
Analysis 05.11. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 11 S-bilirubin (ømol/L) - reported as arithmetic mean or geometric mean

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 11 S-bilirubin (µmol/L) - reported as arithmetic mean or geometric mean

Study	Colchicine N	Colchicine Mean(SD)	Control N	Control Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Arithmetic mean							
Almasio 2000	22	20.95 (15.96)	24	19.67 (16.28)		11.6	1.28 [-8.04, 10.60]
Ikeda 1996	10	14.88 (10.27)	12	16.76 (13.62)		10.0	-1.88 [-11.88, 8.12]
Poupon 1996	37	12.80 (6.69)	37	15.80 (9.73)		69.4	-3.00 [-6.80, 0.80]
Vuoristo 1995	29	23.80 (27.46)	31	15.30 (9.47)		9.0	8.50 [-2.04, 19.04]
Subtotal (95% CI)	98		104			100.0	-1.35 [-4.52, 1.82]
Test for heterogeneity chi-square=4.40 df=3 p=0.22 I² =31.8%							
Test for overall effect z=0.84 p=0.4							
02 Geometric mean							
Kaplan 1986	28	2.86 (1.05)	29	3.30 (1.11)		100.0	-0.44 [-1.00, 0.12]
Subtotal (95% CI)	28		29			100.0	-0.44 [-1.00, 0.12]
Test for heterogeneity: not applicable							
Test for overall effect z=1.54 p=0.1							



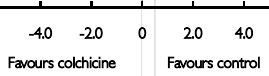
Analysis 05.12. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 12 S-cholesterol (total) (mmol/L) - reported as arithmetic mean or geometric mean

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 12 S-cholesterol (total) (mmol/L) - reported as arithmetic mean or geometric mean

Study	Colchicine N	Colchicine Mean(SD)	Control N	Control Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Arithmetic mean							
Vuoristo 1995	29	6.10 (1.62)	31	6.00 (2.23)		100.0	0.10 [-0.88, 1.08]
Subtotal (95% CI)	29		31			100.0	0.10 [-0.88, 1.08]
Test for heterogeneity: not applicable							
Test for overall effect z=0.20 p=0.8							
02 Geometric mean							
Kaplan 1986	28	1.86 (0.31)	29	1.88 (0.31)		100.0	-0.02 [-0.18, 0.14]
Subtotal (95% CI)	28		29			100.0	-0.02 [-0.18, 0.14]
Test for heterogeneity: not applicable							
Test for overall effect z=0.24 p=0.8							



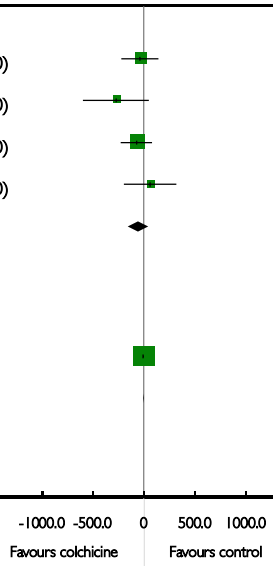
Analysis 05.13. Comparison 05 Subgroup analyses - colchicine versus placebo/no intervention, Outcome 13 S-alkaline phosphatase (ALP) (IU/L) - reported as arithmetic mean or geometric mean

Review: Colchicine for primary biliary cirrhosis

Comparison: 05 Subgroup analyses - colchicine versus placebo/no intervention

Outcome: 13 S-alkaline phosphatase (ALP) (IU/L) - reported as arithmetic mean or geometric mean

Study	Colchicine N	Colchicine Mean(SD)	Control N	Control Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Arithmetic mean							
Almasio 2000	24	556.60 (286.00)	22	587.40 (332.20)		32.1	-30.80 [-210.69, 149.09]
Ikeda 1996	10	297.00 (121.00)	12	563.20 (561.00)		9.8	-266.20 [-592.35, 59.95]
Poupon 1996	35	396.00 (259.60)	37	462.00 (400.40)		43.2	-66.00 [-221.05, 89.05]
Vuoristo 1995	29	892.00 (538.52)	31	826.00 (501.10)		14.9	66.00 [-197.69, 329.69]
Subtotal (95% CI)	98		102			100.0	-54.53 [-156.45, 47.38]
Test for heterogeneity chi-square=2.51 df=3 p=0.47 I² =0.0%							
Test for overall effect z=1.05 p=0.3							
02 Geometric mean							
Kaplan 1986	28	5.66 (0.69)	29	5.89 (0.71)		100.0	-0.23 [-0.59, 0.13]
Subtotal (95% CI)	28		29			100.0	-0.23 [-0.59, 0.13]
Test for heterogeneity: not applicable							
Test for overall effect z=1.24 p=0.2							



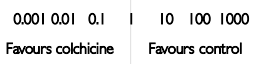
Analysis 06.01. Comparison 06 Sensitivity analyses - colchicine versus placebo/no intervention, Outcome 01 Number of deaths

Review: Colchicine for primary biliary cirrhosis

Comparison: 06 Sensitivity analyses - colchicine versus placebo/no intervention

Outcome: 01 Number of deaths

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Likely scenario					
Almasio 2000	6/46	4/44		19.3	1.43 [0.43, 4.74]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	5/30	6/30		28.3	0.83 [0.28, 2.44]
Poupon 1996	2/37	0/37		2.4	5.00 [0.25, 100.72]
Raedsch 1993	2/14	0/14		2.4	5.00 [0.26, 95.61]
Vuoristo 1995	5/29	5/31		22.8	1.07 [0.34, 3.31]
Warnes 1987	5/34	5/30		25.0	0.88 [0.28, 2.75]
Subtotal (95% CI)	200	198		100.0	1.21 [0.71, 2.06]
Total events: 25 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=2.63 df=5 p=0.76 I^2 =0.0%					
Test for overall effect z=0.70 p=0.5					
02 Best-worst-case scenario					
Almasio 2000	2/46	4/44		19.7	0.48 [0.09, 2.48]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	2/30	6/30		28.9	0.33 [0.07, 1.52]
Poupon 1996	2/37	0/37		2.4	5.00 [0.25, 100.72]
× Raedsch 1993	0/14	0/14		0.0	Not estimable
Vuoristo 1995	1/29	5/31		23.3	0.21 [0.03, 1.72]
Warnes 1987	5/34	5/30		25.6	0.88 [0.28, 2.75]
Subtotal (95% CI)	200	198		100.0	0.59 [0.30, 1.15]
Total events: 12 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=3.94 df=4 p=0.41 I^2 =0.0%					
Test for overall effect z=1.55 p=0.1					
03 Reported scenario					
Almasio 2000	2/46	2/44		18.9	0.96 [0.14, 6.50]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	2/30	1/30		9.3	2.00 [0.19, 20.90]
Poupon 1996	2/37	0/37		4.6	5.00 [0.25, 100.72]



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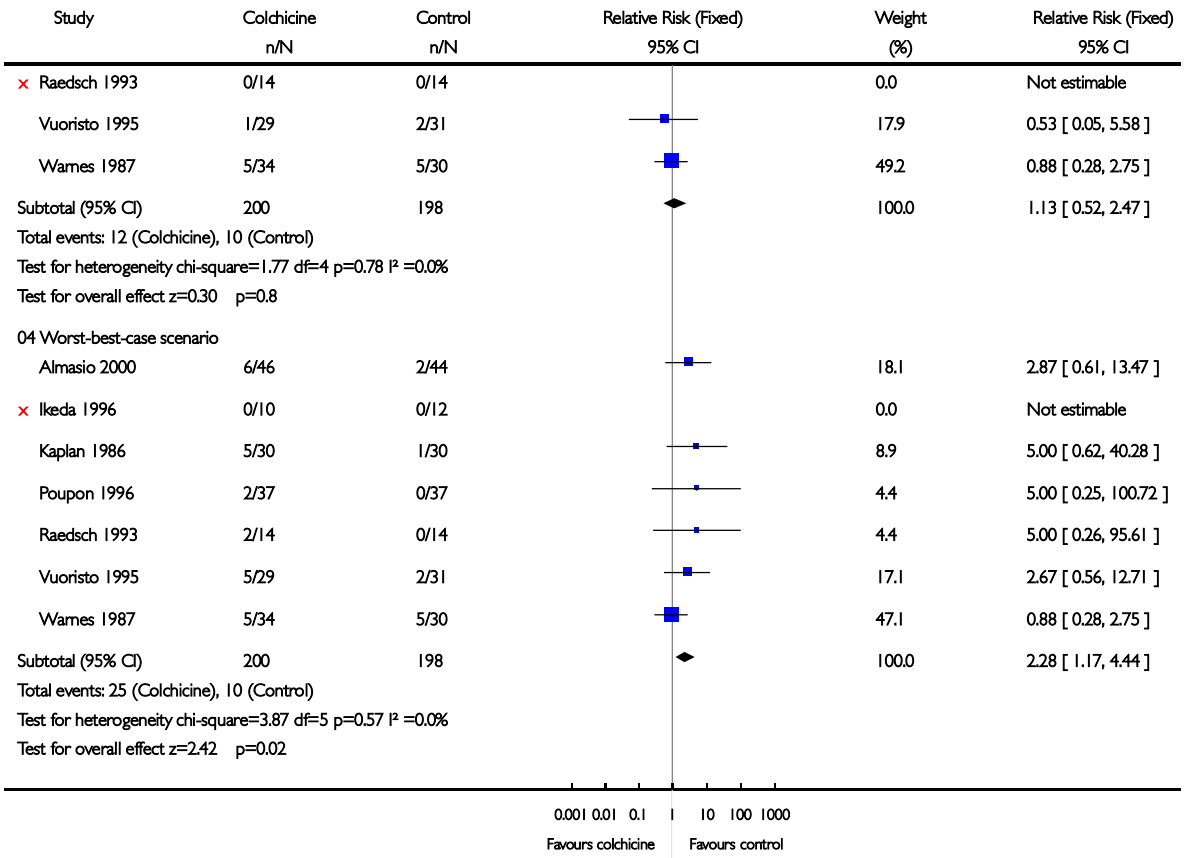


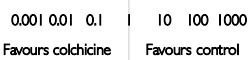
Analysis 06.02. Comparison 06 Sensitivity analyses - colchicine versus placebo/no intervention, Outcome 02 Number of deaths and/or patients who underwent liver transplantation

Review: Colchicine for primary biliary cirrhosis

Comparison: 06 Sensitivity analyses - colchicine versus placebo/no intervention

Outcome: 02 Number of deaths and/or patients who underwent liver transplantation

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Likely scenario					
Almasio 2000	6/46	5/44		13.8	1.15 [0.38, 3.49]
Bodenheimer 1988	10/28	12/29		31.9	0.86 [0.45, 1.67]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	5/30	6/30		16.2	0.83 [0.28, 2.44]
Poupon 1996	2/37	0/37		1.4	5.00 [0.25, 100.72]
Raedsch 1993	2/14	0/14		1.4	5.00 [0.26, 95.61]
Vuoristo 1995	6/29	8/31		20.9	0.80 [0.32, 2.03]
Warnes 1987	5/34	5/30		14.4	0.88 [0.28, 2.75]
Subtotal (95% CI)	228	227		100.0	1.00 [0.67, 1.49]
Total events: 36 (Colchicine), 36 (Control)					
Test for heterogeneity chi-square=2.87 df=6 p=0.82 I² =0.0%					
Test for overall effect z=0.00 p=1					
02 Best-worst-case scenario					
Almasio 2000	2/46	5/44		14.0	0.38 [0.08, 1.87]
Bodenheimer 1988	2/28	12/29		32.3	0.17 [0.04, 0.70]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	2/30	6/30		16.5	0.33 [0.07, 1.52]
Poupon 1996	2/37	0/37		1.4	5.00 [0.25, 100.72]
× Raedsch 1993	0/14	0/14		0.0	Not estimable
Vuoristo 1995	2/29	8/31		21.2	0.27 [0.06, 1.16]
Warnes 1987	5/34	5/30		14.6	0.88 [0.28, 2.75]
Subtotal (95% CI)	228	227		100.0	0.42 [0.24, 0.74]
Total events: 15 (Colchicine), 36 (Control)					
Test for heterogeneity chi-square=6.26 df=5 p=0.28 I² =20.1%					
Test for overall effect z=3.02 p=0.003					
03 Reported scenario					
Almasio 2000	2/46	3/44		14.9	0.64 [0.11, 3.64]
Bodenheimer 1988	2/28	6/29		28.6	0.35 [0.08, 1.57]



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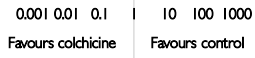
Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	2/30	1/30		4.9	2.00 [0.19, 20.90]
Poupon 1996	2/37	0/37		2.4	5.00 [0.25, 100.72]
× Raedsch 1993	0/14	0/14		0.0	Not estimable
Vuoristo 1995	2/29	5/31		23.5	0.43 [0.09, 2.03]
Warnes 1987	5/34	5/30		25.8	0.88 [0.28, 2.75]
Subtotal (95% CI)	228	227		100.0	0.74 [0.39, 1.39]
Total events: 15 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=3.81 df=5 p=0.58 I² =0.0%					
Test for overall effect z=0.94 p=0.3					
04 Worst-best-case scenario					
Almasio 2000	6/46	3/44		14.5	1.91 [0.51, 7.18]
Bodenheimer 1988	10/28	6/29		27.9	1.73 [0.72, 4.12]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	5/30	1/30		4.7	5.00 [0.62, 40.28]
Poupon 1996	2/37	0/37		2.4	5.00 [0.25, 100.72]
Raedsch 1993	2/14	0/14		2.4	5.00 [0.26, 95.61]
Vuoristo 1995	6/29	5/31		22.9	1.28 [0.44, 3.75]
Warnes 1987	5/34	5/30		25.2	0.88 [0.28, 2.75]
Subtotal (95% CI)	228	227		100.0	1.75 [1.07, 2.86]
Total events: 36 (Colchicine), 20 (Control)					
Test for heterogeneity chi-square=3.66 df=6 p=0.72 I² =0.0%					
Test for overall effect z=2.24 p=0.03					



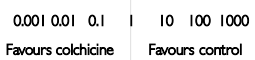
Analysis 06.03. Comparison 06 Sensitivity analyses - colchicine versus placebo/no intervention, Outcome 03 Number of deaths and/or patients who underwent liver transplantation (excluding Bodenheimer 1988)

Review: Colchicine for primary biliary cirrhosis

Comparison: 06 Sensitivity analyses - colchicine versus placebo/no intervention

Outcome: 03 Number of deaths and/or patients who underwent liver transplantation (excluding Bodenheimer 1988)

Study	Colchicine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Likely scenario					
Almasio 2000	6/46	5/44		20.3	1.15 [0.38, 3.49]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	5/30	6/30		23.9	0.83 [0.28, 2.44]
Poupon 1996	2/37	0/37		2.0	5.00 [0.25, 100.72]
Raedsch 1993	2/14	0/14		2.0	5.00 [0.26, 95.61]
Vuoristo 1995	6/29	8/31		30.7	0.80 [0.32, 2.03]
Warnes 1987	5/34	5/30		21.1	0.88 [0.28, 2.75]
Subtotal (95% CI)	200	198		100.0	1.06 [0.65, 1.75]
Total events: 26 (Colchicine), 24 (Control)					
Test for heterogeneity chi-square=2.75 df=5 p=0.74 I² =0.0%					
Test for overall effect z=0.24 p=0.8					
02 Best-case scenario					
Almasio 2000	2/46	5/44		20.7	0.38 [0.08, 1.87]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	2/30	6/30		24.3	0.33 [0.07, 1.52]
Poupon 1996	2/37	0/37		2.0	5.00 [0.25, 100.72]
× Raedsch 1993	0/14	0/14		0.0	Not estimable
Vuoristo 1995	2/29	8/31		31.4	0.27 [0.06, 1.16]
Warnes 1987	5/34	5/30		21.5	0.88 [0.28, 2.75]
Subtotal (95% CI)	200	198		100.0	0.54 [0.28, 1.01]
Total events: 13 (Colchicine), 24 (Control)					
Test for heterogeneity chi-square=4.28 df=4 p=0.37 I² =6.5%					
Test for overall effect z=1.93 p=0.05					
03 Reported scenario					
Almasio 2000	2/46	3/44		20.8	0.64 [0.11, 3.64]
× Ikeda 1996	0/10	0/12		0.0	Not estimable
Kaplan 1986	2/30	1/30		6.8	2.00 [0.19, 20.90]
Poupon 1996	2/37	0/37		3.4	5.00 [0.25, 100.72]



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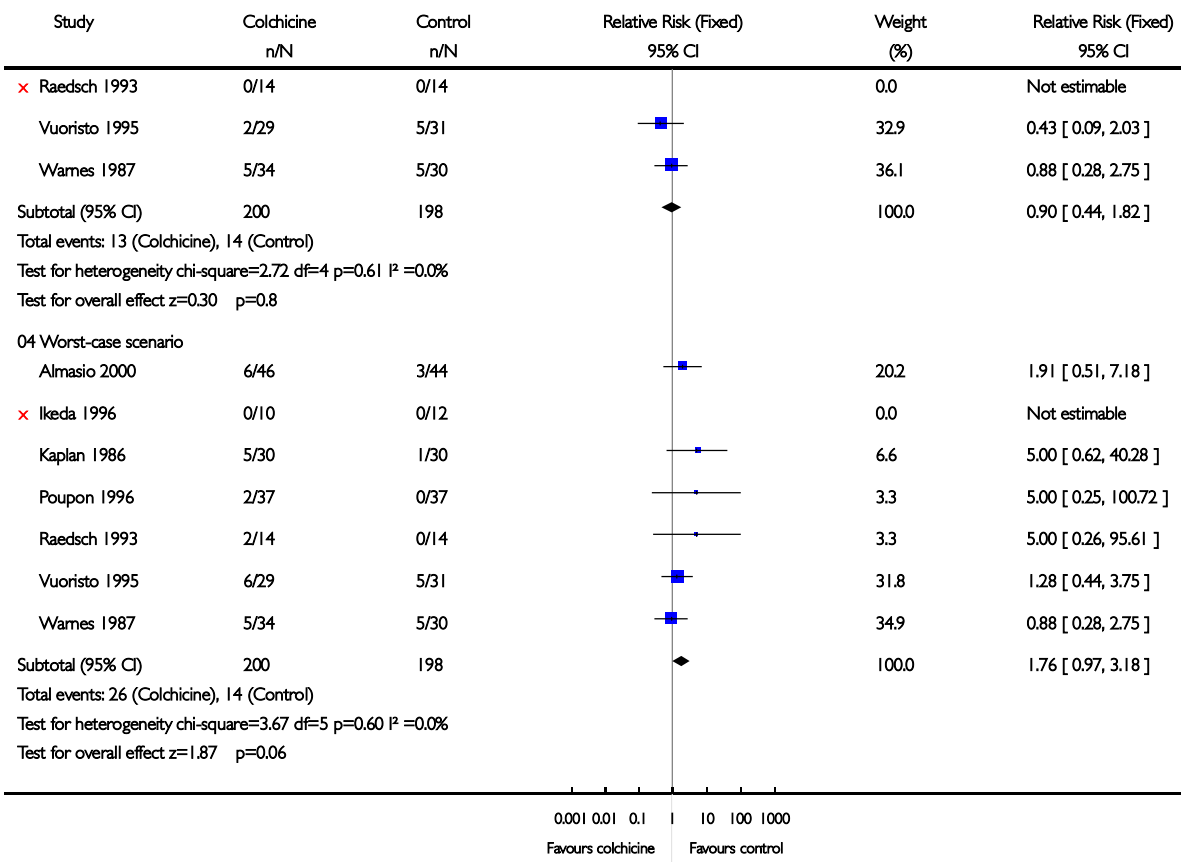

Appendix 3B

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Colchicine for Primary Biliary Cirrhosis: A Cochrane Hepato-biliary Group Systematic Review of Randomized Clinical Trials

Yan Gong, M.D., and Christian Gluud, Pr.Med.Sc.
The Cochrane Hepato-Biliary Group, Copenhagen Trial Unit, Center for Clinical Intervention Research, H:S Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark

OBJECTIVES: Colchicine is used for patients with primary biliary cirrhosis due to its immunomodulatory and antifibrotic potential. The results from randomized clinical trials have, however, been inconsistent. We conducted a systematical review to evaluate the effect of colchicine for primary biliary cirrhosis.

METHODS: We identified randomized clinical trials comparing colchicine with placebo/no intervention. We analyzed effects by fixed and random effects model. We investigated heterogeneity by subgroup and sensitivity analyses.

RESULTS: We included 10 trials involving 631 patients, four of which were high-quality trials. No significant differences were detected between colchicine and placebo/no intervention regarding mortality (relative risk (RR), 1.21; 95% confidence interval (CI), 0.71–2.06), mortality or liver transplantation (RR = 1.00; 95% CI, 0.67–1.49), liver complications, liver biochemical variables, liver histology, or adverse events. Regarding mortality, an extreme case analysis favoring colchicine did not demonstrate beneficial effects of colchicine, whereas an extreme case analysis favoring placebo/no intervention demonstrated a detrimental effect of colchicine (RR = 2.28; 95% CI, 1.17–4.44). The number of patients without improvement of pruritus significantly decreased in the colchicine group (RR = 0.75; 95% CI, 0.65–0.87). However, this estimate was based on only 156 patients from three trials.

CONCLUSIONS: There is insufficient evidence to support the use of colchicine for patients with primary biliary cirrhosis. As we are unable to exclude a risk of increased mortality, we recommend to use colchicine only in randomized clinical trials.

(Am J Gastroenterol 2005;100:1876–1885)

INTRODUCTION

Primary biliary cirrhosis is an uncommon chronic disease of unknown etiology. Over the last 30 yr, prevalence and incidence of primary biliary cirrhosis was substantially increased in many countries (1, 2). Ninety percent of patients with primary biliary cirrhosis are females and the majority are diagnosed after the age of 40 (3). Primary biliary cirrhosis is classically defined on the basis of the triad: antimitochondrial antibodies (4, 5); abnormal liver function tests that are typically cholestatic (with raised activity of alkaline phosphatases being the most frequently seen abnormality); and characteristic liver histological changes in the absence of extrahepatic biliary obstruction (6, 7).

Patients with primary biliary cirrhosis have been subjected to many drugs, for example, ursodeoxycholic acid (UDCA), azathioprine, prednisolone, chlorambucil, cyclosporine, D-penicillamine, methotrexate, and colchicine (8). The clinical effects observed have not led to widespread acceptance of these drugs for primary biliary cirrhosis patients (9).

Colchicine is used for primary biliary cirrhosis patients (10) because it may slow progression (11) and improve liver biochemical tests and immunoglobulin levels (12–14). However, colchicine did not appear to affect clinical symptoms or liver histology (15). Combination therapy with colchicine and UDCA has also been assessed, but the results have been conflicting (16–20). We, therefore, performed a systematic review to assess the effect of colchicine in primary biliary cirrhosis.

METHODS

Search Strategy

We searched for trials in The Cochrane Hepato-Biliary Group Controlled Trials Register (June 2004), The Cochrane Central Register of Controlled Trials on The Cochrane Library (Issue 3, 2004), MEDLINE (January 1966 to August 2004), EMBASE (January 1980 to August 2004), The Chinese Biomedical CD Database (1978–2003), LILACS

(1982–2003), and references of identified studies. We contacted the principal authors and sponsor companies of the identified trials (Eli Lilly, USA, and Shionogi & Co., Ltd., Japan) to obtain any unidentified trials and additional information on identified trials. The details of the search strategy are outlined elsewhere (21).

Eligibility and Data Extraction

We assessed only randomized clinical trials comparing colchicine *versus* placebo/no intervention irrespective of language, year of publication, or publication status (21). Co-interventions were allowed as long as all intervention arms received similar co-interventions. The authors independently scrutinized all articles and extracted data from the trials. Any disagreement about data extraction was resolved by discussion. The authors independently extracted trial data onto a standard form that focused on four aspects of methodological quality in randomized clinical trials (22–24): generation of the allocation sequence; allocation concealment; blinding; follow-up.

Outcome Measures

The primary outcome measures were mortality and mortality or liver transplantation. Our secondary outcome measures were: liver transplantation, pruritus, fatigue, liver complications (variceal bleeding, ascites, hepatic encephalopathy, jaundice, or hepato-renal syndrome), liver biochemistry variables, liver biopsy findings, adverse events (25), quality of life, and cost-effectiveness (21).

Data Analysis

The metaanalysis was performed in Review Manager Software (version 4.2.7) from The Cochrane Collaboration (www.cochrane.org). We calculated an overall weighted estimate of the relative risk (RR) with 95% confidence interval (CI) for binary outcomes and weighted averages of differences between means for continuous outcomes. We examined intervention effects by a random effects model (26) and a fixed effect model (27).

We performed subgroup analyses (28) in which trials were grouped according to methodological quality, dosage of colchicine, trial duration, or combination of colchicine with UDCA. The cut-off for trial duration was the median value of the included trials. We performed sensitivity analyses to assess the impact of missing responses on primary outcomes: "available patients' course analysis," including data on only those whose results are known, using the total number of patients who completed the trial as denominator, and intention-to-treat analysis using imputation (29) on different numerators and all randomized patients as denominators. That is, in the "extreme case favoring colchicine' scenario, we assumed that none of colchicine-group patients but all controls dropouts had the primary outcomes. The "extreme case favoring placebo/no intervention" scenario was opposite: all dropouts from the colchicine group but no controls had primary outcomes. In the "assuming good outcome scenario," we assumed that none of colchicine and controls dropouts had the primary outcomes, whereas in the "assuming poor outcome scenario," we assumed that colchicine and placebo dropouts both had the primary outcomes. For secondary outcomes, we adopted "available patients course analysis." Therefore, in the review, the number of patients in the denominator changed according to the secondary outcomes investigated.

By using the statistical package STATA™, we used the Egger *et al*. regression asymmetry test to assess funnel plot asymmetry indicating the presence of publication bias and other biases (30).

RESULTS

Search Results

We identified 705 references, of which we excluded 673 duplicates, clearly irrelevant references, reviews, and non-randomized clinical studies. The remaining 32 references referred to 10 included randomized clinical trials involving 631 patients. The randomization created comparable intervention groups in the respective trials. The baseline characteristics of the patients are summarized in Table 1.

Methodological Quality of the Trials

The methodological quality of the included trials is summarized in Table 2. Generation of allocation sequence was adequate in four trials (12, 14, 15, 19) and unclear or inadequate in six trials (17, 18, 31–34). Concealment of treatment allocation was adequate in five trials (12, 13, 15, 17, 19) and unclear or inadequate in the other five (18, 31–33, 34). Nine trials reported double-blinding and one trial (17) was not blinded. However, the description of the control in the trials reporting double blinding was not sufficient. Some of the trials (13, 33, 34) did not give any description of the placebo used. Other trials stated that the placebo tablets were identical in appearance or indistinguishable, but did not address smell and taste (12, 15, 18, 19). In total, 48 (8%) patients had been excluded after randomization or were lost to follow-up: 30 (11%) patients in colchicine group and 18 (6%) patients in control group. In one trial, 8 (29%) colchicine patients and 6 (21%) placebo patients were lost to follow-up (31). An intention-to-treat analysis was claimed in four trials (12, 13, 17, 19). Sample size estimation was mentioned in one trial (15), but no estimation was based on mortality. We classified trials with at least two out of three criteria (adequate generation allocation, adequate allocation concealment, and adequate blinding) as high quality. Accordingly, we considered four trials as high quality (12, 13, 15, 19).

Mortality

Data from seven trials with 398 patients were available to estimate the risk of mortality. The available patients' course analysis (RR = 1.12; 95% CI, 0.51–2.46), the analysis assuming poor outcome scenario (RR = 1.21; 95% CI, 0.71–2.06),

Table 1. Characteristics of Included Trials

First Author Publication Year; Country	Trial Duration (Yr)	Intervention	Number of Patients	Patients' Characteristics
Almasio (2000)[19]; Italy	3	Colchicine 1 mg/day + UDCA† 250 mg twice daily	46	Entry data in intervention groups comparable. Mean age: 53.3 yr in colchicine group, 55.5 yr in placebo group.
		Placebo + UDCA 250 mg twice daily	44	Inclusion criteria: 1. An established diagnosis of primary biliary cirrhosis according to Taal *et al*. (40). 2. Pruritus. 3. Serum bilirubin exceeding 2 mg/dl. 4. Histological diagnosis of cirrhosis. Histological stage at entry: Stage I/II: 8 in colchicine group, 8 in control group. Stage III/IV: 28 in colchicine group, 27 in control group. Others were unknown.
Bodenheimer (1988)[31]; USA	4	Colchicine 1.2 mg/day (0.6 mg twice daily)	28	Entry data in intervention groups comparable. Mean age: 53 yr in colchicine group, 51 yr in placebo group.
		Placebo	29	Inclusion criteria: 1. History of chronic cholestatic liver disease. 2. Liver biopsy results compatible with primary biliary cirrhosis. Histological stage at entry: Stage I/II: 8 in colchicine group, 11 in control group. Stage III/IV: 20 in colchicine group, 18 in control group. Others were unknown.
Goddard (1995)*,[32]; UK	2.5	Colchicine 1 mg/day UDCA 10 mg/kg per day Colchicine + UDCA (dose not reported)	57 (total)	Entry data in intervention groups comparable. Mean age: not reported. Inclusion criteria: not reported. Histological stage at entry: unreported.
		UDCA 600 mg/day Placebo	12	Entry data in intervention groups comparable. Mean age: 59.5 yr in colchicine group and 66.5 yr in UDCA group. Inclusion criteria:
Ikeda (1996)[17]; Japan	2	Colchicine 1 mg/day + UDCA 600 mg/day	10	1. Raised alkaline phosphatases. 2. Antimitochondrial antibody test positive. 3. Liver biopsy results compatible with primary biliary cirrhosis. 4. Radiological or ultrasonographic evidence that the bile ducts were patent. Histological stage at randomization: unclear.
Kaplan (1986)[15]; USA	4	Colchicine 1.2 mg/day (0.6 mg twice daily)	30	Entry data in intervention groups comparable. Mean age: not reported.
		Placebo	30	Inclusion criteria: 1. Antimitochondrial antibody test positive. 2. Liver-biopsy proven primary biliary cirrhosis. 3. Radiological or ultrasonographic evidence that bile ducts were patent. Histological stage at entry: Stage I/II: 15 in colchicine group, 15 in control group. Stage III/IV: 15 in colchicine group, 15 in control group. Note: at the end of 2-yr double-blind period, each patient was placed in an open-label study of colchicine for another 2 yr.
Poupon (1996)[18]; France and Canada	2	Colchicine 1 mg/day, 5 days/wk + UDCA 13–15 mg/kg per day	37	Entry data in intervention groups comparable.

Continued

Table 1. (Continued.)

First Author Publication Year; Country	Trial Duration (Yr)	Intervention	Number of Patients	Patients' Characteristics
		Placebo + 13–15 mg/kg per day UDCA	37	Mean age: 55 yr in colchicine group, 52 yr in placebo group. Inclusion criteria: 1. Biopsy-proven primary biliary cirrhosis. 2. No less than eight months previous treatment with UDCA(13–15 mg/kg per day). 3. Alkanline phosphatases activity more than 1.5 times the upper limit of normal. Histological stage at entry: Stage I/II: 21 in colchicine group, 20 in control group. Stage III/IV: 16 in colchicine group, 17 in control group.
Raedsch (1993)[33]; Germany	3	Colchicine 1 mg/day + UDCA 10–12 mg/kg per day	14	Comparability: not reported. Mean age: 54 yr in the patients. Inclusion criteria: 1. Blood biochemistry (details not reported). 2. Antimitochondrial antibody test positive. 3. Liver biopsy results compatible with primary biliary cirrhosis. Histological stage at randomization: unclear.
		Placebo + UDCA 10–12 mg/kg per day	14	
Vuoristo (1995)[13]; Finland	2	Colchicine 1 mg/day	29	Entry data in intervention groups comparable. Mean age: 56, 52, and 57 yr in colchicine, UDCA, and placebo groups. Inclusion criteria: 1. Raised alkaline phosphatases. 2. Liver biopsy compatible with primary biliary cirrhosis. 3. Antimitochondrial antibody test positive. Histological stage at randomization: unreported.
		UDCA 12–15 mg/kg per day	31	
		Placebo	30	
Warnes (1987)[12]; UK	1.5	Colchicine 1 mg/day	34	Entry data in intervention groups Comparable, except that AMA titers were significantly higher in the colchicine group. Mean age: not reported. Inclusion criteria: 1. Raised serum alkaline phosphatases. 2. Antimitochondrial antibody test positive. 3. Liver histology compatible with, or diagnostic of primary biliary cirrhosis. Histological stage at entry: Stage I/II: 6 in colchicine group, 5 in control group. Stage III/IV: 24 in colchicine group, 20 in control group.
		Placebo	30	
Warnes (1996)*,[34]; UK	1.5	Colchicine (dose not specified)	89 (total)	Comparability: not reported. Mean age: not reported. Inclusion criteria: not reported. Histological stage at randomization: unreported.
		Placebo		

†UDCA: ursodeoxycholic acid.
*Note: only a published abstract available.

the extreme case favoring colchicine analysis (RR = 0.59; 95% CI, 0.30–1.15), and the analysis assuming good outcome (RR = 1.13; 95% CI, 0.52–2.47) showed no significant differences between colchicine and placebo/no intervention (Fig. 1). The analysis favoring placebo/no intervention detected a significant detrimental effect of colchicine (RR = 2.28; 95% CI, 1.17–4.44). There was no significant heterogeneity (I^2 = 0%) (35). There are no significant differences across all the subgroups analyses regarding methodological quality of the trials, dosage of colchicine,

Table 2. Assessment of Methodological Quality of Included Trials

Trial	Generation of Allocation Sequence	Allocation of Concealment	Sample Size Estimation	Blinding	Number Lost to Follow-Up (%)	
					Colchicine	Control
Almasio (2000)[19]	Adequate	Adequate	Not reported	Adequate	4 (9%)	2 (5%)
Bodenheimer (1988)[31]	Unclear	Unclear	Not reported	Adequate	8 (29%)	6 (21%)
Goddard (1995)*,[32]	Unclear	Unclear	Not reported	Unclear	Not reported	Not reported
Ikeda (1996)[17]	Unclear	Unclear	Not reported	Not performed	0 (0%)	0 (0%)
Kaplan (1986)[15]	Adequate	Adequate	Yes†	Adequate	3 (10%)	5 (17%)
Poupon (1996)[18]	Unclear	Unclear	Not reported	Adequate	2 (5%)	0 (0%)
Raedsch (1993)[33]	Unclear	Unclear	Not reported	Unclear	2 (7%)	0 (0%)
Vuoristo (1995)[13]	Adequate	Adequate	Not reported	Adequate	3 (10%)	3 (10%)
Warnes (1987)[12]	Adequate	Adequate	Not reported	Adequate	8 (24%)	2 (7%)
Warnes (1996)*,[34]	Unclear	Unclear	Not reported	Unclear	Not reported	Not reported

*Note: only a published abstract available.
†The estimation was based on treatment failure, not mortality.

trial duration, and combination of colchicine with UDCA (Table 3).

Mortality or Liver Transplantation

Data from eight trials with 455 patients were available to estimate the risk of mortality or liver transplantation (Fig. 2). Neither the available patients course analysis and the assuming poor outcome scenario, nor the assuming good outcome scenario showed any significant difference between colchicine and placebo/no intervention. The extreme case favoring colchicine or placebo/no intervention showed the significant effect favoring colchicine or placebo/no intervention. There are no significant differences across the subgroups methodological quality of the trials, dosage of colchicine, trial duration, and combination of colchicine with UDCA (data not shown).

Pruritus, Fatigue, and Liver Complications

Pooling the data from three trials with 156 patients demonstrated that colchicine significantly ameliorated pruritus (RR = 0.75; 95% CI, 0.65–0.87, $p = 0.0002$) (Fig. 3). Colchicine did not significantly influence fatigue (RR = 0.86; 95% CI, 0.72–1.02) or liver complications (RR = 0.37; 95% CI, 0.12–1.10). We were not able to extract data on jaundice.

Liver Biochemical and Histological Outcomes

Colchicine did not lead to any significant effect on the liver biochemical variables (Table 4). There was no significant influence of colchicine on the number of patients experiencing worsening of histological stage (RR = 0.85; 95% CI, 0.41–1.75), fibrosis (RR = 0.60; 95% CI, 0.24–1.49), piecemeal necrosis (RR = 0.58; 95% CI, 0.23–1.44), parenchymal inflammation (RR = 0.69; 95% CI, 0.28–1.72), parenchymal necrosis (RR = 1.00; 95% CI, 0.31–3.18), or histological score (WMD = 0.56; 95% CI, –0.24–1.36).

Adverse Events

Colchicine tended to lead to more adverse events (mostly transient diarrhea, usually resolved by lowering the dose of colchicine), but it is not significant (RR = 1.45; 95% CI, 0.94–2.25). We found no significant difference regarding serious adverse events (RR = 1.17; 95% CI, 0.50–2.75).

Quality of Life and Cost-effectiveness

None of the trials examined specific quality-of-life scales or cost-effectiveness.

Biases Detection

The funnel plots on the primary outcome measures did not show obvious asymmetry (figure not shown). We could not perform linear regression to detect publication bias and other biases due to the low number of trials.

DISCUSSION

This systematic review reveals that colchicine has no significant effects on reducing the risks of mortality and mortality or liver transplantation, compared to placebo/no intervention in patients with primary biliary cirrhosis. These observations were robust to different scenarios when missing responses were considered. The extreme case favoring placebo/no intervention scenario detected a significant detrimental effect of colchicine on mortality.

In the subgroup analyses, considering methodological quality of trials, the dosage of colchicine, or trial duration did not reveal any significant difference regarding the intervention effect of colchicine on mortality or mortality or liver transplantation. Generally, low methodological quality trials overestimate intervention effects (22–24). We did not find this tendency in the present sample of trials, probably due to the relatively small sample size of the individual trials and small number of trials included in this review. The subgroup analyses, stratifying the included trials into monotherapy (*i.e.,*

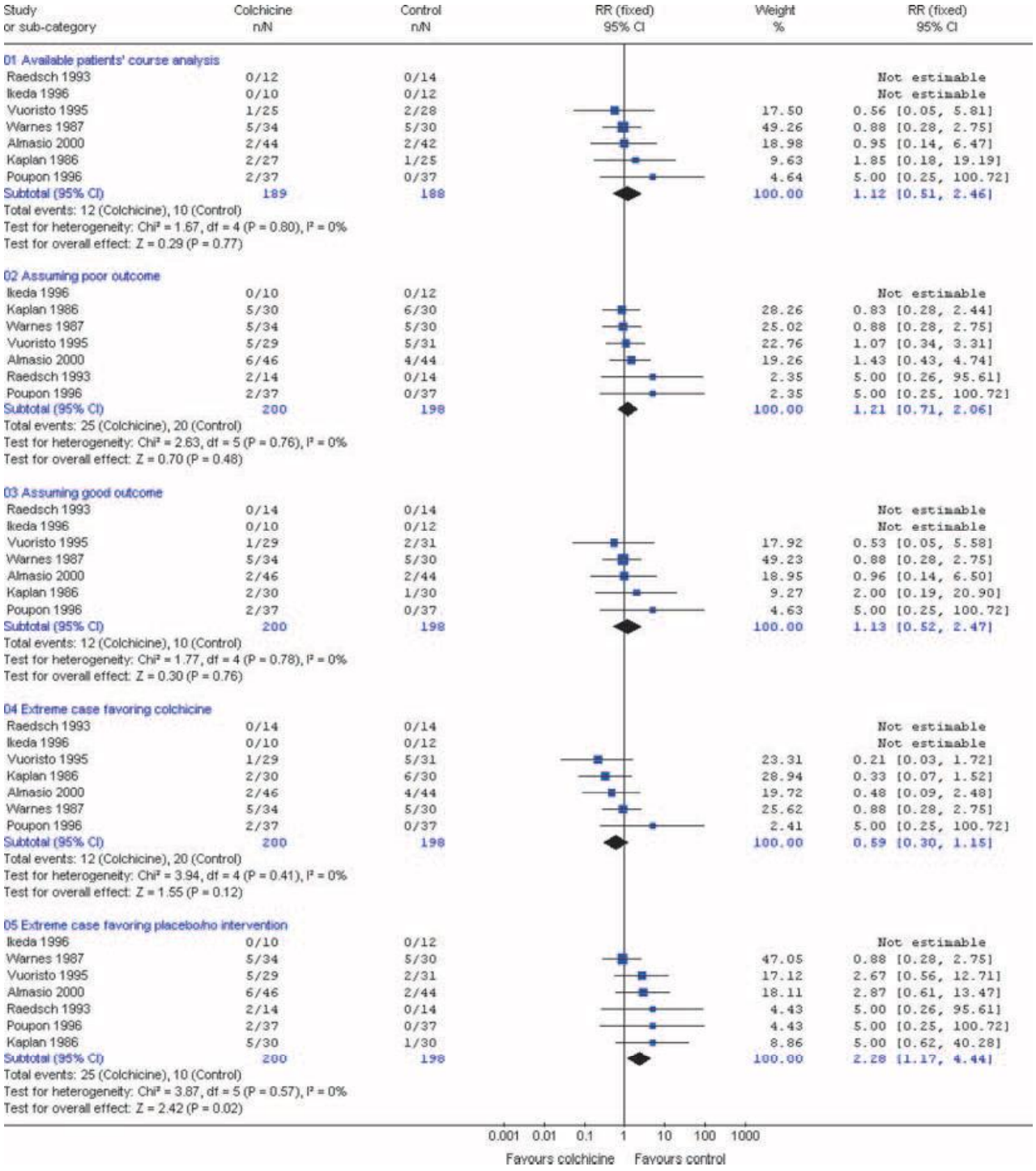

Figure 1. Relative risk of mortality in primary biliary cirrhosis patients randomized to colchicine *versus* placebo/no intervention—sensitivity analysis.

colchicine alone) and combination therapy (*i.e.,* colchicine plus UDCA), did not suggest additional effect of colchicine introduced by the combination with UDCA (Table 3). Further, our observations were robust to sensitivity analyses leaving one trial out at a time (data not shown).

Colchicine seems not to have beneficial effects on liver biochemical parameters. It appeared that the use of colchicine was associated with improvement in hepatic biochemistry in three early trials (12, 15, 31). In these three trials, however, protocol violations regarding the percentage of randomized

Table 3. Relative Risks of Mortality in Primary Biliary Cirrhosis Patients Randomized to Colchicine *versus* Placebo/No Intervention–Subgroup Analyses

Subgroup		Number of Trials	RR	95% CI	Test of Interaction Test
Methodological quality of trials	High*	4[19,15,13,12]	1.02	0.58–1.80	$p = 0.16$
	Low†	3[17,18,33]	5.00	0.61–41.04	
Dosage of colchicine (mg)	1	6[19,17,18,33,13,12]	1.36	0.73–2.52	$p = 0.44$
	1.2	1[15]	0.83	0.28–2.44	
Trial duration (yr)	≤2	5[17,15,18,13,12]	1.04	0.56–1.93	$p = 0.38$
	>2	2[19,33]	1.82	0.62–5.39	
Combination colchicine with UDCA‡	Yes	4[17–19,33]	2.14	0.78–5.87	$p = 0.17$
	No	3[12,13,15]	0.92	0.48–1.75	

*High: trials with adequate generation of allocation sequence, allocation concealment, and blinding.
†Low: trials with unclear or inadequate generation of allocation sequence, allocation concealment, or blinding.
‡UDCA: ursodeoxycholic acid.

patients, who were either lost to follow-up, refused liver biopsy, were noncompliant, or were withdrawn due to adverse events or disease progression, were substantial: 33, 13, and 38%, respectively. Only one of the trials stated having employed the intention-to-treat principle (12). Our meta-analyses show that colchicine does not seem to have major influence on liver biochemistry.

In this review, we were not able to identify any significant effect of colchicine on a number of histological variables. Almasio *et al*. reported a significant reduction in histological grading score in patients administrating colchicine plus UDCA; however, the proportion of patients having liver biopsy was very low, 15 patients out of 90 (19). Thus, its significance could be biased due to the missing responses.

Evidence showed that colchicine may have beneficial effect on pruritus. This finding was based upon only three trials involving 156 patients. A number of arguments may contradict this observation. First, lack of efficient blinding of trials (24) and the subjective nature of pruritus assessment could have biased the estimate. Second, pruritus usually reflects indices of cholestasis (*e.g.,* serum alkaline phosphatases) and a correlation between the severity of pruritus and the presence of florid bile duct lesions in the liver has been reported (36). In our analyses, there were not such consistent findings observed either in any plasma indices of cholestasis or in liver histology. Furthermore, due to the large number of statistical comparisons having been performed some of the comparisons might have come out with a significant difference simply due to 'mass significance' (*i.e.,* spurious significant findings due to repetitive testing). Therefore, we are not convinced that the improvement of pruritus was due to colchicine. The potential beneficial effect of colchicine on pruritus might be worth exploring in future high-quality randomized trials.

In order to examine the effects of colchicine in a broader context, we combined our mortality data with those of patients with alcoholic and non-alcoholic liver fibrosis and cirrhosis (14 randomized clinical trials) (37). The pooled results showed no significant difference on mortality without significant heterogeneity (RR = 0.98; 95% CI, 0.78–1.24; $I^2 = 0\%$). Although this estimate does leave room for both beneficial and harmful effects, it does not function as a strong rationale for conducting further trials.

No study is perfect. Neither is this systematic review. The metaanalyses regarding mortality only involved seven trials with 398 patients. This is a low number of patients (38).

Additionally, compared to the natural history of primary biliary cirrhosis, most of the trials had relatively short period of medication and follow-up. Thus, the risk of type II error is present and a potential beneficial effect of colchicine on survival cannot be reliably excluded. Secondly, since the trial reports did not provide data of stratum according to the patients' baseline stage, we were not able to know whether the effect of colchicine was associated with the severity of the disease. Thirdly, although we have employed considerable search strategies and applied no publication status or language limitations, we are concerned about publication bias and other biases, which leads us to identify 'positive' studies more easily than 'negative' ones (39).

Overall, we did not find convincing evidence showing that colchicine had significant beneficial effects on patients with primary biliary cirrhosis. The combination of colchicine and UDCA did not significantly change the effects of colchicine. We are not able to exclude the possibility that colchicine may reduce mortality by 70%. On the other hand, it may also increase mortality by 344%. Therefore, we think that colchicine should not be used outside randomized clinical trials. Considering that about 2.5 million patients in the world may have primary biliary cirrhosis (1), about 140,000 primary biliary cirrhosis patients may be receiving colchicine (10).

If any researchers have an interest to investigate colchicine for primary biliary cirrhosis, they may consider the following: (1) due to the chronic progression of primary biliary cirrhosis, long-term follow-up is needed; (2) an independent data monitoring and safety committee should be used in order to follow the data closely for any adverse events; (3) study in detail the potential effect of colchicine on pruritus; (4) ensure that enough patients with primary biliary cirrhosis are kept to undergo liver biopsy in order that more data on liver histology become available; (5) include quality of life and cost-effective analyses; (6) adhere to the Consort Statement (www.consort-statement.org).

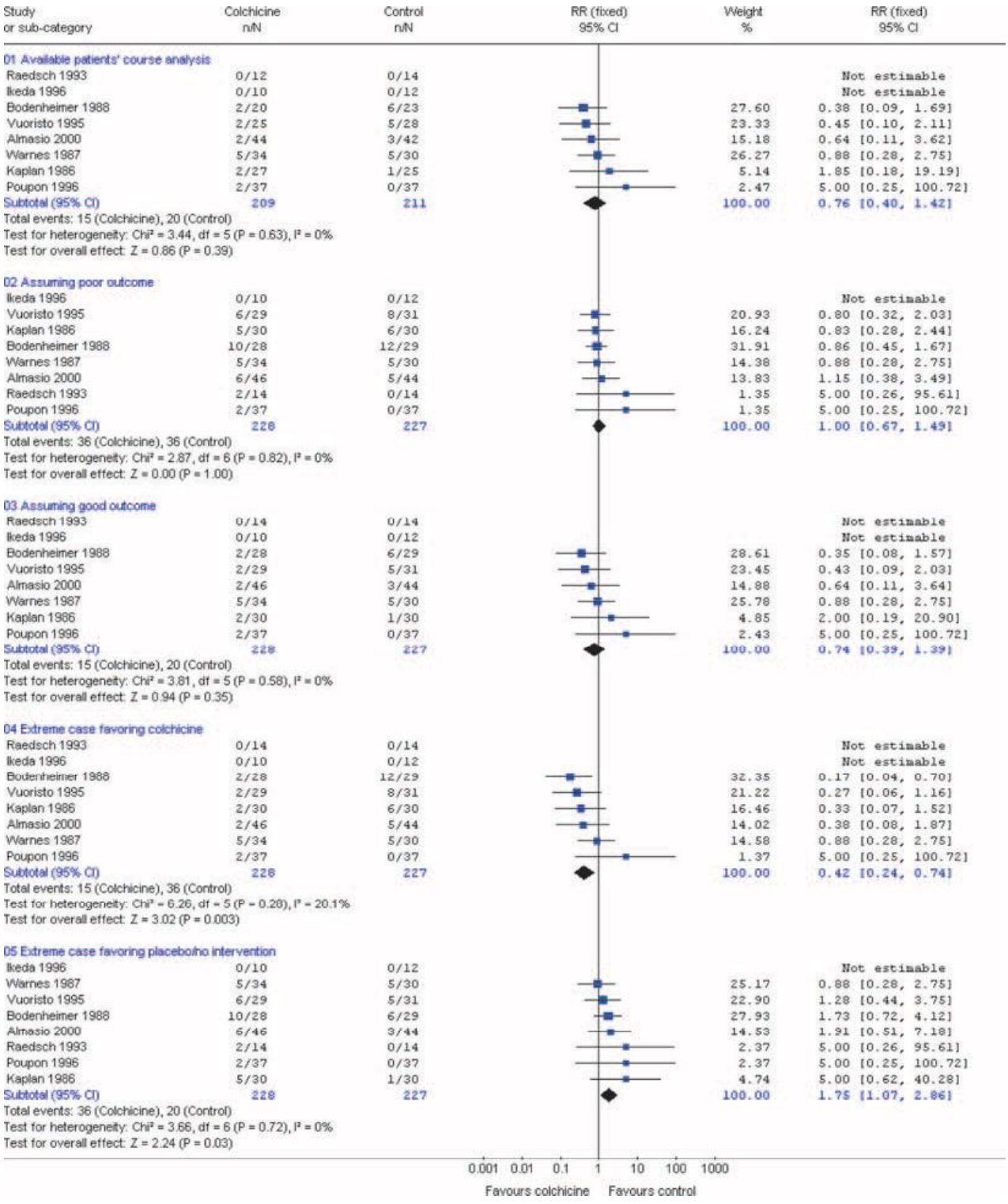

Figure 2. Relative risk of mortality or undergoing liver transplantation in primary biliary cirrhosis patients randomized to colchicine *versus* placebo/no intervention—sensitivity analysis.

ACKNOWLEDGMENTS

We primarily extend our acknowledgments to the patients who took part in and the investigators who designed and conducted the reviewed trials. Furthermore, we give our special gratitude to Piero Almasio, Takaaki Ikeda, Marshal Kaplan, Matti Vuoristo, and Shionogi Pharma for providing us

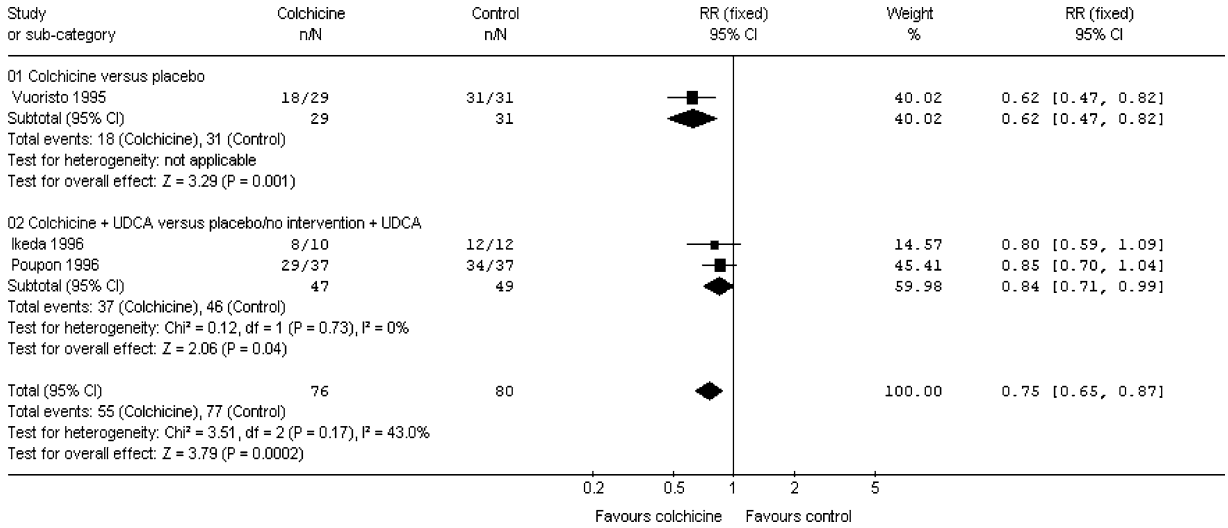

Figure 3. Relative risk of no improvement of pruritus in primary biliary cirrhosis patients randomized to colchicine *versus* placebo/no intervention.

with additional information on their trials. We also thank Libo Tao for his statistical advice. Furthermore, Dimitrinka Nikolova, Nader Salasshahri, and Styrbjørn Birch, all from The Cochrane Hepato-Biliary Group, are thanked for expert assistance during the preparation of this review. This review is published as a Cochrane Review in The Cochrane Library 2004, Issue 2. Cochrane Reviews are regularly updated as new evidence emerges and in response to comments and criticisms. The Cochrane Library should be consulted for the most recent version of the Review. This work was supported by a grant from the Copenhagen Trial Unit, Center for Clinical Intervention Research, H:S Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark and S.C. Van Foundation, Denmark.

Reprint request and correspondence: Yan Gong, The Cochrane Hepato-Biliary Group, Copenhagen Trial Unit, Center for Clinical Intervention Research, Department 7102, H:S Rigshospitalet, Copenhagen University Hospital, Blegdamsvej 9, 2100 Copenhagen, Denmark.

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Table 4. Weighted Mean Difference and Relative Risk with 95% Confidence Interval (CI) of Liver Biochemical and Histological Variables in Primary Biliary Cirrhosis Patients Randomized to Colchicine *versus* Placebo/No Intervention

	Estimates of Effect Measures	
	Weighted Mean Difference (95% CI)	
Variables	Arithmetic Mean, Trials and Patients	Geometric Mean, Trials and Patients*
Liver Biochemical Variables		
Bilirubin (μmol/l)	–1.35 (–4.52–1.82), 4 trials, n = 202	–1.55 (–2.72–1.13), 1 trial, n = 57
Alkaline phosphatases (IU/l)	–55.35 (–158.56–47.85), 4 trials, n = 200	–1.26 (–1.80–1.14), 1 trial, n = 57
Gamma-glutamyltransferase (IU/l)	–25.38 (–73.26–22.50), 4 trails, n = 200	Not estimable
Aspartate aminotransferase (IU/l)	–10.10 (–22.91–2.71), 2 trials, n = 82	Not estimable
Alanine aminotransferase (IU/l)	–2.05 (–8.79–4.68), 4 trials, n = 201	Not estimable
Albumin (g/dl)	0.09 (–0.03–0.21), 4 trials, n = 235	Not estimable
Total cholesterol (mmol/l)	0.10 (–0.88–1.08), 1 trial, n = 60	–1.02 (–1.20–1.15), 1 trial, n = 57
Immunoglobulin M (g/l)	–0.49 (–1.03–0.06), 4 trials, n = 198	Not estimable
Histological variables	Relative Risk (95% CI), Trials and patients	
Worsening of histological stage	0.85 (0.41–1.75), 3 trials, n = 145	
Worsening of histological score†	0.56 (–0.24–1.36), 1 trial, n = 50	
Worsening of piecemeal necrosis	0.58 (0.23–1.44), 2 trials, n = 95	
Worsening of parenchymal inflammation	0.69 (0.28–1.72), 2 trials, n = 95	
Worsening of parenchymal necrosis	1.00 (0.31–3.18), 2 trials, n = 95	
Worsening of fibrosis	0.60 (0.24–1.49), 2 trials, n = 95	

*Based on right skewed data.
†Weighted mean difference.

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Appendix 4

Methotrexate for primary biliary cirrhosis (Review)

Gong Y, Gluud C

This is a reprint of a Cochrane review, prepared and maintained by The Cochrane Collaboration and published in *The Cochrane Library* 2007, Issue 1

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TABLE OF CONTENTS

Methotrexate for primary biliary cirrhosis (Review)

Gong Y, Gluud C

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ABSTRACT

Background
Methotrexate, a folic acid antagonist with immunosuppressive properties, has been used to treat patients with primary biliary cirrhosis. The therapeutic responses to methotrexate in randomised clinical trials have been heterogeneous.

Objectives
To assess the beneficial and harmful effects of methotrexate for patients with primary biliary cirrhosis.

Search strategy
Relevant randomised clinical trials were identified by searching *The Cochrane Hepato-Biliary Group Controlled Trials Register* (June 2004), *The Cochrane Central Register of Controlled Trials* on *The Cochrane Library* (Issue 2, 2004), *MEDLINE* (January 1966 to August 2004), *EMBASE* (January 1980 to August 2004), and manual searches of bibliographies. We contacted authors of trials and pharmaceutical companies.

Selection criteria
Randomised clinical trials comparing methotrexate with placebo, no intervention, or another drug were included irrespective of blinding, language, year of publication, and publication status.

Data collection and analysis
Our primary outcomes were mortality and mortality or liver transplantation. Dichotomous outcomes were reported as relative risk (RR) and hazard ratio (HR) if applicable. Continuous outcomes were reported as weighted mean difference (WMD). We examined intervention effects by using both a random-effects model and a fixed-effect model. Heterogeneity was investigated by subgroup analyses and sensitivity analyses.

Main results
We identified four trials (370 patients) that compared methotrexate with placebo with or without ursodeoxycholic acid as co-intervention. One additional trial (87 patients) compared methotrexate with colchicine without and later with ursodeoxycholic acid as co-intervention. The methodological quality of the trials was low. We did not find significant effects of methotrexate on pruritus, fatigue, liver complications, liver biochemistry, liver histology, or adverse events. The pruritus score (WMD - 0.68, 95% CI - 1.11 to - 0.25), the levels of serum alkaline phosphatases (WMD - 0.41, 95% CI - 0.70 to - 0.12) and plasma immunoglobulin M (WMD - 0.47, 95% CI - 0.74 to - 0.20) were significantly lower in the patients receiving methotrexate.

Authors' conclusions
Methotrexate increased mortality in patients with primary biliary cirrhosis. We do not recommend methotrexate for patients with primary biliary cirrhosis outside randomised trials.

PLAIN LANGUAGE SUMMARY

Methotrexate tended to increase mortality or liver transplantation in patients with primary biliary cirrhosis

Primary biliary cirrhosis is an uncommon chronic liver disease of unknown etiology. Methotrexate, a folic acid antagonist with immunosuppressive properties, has been used to treat patients with primary biliary cirrhosis. However, methotrexate may increase the risk of mortality or the number of patients in need of liver transplantation. The effects of methotrexate on pruritus, fatigue, clinical complications, liver biochemistry levels, liver histology, and adverse events were not significantly different from placebo.

BACKGROUND

Primary biliary cirrhosis is an uncommon chronic liver disease of unknown aetiology. Ninety per cent of patients with primary biliary cirrhosis are females and the majority are diagnosed after the age of 40 years (James 1981). Over the past 30 years, substantial increases in prevalence of primary biliary cirrhosis were noted in the majority of studies examining longitudinal data and several have reported increases in the incidence of primary biliary cirrhosis (Kim 2000). Primary biliary cirrhosis is now a frequent cause of liver morbidity, and patients with primary biliary cirrhosis are significant users of health resources, including liver transplantation (Prince 2003).

Primary biliary cirrhosis is classically defined on the basis of the triad: antimitochondrial antibodies, found in over 95 per cent of patients with primary biliary cirrhosis (Fregeau 1989; Lacerda 1995; Invernizzi 1997; Turchany 1997; Mattalia 1998); abnormal liver function tests that are typically cholestatic (with raised activity of alkaline phosphatases being the most frequently seen abnormality); and characteristic liver histological changes (Scheuer 1967) in the absence of extrahepatic biliary obstruction (Kaplan 1996). Patients may either be diagnosed during a symptomatic phase (the common symptoms being pruritus, fatigue, jaundice, liver enlargement, signs of portal hypertension, sicca complex, and scleroderma-like lesions), in which case survival is significantly decreased, or during an asymptomatic phase, which has a relatively favourable prognosis (Beswick 1985; Balasubramaniam 1990). However, between 40 and 100 per cent of these patients will subsequently develop symptoms of primary biliary cirrhosis (Nyberg 1989; Metcalf 1996; Prince 2000).

Although the aetiology remains unknown, primary biliary cirrhosis is in many respects analogous to the graft-versus-host syndrome in which the immune system is sensitised to foreign proteins. Most primary biliary cirrhosis patients have increased class II human leukocyte antigen (HLA) histocompatibility antigen expression on bile duct cells (Ballardini 1984; Van den Oord 1986), and cytotoxic T-cells are infiltrating the bile duct epithelium (Yamada 1986). Other duct systems of the body with a high concentration of HLA class II antigens on their epithelium, such as the lacrimal and pancreatic glands, may be involved in the disease process (Epstein 1982).

Patients with primary biliary cirrhosis have been subjected to many drugs. Ursodeoxycholic acid, a bile acid, is the most extensively used drug in these patients (Verma 1999). However, we were unable to demonstrate any significant effect of ursodeoxycholic acid on mortality or liver transplantation (Gluud 2002). Over the years, a number of other drugs have been used for primary biliary cirrhosis. Earlier attempts to treat primary biliary cirrhosis using immunomodulatory and other agents such as azathioprine (Heathcote 1976; Christensen 1985), prednisolone (Mitchison 1992), chlorambucil (Hoofnagle 1986), cyclosporine (Wiesner 1990), D-penicillamine (Epstein 1981; Matloff 1982; Dickson 1985; Neuberger 1985), or colchicine (Warnes 1987; Vuoristo 1995; Poupon 1996) did not lead to widespread acceptance of these drugs for primary biliary cirrhosis patients (Kaplan 1994).

In a UK national survey of prescribing habits in primary biliary cirrhosis, 0.3% of the gastroenterologists prescribed methotrexate to patients with primary biliary cirrhosis (Verma 1999). Methotrexate is a folic acid antagonist that blocks nucleic acid synthesis. Additionally, folic acid antagonists are potent inhibitors of cell-mediated (T and B cells) immune reactions and have been employed as immunosuppressive agents, for example, in allogeneic bone marrow and organ transplantation, and for the treatment of dermatomyositis, rheumatoid arthritis, Wegener's granulomatosis, and Crohn's disease (Chu 1995; Feagan 1995). Low-dose methotrexate has immunosuppressive properties that may be mediated through inhibition of human interleukin-1 beta-induced leukocyte proliferation (Miller 1986).

Based on small pilot studies (Kaplan 1988; Bergasa 1996; Kaplan 1997), methotrexate was initially suggested as monotherapy for primary biliary cirrhosis patients since the degree of hepatic inflammation and bile duct injury improved in some patients. The degree of liver fibrosis and histological stage, however, were not improved (Bergasa 1996; Kaplan 1997). The first placebo-controlled trial of methotrexate for primary biliary cirrhosis did not support the clinical use of low-dose methotrexate (Hendrickse 1999). The addition of methotrexate did not seem to confer additional benefit in patients receiving ursodeoxycholic acid (Lindor 1995; Van Steenbergen 1996; Gonzalez-Koch 1997). We have been unable to identify meta-analyses or systematic reviews assessing the effects of methotrexate for patients with primary biliary cirrhosis.

OBJECTIVES

To assess the beneficial and harmful effects of methotrexate for primary biliary cirrhosis patients.

CRITERIA FOR CONSIDERING STUDIES FOR THIS REVIEW

Types of studies

We included randomised clinical trials irrespective of blinding, language, year of publication, or publication status. The randomised clinical trials should have used a proper method of randomisation. Thus, we excluded studies using quasi-randomisation (for example, allocation by date of birth).

Types of participants

Patients with primary biliary cirrhosis, that is, patients having at least two of the following: elevated serum activity of alkaline phosphatases (or other markers of intrahepatic cholestasis), and/or a positive result for serum mitochondrial antibody, and/or liver biopsy findings diagnostic for or compatible with primary biliary cirrhosis.

Types of intervention

Administration of any dose of methotrexate versus placebo or no intervention or other drugs. Co-interventions were allowed as long as intervention arms of the randomised clinical trial received similar co-interventions.

Types of outcome measures

Primary outcome measures were:

- Mortality.
- Mortality or liver transplantation.

Secondary outcome measures were:

- Liver transplantation.
- Pruritus: number of patients without improvement of pruritus or pruritus score.
- Fatigue: number of patients without improvement of fatigue or fatigue score.
- Liver complications: number of patients developing variceal bleeding, ascites, hepatic encephalopathy, jaundice, or hepatorenal syndrome.
- Liver biochemistry: serum (s-)bilirubin; s-alkaline phosphatases; s-gamma-glutamyltransferase; s-aspartate aminotransferase; s-alanine aminotransferase; s-albumin; s-cholesterol (total); plasma immunoglobulins.
- Liver biopsy findings: worsening of liver histological stage or score.
- Quality of life: physical functioning (ability to carry out activities of daily living such as self-care and walking around), psychological functioning (emotional and mental well-being), social functioning (social relationships and participation in social activities), and perception of health, pain, and overall satisfaction with life.
- Adverse events: The adverse event is defined as any untoward medical occurrence in a patient in either of the two arms of the included randomised clinical trials, which did not necessarily have a causal relationship with the treatment, but did, however, result in a dose reduction, discontinuation of treatment, or registration of the advent as an adverse event/side effect (ICH-GCP 1997).

SEARCH METHODS FOR IDENTIFICATION OF STUDIES

See: Cochrane Hepato-Biliary Group methods used in reviews.

We identified relevant randomised clinical trials by searching *The Cochrane Hepato-Biliary Group Controlled Trials Register* (June 2004) involving hand searches of major hepatology journals and conference proceedings, *the Cochrane Central Register of Controlled Trials* on *The Cochrane Library* (Issue 2, 2004), *MEDLINE* (January 1966 to August 2004), and *EMBASE* (January 1980 to August 2004). See Table 01 for the search strategies that we applied to the individual electronic databases.

We identified further trials by reading the reference lists of the identified studies. We wrote to the principal authors of the identified randomised clinical trials and to the researchers active in the field to enquire about additional randomised clinical trials they might know of. We also contacted the pharmaceutical companies that sponsored methotrexate in the included trials to obtain any unidentified or unpublished randomised clinical trial.

METHODS OF THE REVIEW

We performed the meta-analysis following the published protocol (Gong 2003) and the recommendations given by the Cochrane Reviewers' Handbook (Alderson 2004).

Trials selection

Two authors (YG and CG) independently evaluated whether the identified trials fulfilled the inclusion criteria. Excluded trials were listed in 'Characteristics of excluded studies' with the reasons for exclusion. Disagreements were resolved by discussion.

Data extraction

YG extracted data and CG validated the data extraction. We wrote to the authors of the included trials and asked them to specify the data of interest, if the data have not been reported clearly in the reports.

Assessment of methodological quality of included trials

The methodological quality of the randomised clinical trials was assessed using four components (Schulz 1995; Moher 1998; Kjaergard 2001):

Generation of the allocation sequence

- Adequate, if the allocation sequence was generated by a computer or random number table. Drawing of lots, tossing of a coin, shuffling of cards, or throwing dice were considered as adequate if a person who was not otherwise involved in the recruitment of participants performed the procedure;
- Unclear, if the trial was described as randomised, but the method used for the allocation sequence generation was not described.

Allocation concealment

- Adequate, if the allocation of patients involved a central independent unit, on-site locked computer, identically appearing numbered drug bottles or containers prepared by an independent pharmacist or investigator, or sealed envelopes;
- Unclear, if the trial was described as randomised, but the method used to conceal the allocation was not described;
- Inadequate, if the allocation sequence was known to the investigators who assigned participants.

Blinding

- Adequate, if the trial was described as double blind and the method of blinding involved identical placebo or active drug;
- Unclear, if the trial was described as double blind, but the method of blinding was not described;
- Not performed, if the trial was not double blind.

Follow-up

- Adequate, if the numbers and reasons for dropouts and withdrawals in all intervention groups were described or if it was specified that there were no dropouts or withdrawals;
- Unclear, if the report gave the impression that there had been no dropouts or withdrawals, but this was not specifically stated;
- Inadequate, if the number or reasons for dropouts and withdrawals were not described.

Characteristics of patients

Number of patients randomised; patient inclusion and exclusion criteria; mean (or median) age; sex ratio; histological stage; number of patients lost to follow-up.

Characteristics of interventions

Type, dose, and form of methotrexate intervention; type of intervention in the control group and collateral interventions; duration of treatment and follow-up.

Characteristics of outcomes

All outcomes were extracted from each included trial. We analysed outcomes at maximum follow-up.

Statistical methods

We used the statistical package (RevMan Analyses 1.0) provided by The Cochrane Collaboration. Regarding the time-to-event data, for example, death, we extracted hazard ratio (HR) from the trials (Parmar 1998) and pooled it by generic inverse variance. We presented dichotomous data as relative risk (RR) with 95% confidence interval (CI) and continuous outcome measures by weighted mean differences (WMD) with 95% CI.

We examined intervention effects by using both a random-effects model (DerSimonian 1986) and a fixed-effect model (Mantel 1959) with the significant level set at $P < 0.05$. If the results of the two analyses led to the same conclusion, we presented only the results of the fixed-effect analysis. In case of discrepancies of the two models, we reported the results of both models. We explored the presence of statistical heterogeneity by chi-squared test with significance set at $P < 0.10$ and measured the quantities of heterogeneity by I^2(Higgins 2002) .

Subgroup analyses

We performed subgroup analyses, in which trials were grouped according to the methodological quality of the included trials, dosage, and duration of treatment and follow-up. The high methodological quality was confined to adequate generation of the allocation sequence, allocation concealment, blinding, and follow-up. The difference between the estimates of two subgroups was estimated according to Altman 2003.

Sensitivity analyses

Regarding the primary outcome measure, that is, mortality, we included patients with incomplete or missing data in the sensitivity analyses by imputing them into following scenarios (the last four being intention-to-treat analyses) (Hollis 1999):

(1) Available case analysis: data on only those whose results are known, using as denominator the total number of patients who completed the trial;

(2) Assuming poor outcome: dropouts from both the methotrexate and control groups had the primary outcomes;

(3) Assuming good outcome: none of the dropouts from the methotrexate and control groups had the primary outcomes;

(4) Extreme case favouring methotrexate: none of the dropouts from the methotrexate-group but the dropouts from the control group had the primary outcomes;

(5) Extreme case favouring control: all dropouts from the methotrexate-group but none from the control group had the primary outcomes.

For secondary outcomes, we adopted 'available case analysis'. Therefore, in the review, the number of patients in the denominator changed according to the secondary outcomes investigated.

Bias exploration

Funnel plot was used to provide a visual assessment of whether treatment estimates were associated with study size. The performance of the available methods of detecting publication bias and other biases (Begg 1994; Egger 1997; Macaskill 2001) vary with the magnitude of the treatment effect, the distribution of study size, and whether a one- or two-tailed test is used (Macaskill 2001). Therefore, we intended to use the most appropriate method having good trade-off in the sensitivity and specificity, based on characteristics of the trials included in this review.

DESCRIPTION OF STUDIES

Search results

We identified a total of 313 references through electronic searches of *The Cochrane Hepato-Biliary Group Controlled Trials Register* (n = 16), *The Cochrane Controlled Trials Register* in *The Cochrane Library* (n = 15), *MEDLINE* (n = 137), and *EMBASE* (n = 145). We excluded 277 duplicates or clearly irrelevant references through reading abstracts. Accordingly, 36 references were retrieved for further assessment. Of these, we excluded 23 because they were non-randomised trials, reviews, or observational studies. They are listed under 'Characteristic of excluded studies' with reasons for exclusion. Accordingly, 13 references referring to five randomised clinical trials, which fulfilled our inclusion criteria, were included.

Included studies

The five randomised clinical trials were all parallel group trials. Three of them were published as full texts. The other two (Copaci 2001 and Combes 2003) were published as abstracts only.

All the included trials reported random allocation of 457 patients with primary biliary cirrhosis to:

- methotrexate versus placebo (Hendrickse 1999);
- methotrexate plus ursodeoxycholic acid versus placebo (or no intervention) plus ursodeoxycholic acid (Gonzalez-Koch 1997; Copaci 2001; Combes 2003);
- methotrexate versus colchicine (Kaplan 2004)

The mean age of patients in the included trials was 53 years and 96% of the patients were female. About half the patients had liver histological stage I/II and half had stage III/IV in the three trials which reported histological stage at entry. The entry criteria varied across trials, but were generally well-defined, making it highly likely that all patients did in fact have primary biliary cirrhosis. The dosage of methotrexate differed, that is, 7.5 mg/week, 10 mg/week, 15 mg/week, and 15 mg/m^2 body surface (maximal dose 20 mg/week). The duration of methotrexate treatment varied from 48 weeks to 10 years, and the median duration was six years. Details are listed in the table of Characteristics of included studies.

METHODOLOGICAL QUALITY

The methods to generate the allocation sequence and to conceal allocation were considered adequate in two trials (Hendrickse 1999; Kaplan 2004) and unclear in the other three. Blinding was adequate in two trials (Hendrickse 1999; Kaplan 2004) and not performed in the other three. It was stated in Hendrickse 1999 and Kaplan 2004 that a single study monitor was responsible for allocating patients to treatment and control group. The adequate description of follow-up was reported in three trials (Gonzalez-Koch 1997; Hendrickse 1999; Kaplan 2004) and inadequate/unclear in the other two. There was a fairly low rate of missing data across the trials included.

RESULTS

Methotrexate

Mortality (Comparison 01-01)

Two trials (Gonzalez-Koch 1997; Hendrickse 1999) showed that methotrexate had a significantly detrimental effect on mortality (RR 5.00, 95% CI 1.19 to 20.92). The sensitivity analyses did not significantly change the estimate.

Mortality or liver transplantation (Comparison 01-02 to 01-04)

We pooled the estimate of HR from Hendrickse 1999 and Combes 2003 to achieve the overall effect on survival plus liver transplantation (HR 1.44, 95% 0.46 to 4.54, random effects; HR 1.18, 95% 0.64 to 2.16, fixed effect, I^2 = 63.0%). In the six-year Hendrickse 1999 trial, 11/30 patients in the methotrexate group died or underwent liver transplantation versus 7/30 patients in the placebo group. There was no death and/or liver transplantation in the 48-week Gonzalez-Koch 1997 trial. Combining the results of the two trials could not demonstrate significant effect of methotrexate on mortality or liver transplantation (RR 1.57, 95% CI 0.71 to 3.50). This estimate was robust since all the scenario sensitivity analyses did not significantly change the estimate.

Pruritus, fatigue, and jaundice (Comparison 01-05 to 01-07)

Hendrickse 1999 reported pruritus score in each group, and Gonzalez-Koch 1997 reported the number of patients without improvement of pruritus. Both results did not show any significant difference between methotrexate and placebo group (WMD -0.17 score, 95% CI -0.63 to 0.29; RR 6.50, 95% CI 0.37 to 114.12). Gonzalez-Koch 1997 also reported the 'number of patients without improvement of fatigue' and showed no significant difference between methotrexate and placebo groups (RR 0.92, 95% CI 0.06 to 13.18). We were not able to extract data on jaundice.

Liver complications (Comparison 01-08)

Overall, no significant difference was detected on occurrence of liver complications between the methotrexate and placebo group (RR 0.83, 95% CI 0.28 to 2.44). Hendrickse 1999 reported five

patients developing oesophageal varices in the methotrexate group versus six patients in the placebo group.

Liver biochemistry (Comparison 01-09, 01-10)

Neither Gonzalez-Koch 1997 nor Hendrickse 1999 trials provided means and standard deviations for biochemical variables, so it was impossible to pool them in meta-analyses except for serum albumin and prothrombin time. The level of serum albumin or the prothrombin time in the methotrexate group were not significantly different from that in the placebo group (WMD -0.90 g/dl, 95% CI -3.45 to 1.65 and WMD 0.90 second, 95% CI -1.24 to 3.04).

In the Gonzalez-Koch 1997 trial, patients receiving methotrexate plus ursodeoxycholic acid showed a tendency to a larger absolute decrease in immunoglobulin M concentrations and activity of alkaline phosphatases than the patients receiving placebo plus ursodeoxycholic acid. In the methotrexate plus ursodeoxycholic acid group, changes in bilirubin were as follows: in nine patients the level decreased; in two it increased; and in two there was no change. In the placebo plus ursodeoxycholic acid group, bilirubin levels decreased in nine patients and increased in three. The changes from the initial values in albumin and prothrombin time were not significant in either group.

In the Hendrickse 1999 trial, on-treatment serum activities of alkaline phosphatases and gamma-glutamyltransferase and immunoglobulin M and immunoglobulin G were significantly lower in the methotrexate than the placebo group. The serum bilirubin, albumin levels, prothrombin time, and Mayo score deteriorated with time in both groups and the deterioration tended to be greater in the methotrexate group.

Liver biopsy findings (Comparison 01-11, 01-12)

Gonzalez-Koch 1997 found there was no significant improvement in the methotrexate group compared to the placebo group on any of the histologic variables, for example, worsening of histological stage (RR 1.62, 95% CI 0.63 to 4.16), cholestasis (RR 0.31, 95% CI 0.04 to 2.57), or ductular proliferation (RR 0.74, 95% CI 0.26 to 2.12). Ludwig stage, one of the histological variables measured in Hendrickse 1999, was not significantly different between methotrexate and placebo group either after two years (RR -0.61, 95% CI -1.25 to 0.03) or after four to six years.

Quality of life and cost-effectiveness

None of the trials examined quality-of-life scales or cost-effectiveness.

Adverse events (Comparison 01-13)

Hendrickse 1999 reported that 26 patients developed adverse events in the methotrexate group versus 25 in the placebo group. Gonzalez-Koch 1997 summarised that 11 patients in the methotrexate and none in the placebo group developed adverse events. The I^2 is 92.9%. Given the description of adverse events by the authors, it is not possible for us to classify them into non-serious and serious ones.

We also evaluated the harmful effects from observational studies. We identified eight studies and listed all the adverse events in Table 02. Interstitial pneumonitis, aphthous ulcers, transient abdominal discomfort, minor dyspeptic symptoms and mucositis, and marrow depression were most reported as adverse events.

Methotrexate (Comparison 02-01 to 02-14)

Kaplan 2004 compared methotrexate versus colchicine for two years. They observed that 11/42 patients died or underwent liver transplantation in the methotrexate group versus 9/43 patients in the colchicine group (RR 1.25, 95% CI 0.58 to 2.71). The pruritus score was significantly lower in patients receiving methotrexate than than those receiving colchicine (WMD = - 0.68, 95% CI -1.11 to -0.25). Regarding liver biochemical outcomes, the activity of s-alkaline phosphatases and the concentration of plasma immunoglobulin M were significantly lower in the methotrexate group than in the colchicine group (WMD - 0.41, 95% CI -0.70 to -0.12 and WMD -0.47, 95% CI -0.74 to -0.20). For other outcomes, no significant differences were detected.

Bias exploration

We did not perform funnel plot and did not apply the three statistical methods to detect publication bias and other biases because the power of those would have been low and inconsistent due to the small number of included trials.

DISCUSSION

Evidence showed that methotrexate tended to increase mortality or liver transplantation in patients with primary biliary cirrhosis. We advise that any new trials with methotrexate for patients with primary biliary cirrhosis should monitor harmful effects closely and that methotrexate should not be used outside randomised trials.

The systematic review has several limitations. Only four trials involving 370 patients were analysed on methotrexate versus placebo or no intervention. This is a low number of patients (Ioannidis 2001). None of the trials reported a sample size estimate. The methodological trial quality was generally low, which may significantly overestimate intervention effects (Schulz 1995; Moher 1998; Kjaergard 2001). If the same overestimation is valid for the present sample of trials, the prospects for methotrexate for primary biliary cirrhosis look even worse than our results may indicate. On the other hand, we cannot preclude that methotrexate may have beneficial effects in certain patient groups. Further, half of the trials have shorter follow-up than the estimated median survival of 10 to 15 years (Prince 2002). Therefore, it is difficult to detect a significant difference on mortality.

Combining the results of Gonzalez-Koch 1997 and Hendrickse 1999, we found significant detrimental effect of methotrexate on mortality. The sensitivity analyses (intention-to-treat analysis us-

ing imputation) showed that the estimate was not changed under the different imputations.

Methotrexate tended to increase the risk of mortality or liver transplantation in patients with primary biliary cirrhosis. Heterogeneity across the two included trials (Hendrickse 1999; Combes 2003) was considerable (I^2 = 63.0%). The possible explanation could be different follow-up periods (six years and 10 years), methotrexate was given with or without co-intervention (ursodeoxycholic acid was added to both intervention arms of the Combes 2003 trial), etc. The use of composite outcomes is much debated (Lubsen 2002). The referrals for liver transplantation occurred mainly after the blinding of the randomised clinical trials had been removed. Unblinded comparisons may exaggerate intervention efficacy significantly (Schulz 1995; Kjaergard 2001). Thus, the risk of mortality and/or liver transplantation due to methotrexate may actually be worse than observed in the review.

Methotrexate did not show any significant effect on pruritus, fatigue, or liver complications when compared with placebo. When compared with colchicine (Kaplan 2004), methotrexate seemed to improve the severity of pruritus (WMD -0.68, 95% CI -1.11 to -0.25). This finding needs further confirmation. First, this finding was from one trial with only 87 patients. Second, due to the large number of statistical comparisons having been performed some of the comparisons might have come out with a significant difference simply due to the repetitive testing ('mass significance').

In a non-randomised study, Bergasa et al reported that methotrexate increased the number of patients with fibrosis (Bergasa 1996). Kaplan et al reported, in another non-randomised study, that methotrexate slowed the progression of primary biliary cirrhosis (Kaplan 1997). Based on the included trials (Gonzalez-Koch 1997; Hendrickse 1999), we did not observe any significant improvement in the liver histologic variables in the methotrexate group. For example, Knodell inflammatory score, Knodell fibrosis score, Ludwig stage, and bile duct/portal tract ratio, were not significantly different between the methotrexate and the placebo groups, neither after two years nor after four to six years.

The total dose in the Gonzalez-Koch 1997 trial was 10 mg/week, while in Hendrickse 1999 was 7.5 mg/week. Failure to show a beneficial effect of methotrexate on clinical, biochemical, or histologic evolution may point to insufficient dose of methotrexate. Since there was no death and/or liver transplantation in Gonzalez-Koch 1997, it is impossible to do a subgroup analysis by dosage. In an unblinded study (Conjeervaram 1995), patients given 7.5 mg and 15 mg methotrexate per week had similar biochemical and symptomatic responses.

The analysis of adverse events showed substantial heterogeneity (I^2 = 92.9%). Hendrickse 1999, a six-year randomised clinical trial applying methotrexate with 7.5 mg/week, observed a non-significant difference in adverse events. However, in Gonzalez-Koch 1997 trail with the dosage of 10 mg/week and 48-week follow-up, 11/13 patients given methotrexate did develop adverse reactions while 0/12 patients given placebo complained of any adverse reactions. This contrast might be because the toxicity of methotrexate could appear in the early response and well-tolerated in the later long period. It might also be because the two trials applied different definitions to adverse events.

We evaluated the safety issue in the relevant non-randomised studies and observational studies. We found that interstitial pneumonitis, aphthous ulcers, transient abdominal discomfort, minor dyspeptic symptoms, and mucositis and marrow depression were the most frequent adverse events. In our sample of the included trials, we could not rule out the hepatotoxity of methotrexate because of the liver-cirrhosis-related nature of primary biliary cirrhosis and its reflection on the tendency of decreased survival or liver transplantation. Four Cochrane reviews have examined the hepatotoxic effect of methotrexate in patients with asthma (Davies 2001), rheumatoid arthritis (Suarez-Almazor 1997), juvenile idiopathic arthritis (Takken 2001), and Crohn's disease (Alfadhli 2003). Further, a narrative review on patients with psoriasis (Tang 1996) has also explored the heptatotoxicity of methotrexate. Hepatotoxicity was a common adverse effect with methotrexate (8 trials using 15 mg/week, 1 using 30 mg/week) compared to placebo in adults with asthma (RR 6.61, 95% CI 2.36 to 18.53, 9 trials) (Davies 2001). The most common cause for discontinuation of methotrexate (7.5 to 15 mg/week) in patients with rheumatoid arthritis was the presence of liver enzyme abnormalities (RR 4.45, 95% CI 1.57 to 12.66, 5 trials) (Suarez-Almazor 1997). The RR for overall withdrawals from methotrexate therapy (10 to15 mg/m^2/week) for patients with juvenile idiopathic arthritis was 1.60 (95% CI 0.66 to 3.87, two trials) compared to placebo, suggesting there is no difference between methotrexate and placebo in terms of harmful effects. However, the type of adverse effects was not specified (Takken 2001). The adverse effects in methotrexate patients (12.5 mg, 22.5 mg, 25 mg per week) with refractory Crohn's disease were significantly more common than with placebo (RR 6.97, 95% 1.61 to 30.10, three trials) (Alfadhli 2003). The most common reasons for withdrawal were nausea and vomiting and asymptomatic elevation of liver enzymes. The evidence is sufficiently strong to support that in psoriatic patients, low-dose methotrexate may be hepatotoxic, eventually leading to hepatic fibrosis and cirrhosis (Tang 1996). More information is needed about the methotrexate-induced hepatotoxity. If the problem is limited to an increase of biological parameters that disappear when the drug is stopped, it is manageable, but if there is clinical complication related to the hepatoxicity or major histological lesion the issue is different.

We have also systematically reviewed the effects of other immunosuppressants compared with placebo/no intervention, that is, colchicine and D-penicillamine on survival. We did not find evidence either to support or refute the use of colchicine for patients with primary biliary cirrhosis (Gong 2004a). D-penicillamine did not reduce the risk of mortality, and it significantly increased the

occurrences of adverse events (Gong 2004b).

AUTHORS' CONCLUSIONS

Implications for practice

Evidence showed that methotrexate increased mortality in patients with primary biliary cirrhosis from two long-period randomised clinical trials. We do not advocate the use of methotrexate for patients with primary biliary cirrhosis.

Implications for research

Although the majority of the evidence did not point to a beneficial effect of methotrexate for patients with primary biliary cirrhosis we are not able to exclude the possibility for a beneficial effect in certain patient groups. We advise that any new placebo-controlled trials with methotrexate for patients with primary biliary cirrhosis should monitor harmful effects closely. Further trials on patients with primary biliary cirrhosis ought to be conducted and reported according to the CONSORT guidelines (www.consort-statement.org)

FEEDBACK

Methotrexate for primary biliary cirrhosis

Summary

Date of Submission: 12-Jun-2006
Name: Roger Pepin
Email Address: r.pepin@elsevier.com
Personal Description: Occupation EBM Editor

Feedback: I find sentence three and sentence four in the Main Results section rather confusing and contradictory, saying in the first instance "We did not find significant effects of methotrexate on pruritus...,liver biochemistry.." and then following this with " The pruritus score..and levels of serum alkaline phosphatases...were significantly lower in the patients receiving methotrexate." I hope this can be clarified in subsequent revisions.

Submitter agrees with default conflict of interest statement:
I certify that I have no affiliations with or involvement in any organization or entity with a financial interest in the subject matter of my feedback.

Author's reply

We are very sorry for this inaccuracy. 'The pruritus score..and levels of serum alkaline phosphatases...were significantly lower in the patients receiving methotrexate' should be changed into: 'The pruritus score..and levels of serum alkaline phosphatases...were significantly lower in the patients receiving methotrexate than those receiving colchicine.' We wrote this clearly in results section, but we forgot to add 'than colchicine' in the 'main results' in the abstract of the review.

Contributors

Yan Gong, primary author
23 August 2006
Copenhagen, Denmark

Christian Gluud, author and Criticism Editor
23 August 2006
Copenhagen, Denmark

POTENTIAL CONFLICT OF INTEREST

None known.

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* *Indicates the major publication for the study*

TABLES

Characteristics of included studies

Study	**Combes 2003**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: unclear. Blinding: unclear. Follow-up: unclear.
Participants	Country: USA. Mean age: not reported. Female/Male: not reported. PBC stage status: stage I or II: 62 in MTX group, 64 in placebo group. stage III or IV: 70 in MTX group, 69 in placebo group. Inclusion criteria: 1. AMA positive PBC patients without ascites, variceal bleeding, or encephalopathy. 2. A serum bilirubin less than 3 mg/dl prior to and while on UDCA before randomization and serum albumin 3 g/dl or greater. 3. On UDCA 15 mg/kg/day for at least 6 months. Exclusion criteria: 1. Other forms of liver disease.
Interventions	a) MTX plus UDCA (n = 132): 15 mg/sq. m. body surface area (maximal dose 20 mg) once a week while UDCA is administered as 15 mg/kg/day. b) Placebo plus UDCA (n = 133): no description of placebo. The mean duration of therapy has been 89.4 months - the longest duration was117 months.
Outcomes	1. Death without liver transplantation. 2. Liver transplantation. 3. Variceal bleeding. 4. Development of ascites, encephalopathy, varices. 5. A doubling of bilirubin to 2.5 mg/dl or greater, a fall in serum albumin to 2.5 mg/dl or less. 6. Histologic progression by 2 stages or to cirrhosis. 7. Voluntary discontinuation of and/or inability to tolerate study medication.
Notes	1. All the patients were treated with UDCA for six months before randomization. 2. The trial was conducted with a stopping rule and was stopped early by their Data Monitoring Board for futility. 3. It was an abstract. Sent letter (7 March 2003). Dr. Combes replied that the manuscript was being written for publication.
Allocation concealment	B – Unclear

Study	**Copaci 2001**
Methods	Generation of the allocation sequence: unclear.

Characteristics of included studies (*Continued*)

	Allocation concealment: unclear. Blinding: not performed. Follow-up: unclear.
Participants	Country: Romania. Mean age: not reported. Female/Male: not reported. PBC stage status: not reported. Inclusion criteria were: elevated ALP, positive AMA, liver histology. Patients with bilirubin higher than 3 mg/dL and or decompensated liver disease were excluded.
Interventions	a) MTX plus UDCA (n = 8); b) UDCA (n = 12) Mean follow-up was 5.0 ± 1.8 years.
Outcomes	Major outcomes were the development of liver decompensation and liver related death.
Notes	1. Only published as an abstract. 2. Sent letter (14 Mar. 2003), but no reply has been received.
Allocation concealment	B – Unclear

Study	**Gonzalez-Koch 1997**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: unclear. Blinding: unclear. Follow-up: adequate, none of patients in both groups were lost to follow-up.
Participants	Country: Chile. Mean age: 49 ± 8 years in MTX plus UDCA group; 52 ± 13 years in placebo plus UDCA group. Female/Male: 25/0. PBC stage status: stage I or II: 8 in MTX group, 5 in placebo group. stage III: 5 in MTX group, 7 in placebo group. Inclusion criteria: 1. ALP or GGT levels at least 1.5 times the normal upper limit. 2. A positive AMA test 3. Biopsy-proved PBC. Exclusion criteria: 1. Feature suggestive of other concomitant liver or biliary disease. 2. Decompensated cirrhosis. 3. Presense of other serious diseases. 4. The need to use additional medications. 5. Pregnancy. 6. Any pharmacological therapy during the previous six months.
Interventions	MTX plus UDCA (n = 13): MTX: weekly oral pulse therapy in four doses over 48 hours (total dose 10 mg/week) UDCA: 250 mg twice a day; Placebo plus UDCA (n=12): Duration of medication: 48 weeks.
Outcomes	1. Liver biochemical variables. 2. Major clinical outcomes: death, liver transplantation, etc.

Characteristics of included studies (*Continued*)

	3. Liver biopsy findings. 4. Clinical symptoms: fatigue, pruritus, etc.
Notes	1. Sent letter (14 March 2003), but no reply has been received.
Allocation concealment	B – Unclear

Study	**Hendrickse 1999**
Methods	Generation of the allocation sequence: adequate, a random-number technique. Allocation concealment: adequate, hospital pharmacy. Blinding: adequate, double-blinding using an identical placebo. Follow-up: adequate, 1 in MTX group and 0 in placebo group were lost to follow-up.
Participants	Country: England. Mean age: 57±1.8 years in MTX group, 57± 1.7 years in placebo group. Female/Male: 55/5. PBC stage I/II/III/IV: 11/7/13/7 in MTX group; 10/7/13/6 in placebo group. Inclusion criteria: 1. Results of liver biopsy consistent with PBC. 2. Exclusion of extrahepatic obstruction. 3. At least one of the following laboratory abnormalities: cholestatic pattern of liver enzyme abnormalities persisting over several months; serum positive for AMA; increased serum Ig M level. Exclusion criteria: 1. Advanced liver disease. 2. Continuing or recent alcohol abuse. 3. Immunosuppressive drugs received in the proceeding six months. 4. Contemplation of pregnancy. 5. Hematologic abnormalities. 6. Other serious medical illness.
Interventions	a) MTX (n = 30): 7.5 mg/week. b) Placebo (n = 30): identical placebo. Duration of medication: six years.
Outcomes	1. Major clinical outcomes: death, liver transplantation, etc. 2. Liver biochemical variables. 3. Mayo score. 4. Clinical outcomes: pruritus, fatigue, complications of liver disease, etc. 5. Liver biopsy findings. 6. Adverse events.
Notes	1. Sent letter (8 March 2003), but no reply has been received.
Allocation concealment	A – Adequate

Study	**Kaplan 2004**
Methods	Generation of the allocation sequence: adequate, computer-generated list. Allocation concealment: adequate, a single study monitor. Blinding: adequate, double-blinding and double-dummy. Follow-up: inadequate.
Participants	Country: USA. Mean age: 51 ± 1.4 years in colchicine group, 51 ± 1.5 years in methotrexate group. Female/Male: 82/3. PBC stage IV: 23 in colchicine group, 16 in methotrexate, others unknown.

	Inclusion criteria: 1. Serum ALP level of at least 2 times greater than the upper limit of normal. 2. Serum bilirubin level not greater than 10 mg/dL. 3. Liver biopsy performed consistent with PBC. 4. Radiological or ultrasonic evidence.
Interventions	a) Methotrexate: 15 mg/week, 5 mg every 12 hours 3 times. b) Colchicine: 0.6 mg colchicine twice daily. Duration of the medication: 2 years
Outcomes	1. Biochemical variables. 2. IgM. 3. Puritus and fatigue. 4. Liver histological evidence.
Notes	1. It is the interim analysis of the ten-year trial starting in 1988. 2. 2/87 withdrew from the trial immediately after randomisation before they received any drugs, did not return for follow-up testings, and were not included in the analyses. Ten patients dropped out of the trial. The reasons were specified, but the number in each group was not given. 3. Sent letter (4 November 2002). M. Kaplan responded, but did not provide additional information.
Allocation concealment	A – Adequate

UDCA: ursodeoxycholic acid
MTX: methotrexate
AASLD: American Association for the Study of Liver Diseases
ALP: alkaline phosphatases
AMA: antimitochondrial antibody
GGT: gamma-glutamyltransferase
PBC: primary biliary cirrhosis
Ig: immunoglolubin

Characteristics of excluded studies

Study	Reason for exclusion
Bach 1997	A case series. A total of 48 complete liver biopsies were obtained from 68 patients with PBC before methotrexate was started and after two years of therapy.
Bergasa 1996	A case series of ten patients with PBC treated with methotrexate.
Bonis 1999	A case series with ten patients with PBC.
Buscher 1993	A case series of eight patients with PBC. Methotrexate (2.5mg/day) was given to eight female patients with PBC treated with UDCA (10.15mg/kg per day).
Hoofnagle 1991	A case report of nine PBC patients treated with methotrexate for 12 months.
Kaplan 1988	A case report of two PBC cases.
Kaplan 1992	A historically controlled clinical study. It contained two before-after comparisons (with no randomisation) in order to evaluate whether combination therapy (UDCA plus methotrexate) was more effective than the individual drugs given alone.
Kaplan 1997	A case series of 19 patient with PBC treated with methotrexate, 15 mg/wk.
Lindor 1995	A historically controlled clinical study. Thirty-two patients with PBC were entered into a pilot study and received methotrexate plus UDCA. The results of this treatment were compared with those obtained from 180 patients with PBC studied in a placebo-controlled trial of UDCA alone.
Van Steenbergen 1992	A historically controlled clinical study. Thirteen patients were treated with methotrexate treatment for one year to assess its biochemical and histological efficacy before and after.
Van Steenbergen 1994	A pilot study comparing methotrexate with methotrexate with ursodeoxycholic acid.

Characteristics of excluded studies (*Continued*)

Van Steenbergen 1995	A presentation on the mechanisms of methotrexate in PBC at the Rotterdam liver day.
Van Steenbergen 1996	An open label randomised clinical trial. It compared the clinical, biochemical and histologic evolution in six untreated patients with those in eight patients treated with methotrexate in association with UDCA.
Vandeputte 1997	A retrospective cohort study. It reviewed 20 non-cirrhotic PBC patients who received methotrexate association with UDCA.

PBC: primary biliary cirrhosis
UDCA: ursodeoxycholic acid

ADDITIONAL TABLES

Table 01. Search strategy for identification of studies

Database	Time of search	Searched items
The Cochrane Hepato-Biliary Group Controlled Trials Register	Jan 2004	#1 = 'RCT' and ' PRIMARY BILIARY CIRRHOSIS' and ' METHOTREXATE'
The Cochrane Library (CENTRAL)	Issue 1, 2004	#1 = LIVER-CIRRHOSIS-BILIARY*: MESH #2 = (PRIMARY and BILIARY and CIRRHOSIS) or PBC #3 = METHOTREXATE: MESH #4 = IMMUNOSUPPRES* : MESH #5 = URSODEOXYCHOLIC-ACID: MESH #6 = METHOTREXATE or IMMUNOSUPPRES* or (URSODEOXYCHOLIC and ACID) #7 = #3 or #4 or #5 or #6 #8 = (#1 and #7) #9 = (#2 and #7) #10 = (#8 or #9)
MEDLINE	January 1966 to Jan 2004	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 = (PRIMARY and BILIARY and CIRRHOSIS) or PBC #3 = "PRIMARY BILIARY CIRRHOSIS" or PBC #4 = #2 or #3 #5 = METHOTREXATE #6 = IMMUNOSUPPRES* #7 = URSODEOXYCHOLIC #8 = ACID #9 = #5 or #6 or (#7 and #8) #10 = METHOTREXATE: MESH #11 = IMMUNOSUPPRESS*: MESH #12 = URSODEOXYCHOLIC-ACID: MESH #13 = #9 or #10 or #11 or #12 #14 = #1 and #13 #15 = #4 and #13 #16 = #14 or #15 #17 = random* #18 = placebo* #19 = blind* #20 = meta-analysis #21 = #17 or #18 or #19 or #20

Table 01. Search strategy for identification of studies (*Continued*)

Database	Time of search	Searched items
		#22 = #16 and # 21
EMBASE	January 1980 to Jan 2004	#1 = PRIMARY-BILIARY-CIRRHOSIS: MESH #2 = (PRIMARY and BILIARY and CIRRHOSIS) or PBC #3 = "PRIMARY BILIARY CIRRHOSIS" or PBC #4 = #2 or #3 #5 = METHOTREXATE #6 = IMMUNOSUPPRES* #7 = URSODEOXYCHOLIC #8 = ACID #9 = #5 or #6 or (#7 and #8) #10 = METHOTREXATE: MESH #11 = IMMUNOSUPPRESS*: MESH #12 = URSOXYCHOLIC-ACID: MESH #13 = #9 or #10 or #11 or #12 #14 = #1 and #13 #15 = #4 and #13 #16 = #14 or #15 #17 = random* #18 = placebo* #19 = blind* #20 = meta-analysis #21 = #17 or #18 or #19 or #20 #22 = #16 and # 21

Table 02. Adverse Events (AE) in patients (pts) with PBC

Study	Pts. in experimental	Pts. in control	AE in experimental	AE in control	Author's conclusion
Combes 1998	132	131	No information.	No information.	Frequency of respiratory adverse events was similiar in both groups of patients.
Gonzalez-Koch 1997	13	12	Diarrhea (5 pts), abdominal discomfort (4 pts), vomiting (4 pts), nausea (2 pts), glossitis (1 pt).	No AE.	The combination of ursodeoxycholic acid plus methotrexate was not associated with substantial adverse effects, which may be due to the lower dose of methotrexate used.
Hendrickse 1999	30	30	Fatal variceal bleed (1 pt), worsening liver function (5 pts), shortness of breath	Worsening liver function (5 pts), shortness of breath (5 pts), recurrent	Although we found no evidence that methotrexate accelerated fibrosis,

Table 02. Adverse Events (AE) in patients (pts) with PBC (*Continued*)

Study	Pts. in experimental	Pts. in control	AE in experimental	AE in control	Author's conclusion
			(2 pts), recurrent infection (3 pts), alopecia (3 pts), marrow depression (5 pts), nausea (1pt), depression (2 pts), extrahepatic disease (3 pts).	infection (1 pt), marrow depression (3 pts), nausea (3 pts), depression (2 pts), extrahepatic disease (3 pts).	it is possible that methotrexate aggravated liver damage, with or without fibrosis in some patients.
Kaplan 1999	43	42	Interstitial pneumonitis (6 pts).	No information.	We believe that interstitial pneumonitis is a potential problem with methotrexate and its prevalence will be approximately 1 to 3%.
Bach 1997	68	0	Recurrent mouth uclers (1 pt), acceleration bone loss (1 pt).	Not applicable.	A positive finding of this study was lack of evidence of methotrexate hepatotoxicity at least at the two-year mark or after a cumulative dose of methotrexate ranging between 1.5 to 2 g.
Bergasa 1996	10	0	Minor dyspeptic symptoms and mucositis (10 pts), aphthous uclers (2 pts), pneumonitis (1 pt), lymphadenopathy (1 pt).	Not applicable.	No comments.
Bonis 1999	10	0	Oral aphthous ulcers (1 pt), thrombocytopenia and leukopenia (1 pt).	Not applicable.	The long-term safety of methotrexate in the treatment of primary biliary cirrhosis has not been established.
Buscher 1993	8	0	Increased fatigue (7 pts), transient abdominal discomfort (2 pts).	Not applicable.	Ursodeoxycholic acid treatment may be advantageous in the first four

Table 02. Adverse Events (AE) in patients (pts) with PBC (*Continued*)

Study	Pts. in experimental	Pts. in control	AE in experimental	AE in control	Author's conclusion
					to six weeks of methotrexate therapy due to its liver-protecting effects. The combination of ursodeoxycholic acid and methotrexate indicates an acceptable risk/benefit ratio with the respect to methotrexate-induced hepatotoxity.
Kaplan 1992	9	5	No information.	No information.	Ursodeoxycholic acid and methotrexate alone and in combination were well tolerated by all patients.
Kaplan 1997	19	0	Pancytopenia (1 pt), complications of diabetes mellitus (1 pt), thrombocytopenia (1 pt), leukopenia and thrombocytopenia (1 pt), intention to become pregnant (1 pt), desire to use only herbal medicine (1 pt).	Not applicable.	Methotrexate must still be used with caution in patients with primary biliary cirrhosis because it is not well tolerated by all patients. Hepatotoxicity seems unlikely in these patiens. Reversible bone marrow suppression and interstitial pneumonitis are potential problems.
Lindor 1995	32	0	Pulmonary toxicity (4 pts), mouth ulcer (1 pt), hair loss (1 pt), gastrointestinal upset (2 pts).	Not applicable.	In our patients with primary biliary cirrhosis, we were unable to detect any evidence of accelerated hepatic fibrosis.
Van Steenbergen 1992	13	0	Severe pneumonitis (1 pt).	Not applicable.	Oral low pulse dose treatment with methotrexate is generally well

Table 02. Adverse Events (AE) in patients (pts) with PBC (*Continued*)

Study	Pts. in experimental	Pts. in control	AE in experimental	AE in control	Author's conclusion
					tolerated.

ANALYSES

Comparison 01. Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Mortality	2	85	Relative Risk (Fixed) 95% CI	5.00 [1.19, 20.92]
02 Mortality or liver transplantation (expressed as relative risk)	2	85	Relative Risk (Fixed) 95% CI	1.57 [0.71, 3.50]
03 Mortality or liver transplantation (expressed as hazard ratio) - random effects	2	325	Hazard ratio (Random) 95% CI	1.44 [0.46, 4.54]
04 Mortality or liver transplantation (expressed as hazard ratio) - fixed effect	2	325	Hazard ratio (Fixed) 95% CI	1.18 [0.64, 2.16]
05 Number of patients without improvement of pruritus	1	25	Relative Risk (Fixed) 95% CI	6.50 [0.37, 114.12]
06 Pruritus score	1	60	Weighted Mean Difference (Fixed) 95% CI	-0.17 [-0.63, 0.29]
07 Number of patients without improvement of fatigue	1	25	Relative Risk (Fixed) 95% CI	0.92 [0.06, 13.18]
08 Number of patients developing liver complications	2	85	Relative Risk (Fixed) 95% CI	0.83 [0.28, 2.44]
09 S-albumin (g/dl)	1	57	Weighted Mean Difference (Fixed) 95% CI	-0.90 [-3.45, 1.65]
10 Prothrombin time (second)	1	52	Weighted Mean Difference (Fixed) 95% CI	0.90 [-1.24, 3.04]
11 Liver biopsy findings - dichotomous variables			Relative Risk (Fixed) 95% CI	Subtotals only
12 Liver biopsy findings - histological stage	1	42	Weighted Mean Difference (Fixed) 95% CI	-0.61 [-1.25, 0.03]
13 Adverse events			Relative Risk (Random) 95% CI	Subtotals only

Comparison 02. Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Mortality or liver transplantation			Relative Risk (Fixed) 95% CI	Subtotals only
02 Pruritus score			Weighted Mean Difference (Fixed) 95% CI	Subtotals only
03 Fatigue score			Weighted Mean Difference (Fixed) 95% CI	Subtotals only
04 S-bilirubin (µmol/L) (presented as log scaled)			Weighted Mean Difference (Fixed) 95% CI	Subtotals only
05 S-alkaline phosphatases (ALP)(IU/L) (presented as log scaled)			Weighted Mean Difference (Fixed) 95% CI	Subtotals only

06 S-aspartate aminotransferase (AST)(IU/L) (presented as log scaled)	Weighted Mean Difference (Fixed) 95% CI	Subtotals only
07 S-alanine aminotransferase (ALT)(IU/L) (presented as log scaled)	Weighted Mean Difference (Fixed) 95% CI	Subtotals only
08 S-albumin (g/dl)	Weighted Mean Difference (Fixed) 95% CI	Subtotals only
09 S-cholesterol (total) (mmol/L) (presented as log scaled)	Weighted Mean Difference (Fixed) 95% CI	Subtotals only
10 Plasma immunoglobulin M (g/L) (presented as log scaled)	Weighted Mean Difference (Fixed) 95% CI	Subtotals only
11 Prothrombin time (second)	Weighted Mean Difference (Fixed) 95% CI	Subtotals only
12 Liver biopsy findings - histological stage	Weighted Mean Difference (Fixed) 95% CI	Subtotals only
13 Liver biopsy findings - histological score	Weighted Mean Difference (Fixed) 95% CI	Subtotals only
14 Adverse events	Relative Risk (Fixed) 95% CI	Subtotals only

INDEX TERMS

Medical Subject Headings (MeSH)

Folic Acid Antagonists [adverse effects; *therapeutic use]; Immunosuppressive Agents [adverse effects; *therapeutic use]; Liver Cirrhosis, Biliary [*drug therapy; mortality]; Liver Transplantation; Methotrexate [adverse effects; *therapeutic use]; Randomized Controlled Trials

MeSH check words

Humans

COVER SHEET

Title	Methotrexate for primary biliary cirrhosis
Authors	Gong Y, Gluud C
Contribution of author(s)	YG drafted the protocol, performed the searches, selected trials for inclusion, wrote to authors and pharmaceutical companies, performed data extraction and data analyses, and drafted systematic review. CG formulated the idea of this review and revised the protocol, selected trials for inclusion, solved discrepancy of data extraction, validated data analysis, and revised the review.
Issue protocol first published	2003/3
Review first published	2005/3
Date of most recent amendment	23 August 2006
Date of most recent SUBSTANTIVE amendment	14 March 2005
What's New	Measurement of serum immunoglobulins generally reveals an elevated immunoglobulin M value. Therefore, we have chosen it to replace immunoglobulin to which we referred in the published protocol with the biomedical outcome measure 'Plasma immnunoglobulin M'.
Date new studies sought but none found	27 January 2004

Date new studies found but not yet included/excluded	Information not supplied by author
Date new studies found and included/excluded	27 January 2004
Date authors' conclusions section amended	15 March 2004
Contact address	Dr Yan Gong Copenhagen Trial Unit Centre for Clinical Intervention Research, Copenhagen University Hospital Dept. 7102, Blegdamsvej 9 H:S Rigshospitalet Copenhagen DK-2100 DENMARK E-mail: ygong@ctu.rh.dk Tel: +45 3545 7161 Fax: +45 3545 7101
DOI	10.1002/14651858.CD004385.pub2
Cochrane Library number	CD004385
Editorial group	Cochrane Hepato-Biliary Group
Editorial group code	HM-LIVER

GRAPHS AND OTHER TABLES

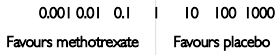
Analysis 01.01. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 01 Mortality

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 01 Mortality

Study	Methotrexate n/N	Placebo n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Methotrexate versus placebo					
Hendrickse 1999	10/30	2/30		100.0	5.00 [1.19, 20.92]
Subtotal (95% CI)	30	30		100.0	5.00 [1.19, 20.92]
Total events: 10 (Methotrexate), 2 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect z=2.20 p=0.03					
02 Methotrexate + UDCA versus placebo/no intervention + UDCA					
× Gonzalez-Koch 1997	0/13	0/12		0.0	Not estimable
Subtotal (95% CI)	13	12		0.0	Not estimable
Total events: 0 (Methotrexate), 0 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect: not applicable					
Total (95% CI)	43	42		100.0	5.00 [1.19, 20.92]
Total events: 10 (Methotrexate), 2 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect z=2.20 p=0.03					



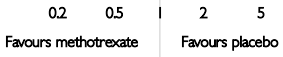
Analysis 01.02. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 02 Mortality or liver transplantation (expressed as relative risk)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 02 Mortality or liver transplantation (expressed as relative risk)

Study	Methotrexate n/N	Placebo n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Methotrexate versus placebo					
Hendrickse 1999	11/30	7/30		100.0	1.57 [0.71, 3.50]
Subtotal (95% CI)	30	30		100.0	1.57 [0.71, 3.50]
Total events: 11 (Methotrexate), 7 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.11 p=0.3					
02 Methotrexate + UDCA versus placebo/no intervention + UDCA					
× Gonzalez-Koch 1997	0/13	0/12		0.0	Not estimable
Subtotal (95% CI)	13	12		0.0	Not estimable
Total events: 0 (Methotrexate), 0 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect: not applicable					
Total (95% CI)	43	42		100.0	1.57 [0.71, 3.50]
Total events: 11 (Methotrexate), 7 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.11 p=0.3					

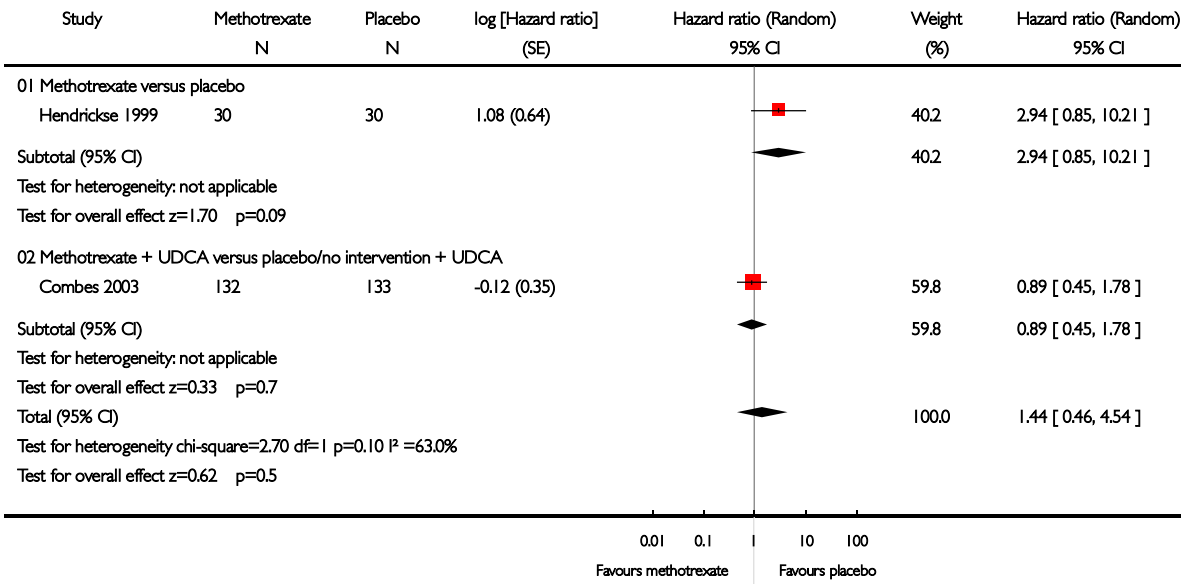


Analysis 01.03. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 03 Mortality or liver transplantation (expressed as hazard ratio) - random effects

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 03 Mortality or liver transplantation (expressed as hazard ratio) - random effects



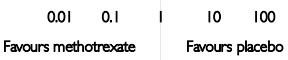
Analysis 01.04. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 04 Mortality or liver transplantation (expressed as hazard ratio) - fixed effect

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 04 Mortality or liver transplantation (expressed as hazard ratio) - fixed effect

Study	Methotrexate N	Placebo N	log [Hazard ratio] (SE)	Hazard ratio (Fixed) 95% CI	Weight (%)	Hazard ratio (Fixed) 95% CI
01 Methotrexate versus placebo						
Hendrickse 1999	30	30	1.08 (0.64)		23.7	2.94 [0.85, 10.21]
Subtotal (95% CI)					23.7	2.94 [0.85, 10.21]
Test for heterogeneity: not applicable						
Test for overall effect z=1.70 p=0.09						
02 Methotrexate + UDCA versus placebo/no intervention + UDCA						
Combes 2003	132	133	-0.12 (0.35)		76.3	0.89 [0.45, 1.78]
Subtotal (95% CI)					76.3	0.89 [0.45, 1.78]
Test for heterogeneity: not applicable						
Test for overall effect z=0.33 p=0.7						
Total (95% CI)					100.0	1.18 [0.64, 2.16]
Test for heterogeneity chi-square=2.70 df=1 p=0.10 I^2 =63.0%						
Test for overall effect z=0.54 p=0.6						



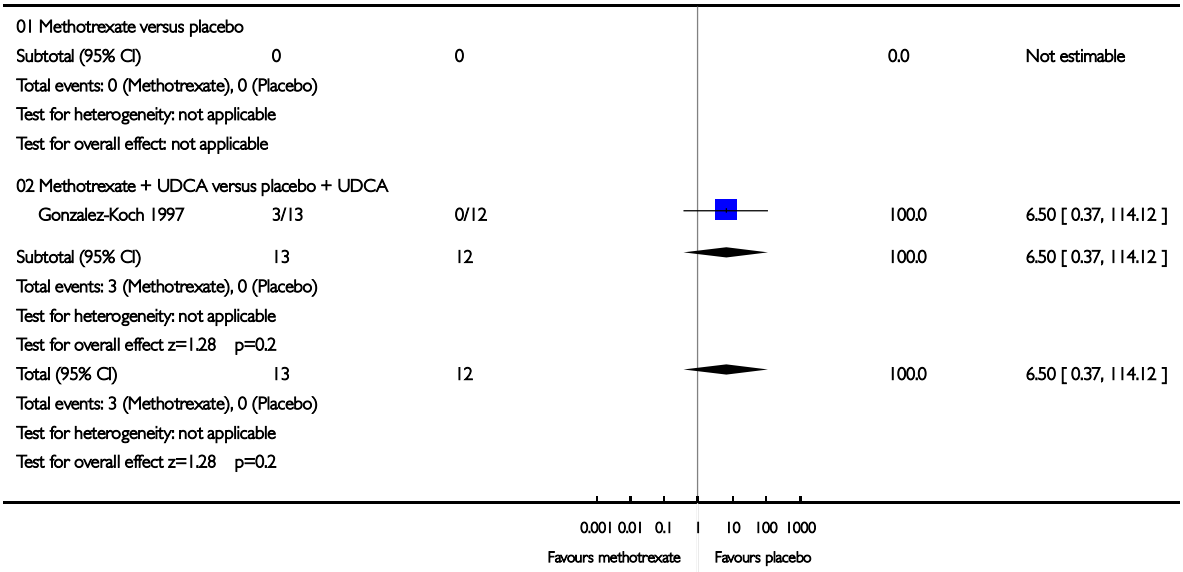
Analysis 01.05. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 05 Number of patients without improvement of pruritus

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 05 Number of patients without improvement of pruritus

Study	Methotrexate n/N	Placebo n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Methotrexate versus placebo					
Subtotal (95% CI)	0	0		0.0	Not estimable
Total events: 0 (Methotrexate), 0 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect: not applicable					
02 Methotrexate + UDCA versus placebo + UDCA					
Gonzalez-Koch 1997	3/13	0/12		100.0	6.50 [0.37, 114.12]
Subtotal (95% CI)	13	12		100.0	6.50 [0.37, 114.12]
Total events: 3 (Methotrexate), 0 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.28 p=0.2					
Total (95% CI)	13	12		100.0	6.50 [0.37, 114.12]
Total events: 3 (Methotrexate), 0 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect z=1.28 p=0.2					



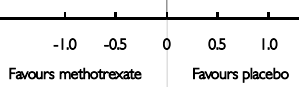
Analysis 01.06. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 06 Pruritus score

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 06 Pruritus score

Study	Methotrexate N	Methotrexate Mean(SD)	Placebo N	Placebo Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Methotrexate versus placebo							
Hendrickse 1999	30	0.87 (0.93)	30	1.04 (0.88)		100.0	-0.17 [-0.63, 0.29]
Subtotal (95% CI)	30		30			100.0	-0.17 [-0.63, 0.29]
Test for heterogeneity: not applicable							
Test for overall effect z=0.73 p=0.5							
02 Methotrexate + UDCA versus placebo/no intervention + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							
Total (95% CI)	30		30			100.0	-0.17 [-0.63, 0.29]
Test for heterogeneity: not applicable							
Test for overall effect z=0.73 p=0.5							



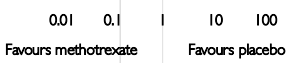
Analysis 01.07. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 07 Number of patients without improvement of fatigue

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 07 Number of patients without improvement of fatigue

Study	Methotrexate n/N	Placebo n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Methotrexate versus placebo					
Subtotal (95% CI)	0	0		0.0	Not estimable
Total events: 0 (Methotrexate), 0 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect: not applicable					
02 Methotrexate + UDCA versus placebo/no intervention + UDCA					
Gonzalez-Koch 1997	1/13	1/12		100.0	0.92 [0.06, 13.18]
Subtotal (95% CI)	13	12		100.0	0.92 [0.06, 13.18]
Total events: 1 (Methotrexate), 1 (Placebo)					
Test for heterogeneity: not applicable					
Test for overall effect z=0.06 p=1					



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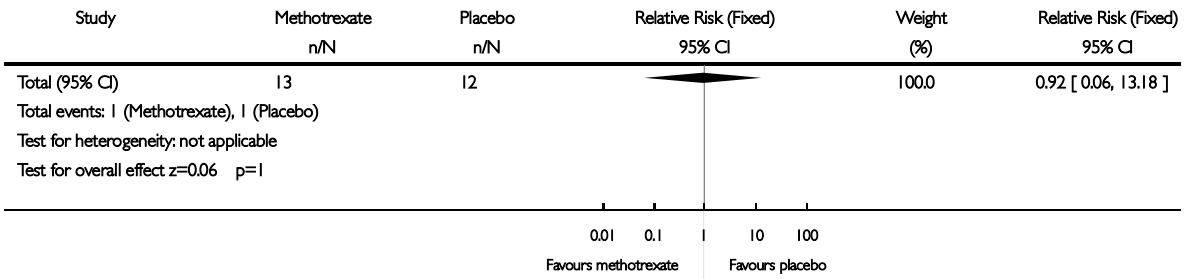
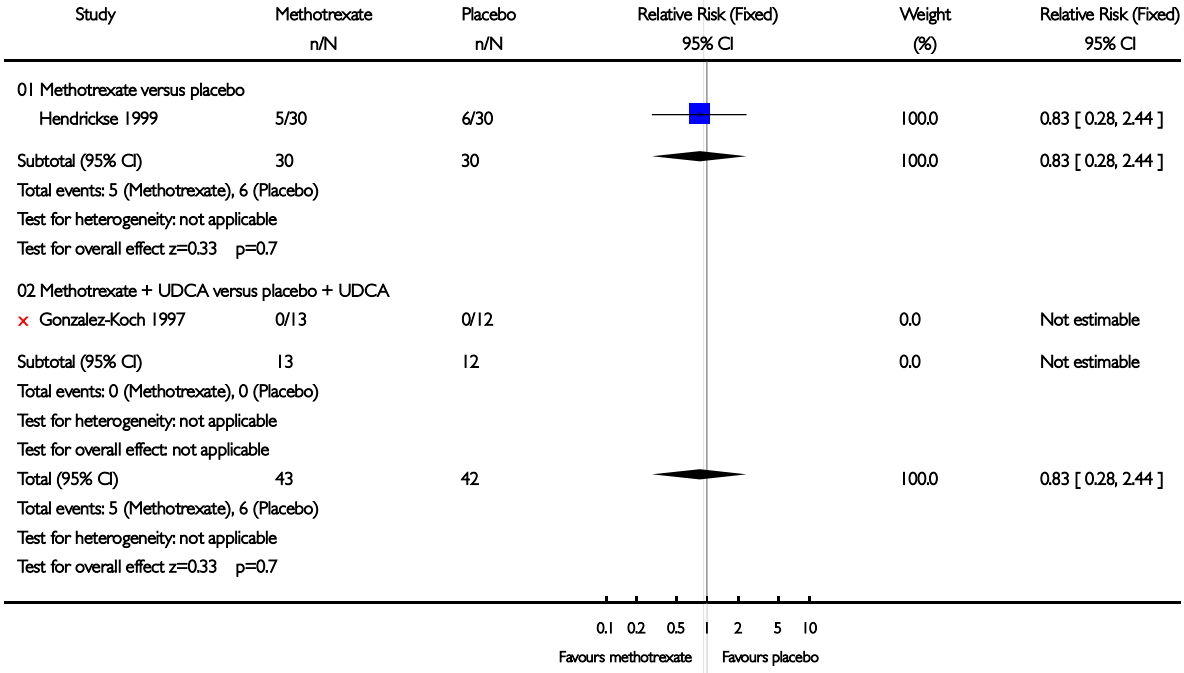


Analysis 01.08. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 08 Number of patients developing liver complications

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 08 Number of patients developing liver complications



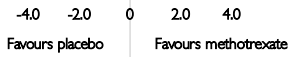
Analysis 01.09. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 09 S-albumin (g/dl)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 09 S-albumin (g/dl)

Study	Methotrexate N	Methotrexate Mean(SD)	Placebo N	Placebo Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Methotrexate versus placebo							
Hendrickse 1999	29	37.40 (5.92)	28	38.30 (3.70)		100.0	-0.90 [-3.45, 1.65]
Subtotal (95% CI)	29		28			100.0	-0.90 [-3.45, 1.65]
Test for heterogeneity: not applicable							
Test for overall effect z=0.69 p=0.5							
02 Methotrexate + UDCA versus placebo/no intervention + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							
Total (95% CI)	29		28			100.0	-0.90 [-3.45, 1.65]
Test for heterogeneity: not applicable							
Test for overall effect z=0.69 p=0.5							



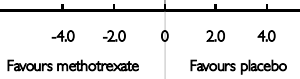
Analysis 01.10. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 10 Prothrombin time (second)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 10 Prothrombin time (second)

Study	Methotrexate N	Methotrexate Mean(SD)	Placebo N	Placebo Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Methotrexate versus placebo							
Hendrickse 1999	25	14.20 (3.70)	27	13.30 (4.16)		100.0	0.90 [-1.24, 3.04]
Subtotal (95% CI)	25		27			100.0	0.90 [-1.24, 3.04]
Test for heterogeneity: not applicable							
Test for overall effect z=0.83 p=0.4							
02 Methotrexate + UDCA versus placebo/no intervention + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							
Total (95% CI)	25		27			100.0	0.90 [-1.24, 3.04]
Test for heterogeneity: not applicable							
Test for overall effect z=0.83 p=0.4							

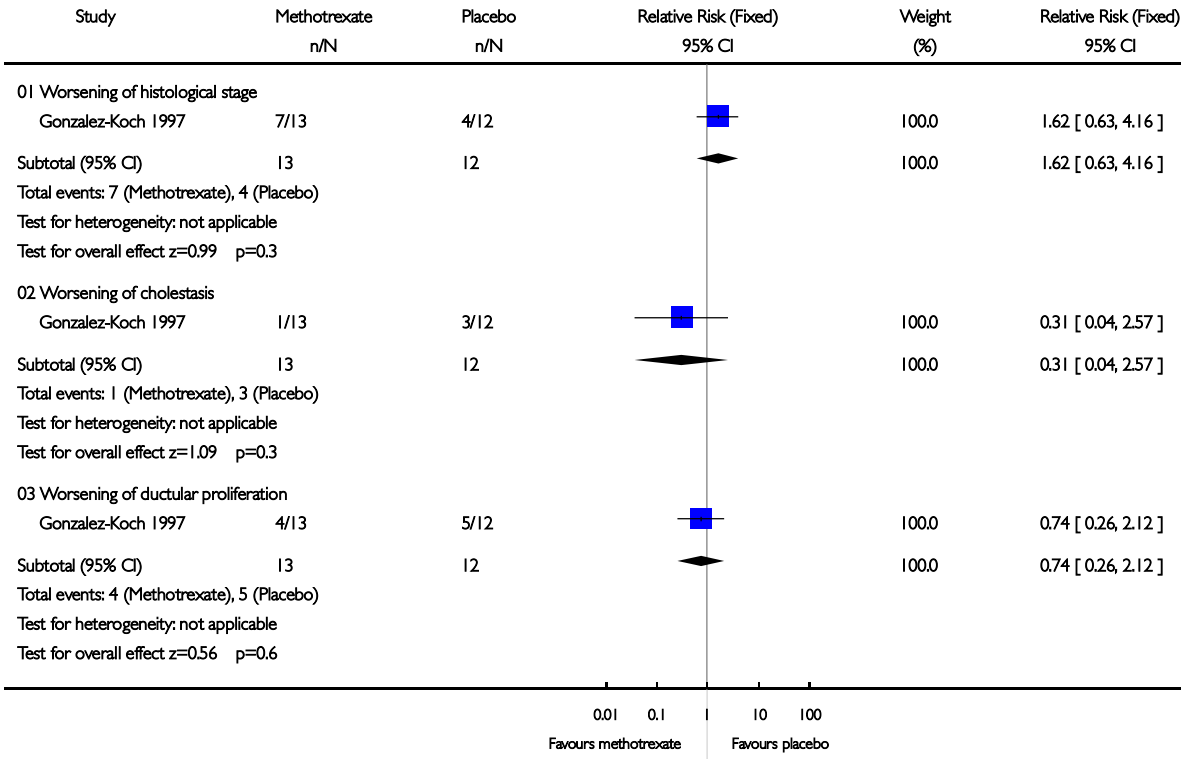


Analysis 01.11. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 11 Liver biopsy findings - dichotomous variables

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 11 Liver biopsy findings - dichotomous variables

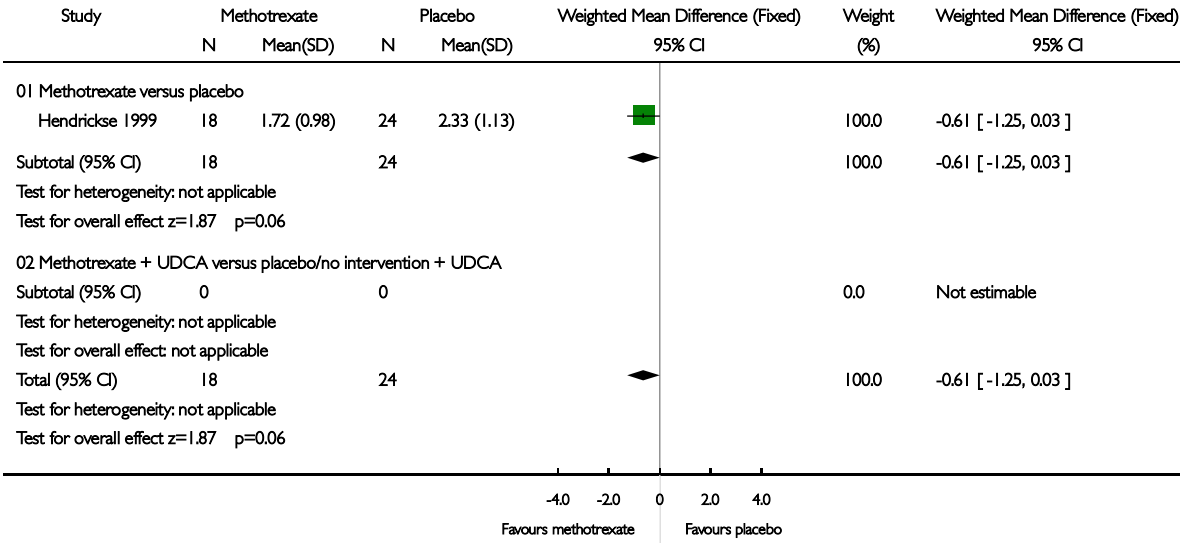


Analysis 01.12. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 12 Liver biopsy findings - histological stage

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 12 Liver biopsy findings - histological stage

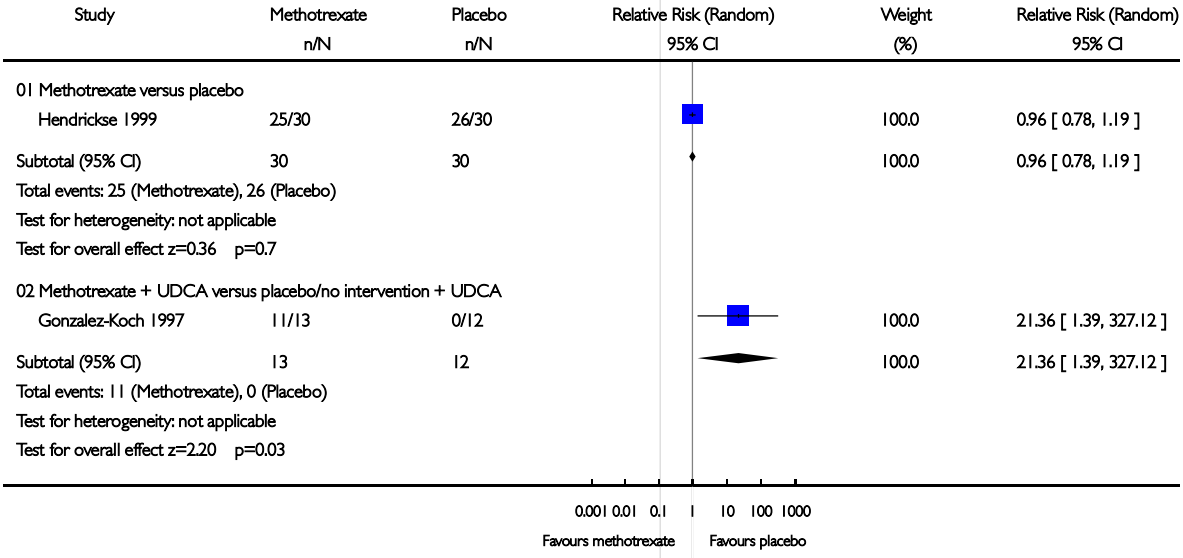


Analysis 01.13. Comparison 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid), Outcome 13 Adverse events

Review: Methotrexate for primary biliary cirrhosis

Comparison: 01 Methotrexate (with or without ursodeoxycholic acid) versus placebo (with or without ursodeoxycholic acid)

Outcome: 13 Adverse events



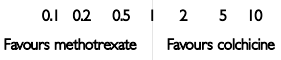
Analysis 02.01. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 01 Mortality or liver transplantation

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 01 Mortality or liver transplantation

Study	Methotrexate n/N	Colchicine n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Methotrexate versus colchicine					
Kaplan 2004	11/42	9/43		100.0	1.25 [0.58, 2.71]
Subtotal (95% CI)	42	43		100.0	1.25 [0.58, 2.71]
Total events: 11 (Methotrexate), 9 (Colchicine)					
Test for heterogeneity: not applicable					
Test for overall effect z=0.57 p=0.6					
02 Methotrexate + UDCA versus colchicine + UDCA					
Subtotal (95% CI)	0	0		0.0	Not estimable
Total events: 0 (Methotrexate), 0 (Colchicine)					
Test for heterogeneity: not applicable					
Test for overall effect: not applicable					



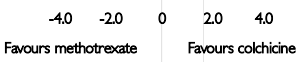
Analysis 02.02. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 02 Pruritus score

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 02 Pruritus score

Study	Methotrexate N	Mean(SD)	Colchicine N	Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Methotrexate versus colchicine							
Kaplan 2004	42	0.44 (0.71)	43	1.12 (1.25)		100.0	-0.68 [-1.11, -0.25]
Subtotal (95% CI)	42		43			100.0	-0.68 [-1.11, -0.25]
Test for heterogeneity: not applicable							
Test for overall effect z=3.09 p=0.002							
02 Methotrexate + UDCA versus colchicine + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							

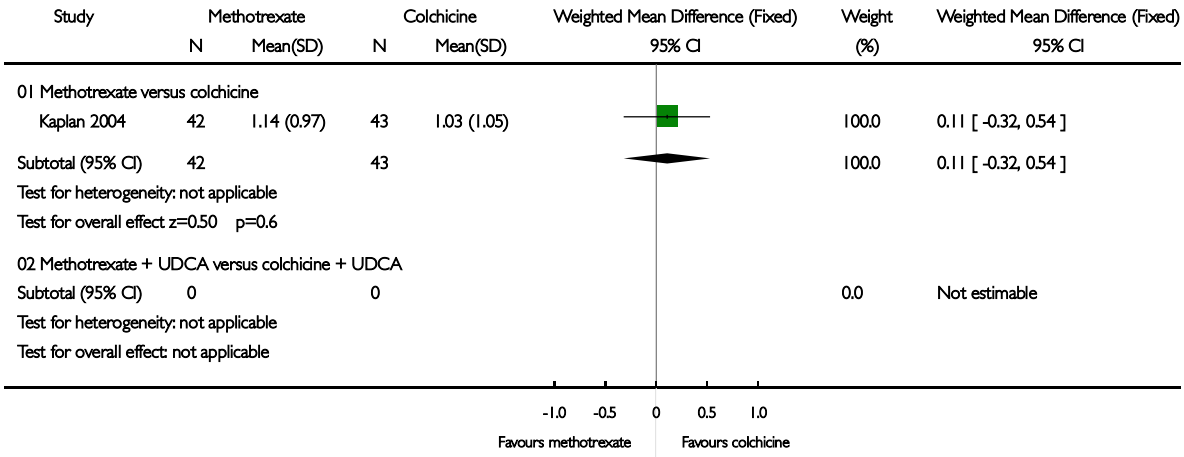


Analysis 02.03. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 03 Fatigue score

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 03 Fatigue score

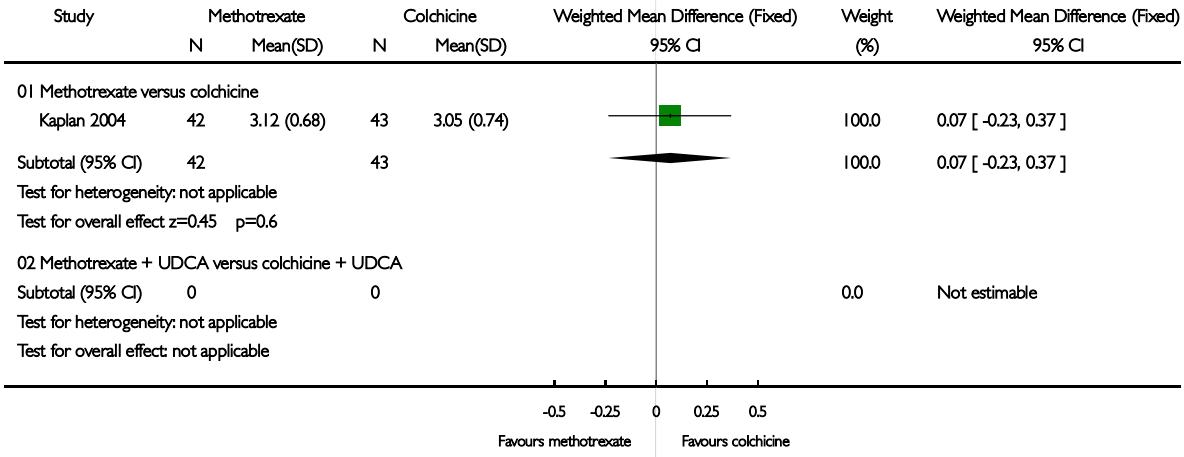


Analysis 02.04. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 04 S-bilirubin (ømol/L) (presented as log scaled)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 04 S-bilirubin (µmol/L) (presented as log scaled)



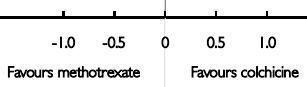
Analysis 02.05. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 05 S-alkaline phosphatases (ALP)(IU/L) (presented as log scaled)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 05 S-alkaline phosphatases (ALP)(IU/L) (presented as log scaled)

Study	Methotrexate N	Methotrexate Mean(SD)	Colchicine N	Colchicine Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Methotrexate versus colchicine							
Kaplan 2004	42	5.46 (0.79)	43	5.87 (0.57)		100.0	-0.41 [-0.70, -0.12]
Subtotal (95% CI)	42		43			100.0	-0.41 [-0.70, -0.12]
Test for heterogeneity: not applicable							
Test for overall effect z=2.74 p=0.006							
02 Methotrexate + UDCA versus colchicine + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							



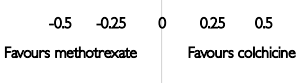
Analysis 02.06. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 06 S-aspartate aminotransferase (AST)(IU/L) (presented as log scaled)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 06 S-aspartate aminotransferase (AST)(IU/L) (presented as log scaled)

Study	Methotrexate N	Methotrexate Mean(SD)	Colchicine N	Colchicine Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Methotrexate versus colchicine							
Kaplan 2004	42	4.13 (0.56)	43	4.19 (0.57)		100.0	-0.06 [-0.30, 0.18]
Subtotal (95% CI)	42		43			100.0	-0.06 [-0.30, 0.18]
Test for heterogeneity: not applicable							
Test for overall effect z=0.49 p=0.6							
02 Methotrexate + UDCA versus colchicine + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							

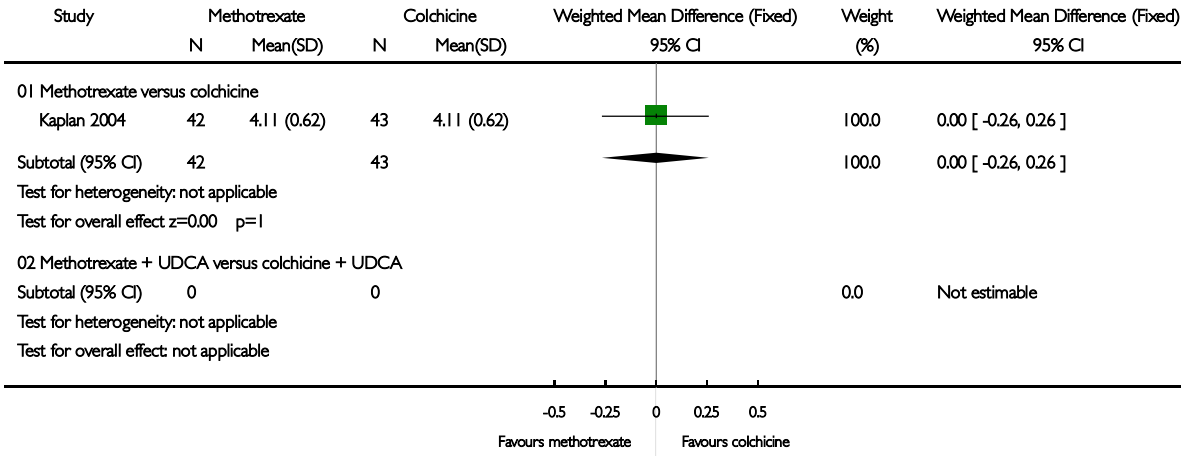


Analysis 02.07. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 07 S-alanine aminotransferase (ALT)(IU/L) (presented as log scaled)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 07 S-alanine aminotransferase (ALT)(IU/L) (presented as log scaled)

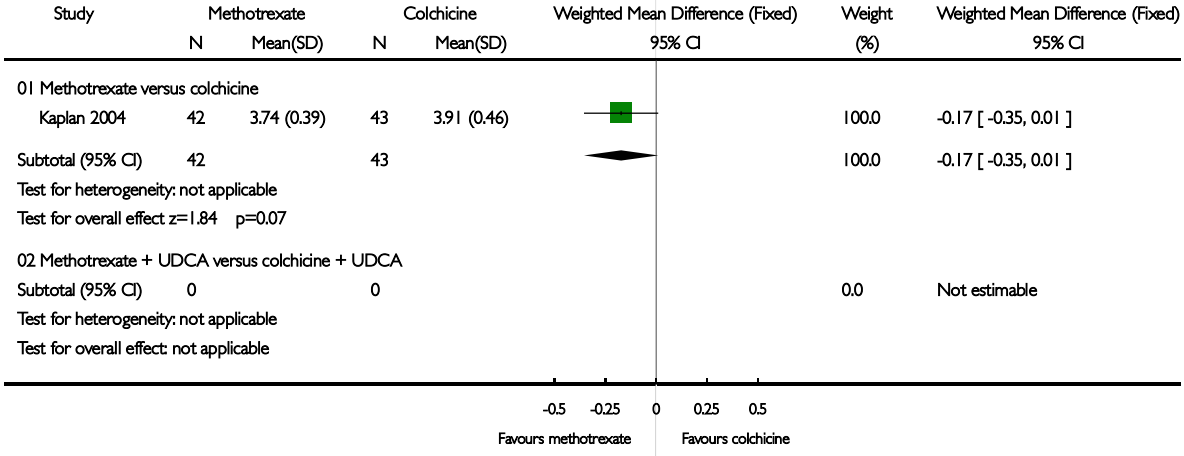


Analysis 02.08. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 08 S-albumin (g/dl)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 08 S-albumin (g/dl)



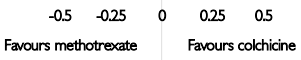
Analysis 02.09. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 09 S-cholesterol (total) (mmol/L) (presented as log scaled)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 09 S-cholesterol (total) (mmol/L) (presented as log scaled)

Study	Methotrexate N	Methotrexate Mean(SD)	Colchicine N	Colchicine Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Methotrexate versus colchicine							
Kaplan 2004	42	1.74 (0.25)	43	1.83 (0.32)		100.0	-0.09 [-0.21, 0.03]
Subtotal (95% CI)	42		43			100.0	-0.09 [-0.21, 0.03]
Test for heterogeneity: not applicable							
Test for overall effect z=1.45 p=0.1							
02 Methotrexate + UDCA versus colchicine + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							



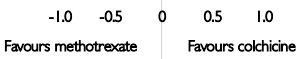
Analysis 02.10. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 10 Plasma immunoglobulin M (g/L) (presented as log scaled)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 10 Plasma immunoglobulin M (g/L) (presented as log scaled)

Study	Methotrexate N	Methotrexate Mean(SD)	Colchicine N	Colchicine Mean(SD)	Weighted Mean Difference (Fixed) 95% CI	Weight (%)	Weighted Mean Difference (Fixed) 95% CI
01 Methotrexate versus colchicine							
Kaplan 2004	42	1.00 (0.68)	43	1.47 (0.57)		100.0	-0.47 [-0.74, -0.20]
Subtotal (95% CI)	42		43			100.0	-0.47 [-0.74, -0.20]
Test for heterogeneity: not applicable							
Test for overall effect z=3.45 p=0.0006							
02 Methotrexate + UDCA versus colchicine + UDCA							
Subtotal (95% CI)	0		0			0.0	Not estimable
Test for heterogeneity: not applicable							
Test for overall effect: not applicable							

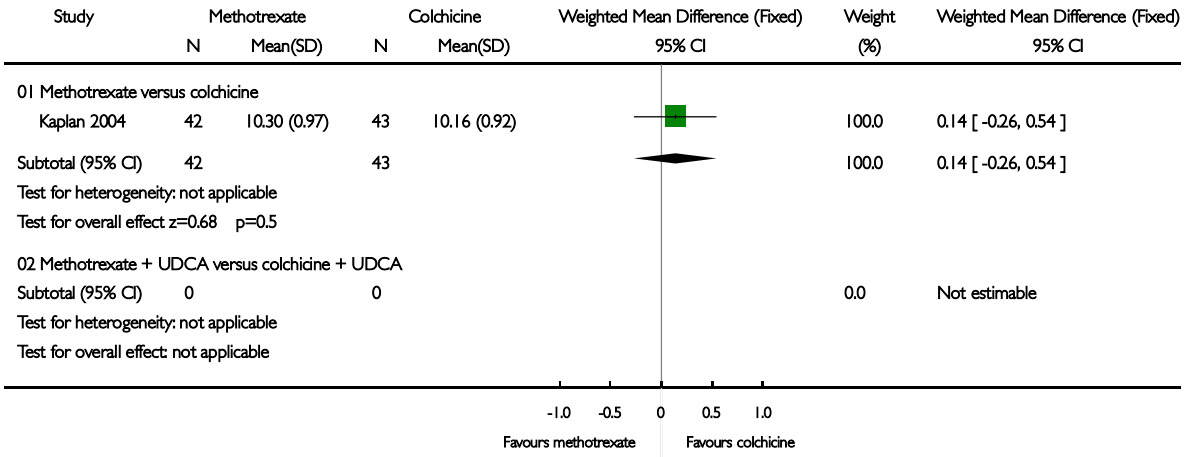


Analysis 02.11. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 11 Prothrombin time (second)

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 11 Prothrombin time (second)

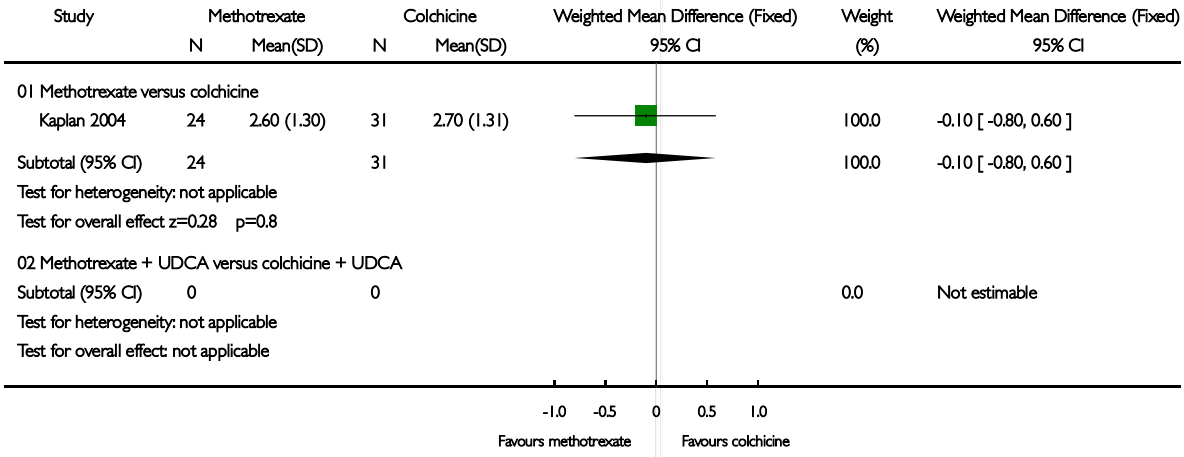


Analysis 02.12. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 12 Liver biopsy findings - histological stage

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 12 Liver biopsy findings - histological stage

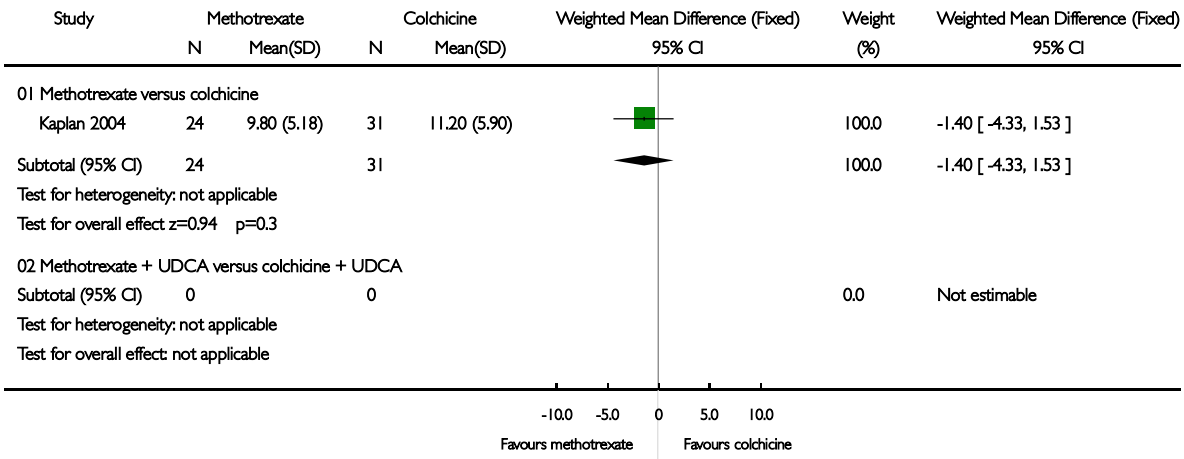


Analysis 02.13. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 13 Liver biopsy findings - histological score

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 13 Liver biopsy findings - histological score

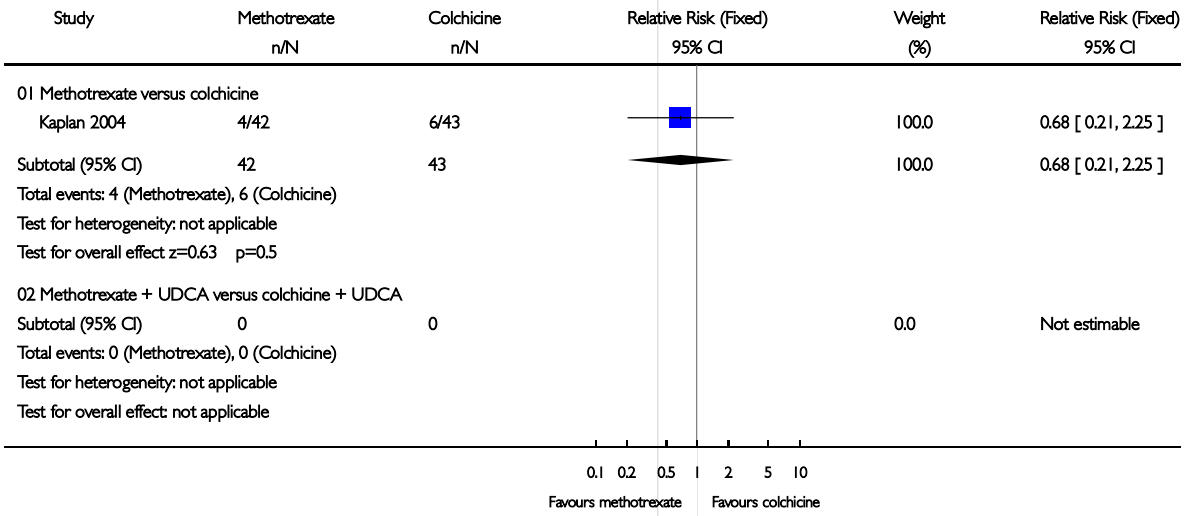


Analysis 02.14. Comparison 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid), Outcome 14 Adverse events

Review: Methotrexate for primary biliary cirrhosis

Comparison: 02 Methotrexate (with or without ursodeoxycholic acid) versus colchicine (with or without ursodeoxycholic acid)

Outcome: 14 Adverse events



Appendix 5

Azathioprine for primary biliary cirrhosis (Review)

Gong Y, Christensen E, Gluud C

This is a reprint of a Cochrane review, prepared and maintained by The Cochrane Collaboration and published in *The Cochrane Library* 2007, Issue 3

http://www.thecochranelibrary.com

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Azathioprine for primary biliary cirrhosis (Review)

Gong Y, Christensen E, Gluud C

Status: *New*

This record should be cited as:
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Date of most recent substantive amendment: 21 May 2007

ABSTRACT

Background
Azathioprine is used for patients with primary biliary cirrhosis, but the therapeutic responses in randomised clinical trials have been conflicting.

Objectives
To assess the benefits and harms of azathioprine for patients with primary biliary cirrhosis.

Search strategy
Randomised clinical trials were identified by searching *The Cochrane Hepato-Biliary Group Controlled Trials Register*, the *Cochrane Central Register of Controlled Trials (CENTRAL)* in *The Cochrane Library*, *MEDLINE*, *EMBASE*, *Science Citation Index Expanded*, *The Chinese Biomedical Database*, and *LILACS*, and manual searches of bibliographies to September 2005.

Selection criteria
Randomised clinical trials comparing azathioprine versus placebo, no intervention, or another drug were included irrespective of blinding, language, year of publication, and publication status.

Data collection and analysis
Our primary outcomes were mortality, and mortality or liver transplantation. Dichotomous outcomes were reported as relative risk (RR) with 95% confidence interval (CI). Continuous outcomes were reported as weighted mean difference (WMD) or standardised mean difference (SMD). We examined the intervention effects by random-effects and fixed-effect models.

Main results
We identified two randomised clinical trials with 293 patients. Only one of the trials was regarded as having low bias risk. Azathioprine did not significantly decrease mortality (RR 0.80, 95% CI 0.49 to 1.31, 2 trials). Azathioprine did not improve pruritus at one-year intervention (RR 0.71, 95% CI 0.28 to 1.84, 1 trial), cirrhosis development, or quality of life. Patients given azathioprine experienced significantly more adverse events than patients given no intervention or placebo (RR 2.44, 95% CI 1.14 to 5.20, 2 trials). The common adverse events were rash, severe diarrhoea, and bone marrow depression.

Authors' conclusions
There is no evidence to support the use of azathioprine for patients with primary biliary cirrhosis. Researchers who are interested in performing further randomised clinical trials should be aware of the risks of adverse events.

PLAIN LANGUAGE SUMMARY

There is no evidence to support azathioprine for patients with primary biliary cirrhosis

Primary biliary cirrhosis (PBC) is a chronic disease of the liver that is characterised by destruction of bile ducts. Estimates of annual incidence range from 2 to 24 patients per million population, and estimates of prevalence range from 19 to 240 patients per million population. PBC primarily affects middle-aged women. The forecast for the symptomatic patient after diagnosis is between 10 and 15 years. The cause of PBC is unknown, but the dynamics of the disease resemble the group 'autoimmune disease'. Therefore, one might expect a noticeable effect of administering an immune repressing drug (immunosuppressant). This review evaluates all clinical data on the immunosuppressant azathioprine in relation to PBC.

The findings of this review are based on two clinical trials with 293 patients. The drug azathioprine was tested versus placebo or no intervention. The primary findings of the review are that azathioprine has no effect on survival, itching, progression of the disease (cirrhosis development), or quality of life. Patients given azathioprine experienced more adverse events than patients given placebo.

BACKGROUND

Primary biliary cirrhosis is a chronic liver disease of unknown aetiology. Ninety per cent of patients with primary biliary cirrhosis are females and the majority are diagnosed after the age of 40 years (James 1981). Over the past 30 years, substantial increases in the prevalence of primary biliary cirrhosis have been observed (Kim 2000). Primary biliary cirrhosis is now a frequent cause of liver morbidity, and patients with primary biliary cirrhosis are significant users of health resources, including liver transplantation (Prince 2003).

Primary biliary cirrhosis is classically defined on the basis of the triad: antimitochondrial antibodies, found in over 95 per cent of patients with primary biliary cirrhosis (Fregeau 1989; Lacerda 1995; Invernizzi 1997; Turchany 1997; Mattalia 1998); abnormal liver function tests that are typically cholestatic (with raised activity of alkaline phosphatases being the most frequently seen abnormality); and characteristic liver histological changes (Scheuer 1967) in the absence of extrahepatic biliary obstruction (Kaplan 1996). Patients may either be diagnosed during a symptomatic phase (the common symptoms being pruritus, fatigue, jaundice, liver enlargement, signs of portal hypertension, sicca complex, and scleroderma-like lesions), in which case survival is significantly decreased, or during an asymptomatic phase, in which one has a relatively favourable prognosis (Beswick 1985; Balasubramaniam 1990). However, 40% to 100% of these patients will subsequently develop symptoms of primary biliary cirrhosis (Nyberg 1989; Metcalf 1996; Prince 2000).

Although the aetiology remains unknown, primary biliary cirrhosis is analogous to the graft-versus-host syndrome in which the immune system is sensitised to foreign proteins. Most primary biliary cirrhosis patients have increased class II human leukocyte antigen (HLA) histocompatibility antigen expression on bile duct cells (Ballardini 1984; Van den Oord 1986), and cytotoxic T-cells are infiltrating the bile duct epithelium (Yamada 1986). Other duct systems of the body with a high concentration of HLA class II antigens on their epithelium, such as the lacrimal and pancreatic glands, may be involved in the disease process (Epstein 1982).

Patients with primary biliary cirrhosis have been subjected to many drugs. Ursodeoxycholic acid (a bile acid) is the most extensively used drug in these patients (Verma 1999). Other drugs have been immunomodulatory and other agents, such as colchicine (Warnes 1987; Vuoristo 1995; Poupon 1996; Gong 2005b), prednisolone (Mitchison 1992; Prince 2005), chlorambucil (Hoofnagle 1986), cyclosporin A (Minuk 1988; Wiesner 1990; Gong 2005c), D-penicillamine (Dickson 1985; Neuberger 1985; Gong 2004), methotrexate (Kaplan 1991; Lindor 1995; Gong 2005a), or azathioprine (Heathcote 1976; Christensen 1985).

Azathioprine is an immunosuppressant, suppressing delayed hypersensitivity and cellular cytotoxicity more than antibody responses. The immunosuppressive action of azathioprine depends on its conversion to active 6-mercaptopurine by thiopurine S-methyl-transferase (Lennard 1992). Azathioprine is used for Crohn's disease (Pearson 1998), renal homotransplantation (Sandrini 2000), and severe, active rheumatoid arthritis (Suarez-Almazor 2000). The first controlled therapeutic trial of azathioprine in primary biliary cirrhosis showed no efficacy and suggested the possibility of significant toxicity of azathioprine therapy (Heathcote 1976). In contrast, a large multicenter trial showed evidence of efficacy with very little toxicity (Christensen 1985). We have been unable to identify meta-analyses or systematic reviews on the beneficial and harmful effects of azathioprine in primary biliary cirrhosis.

OBJECTIVES

To systematically assess the benefits and harms of azathioprine for primary biliary cirrhosis.

CRITERIA FOR CONSIDERING STUDIES FOR THIS REVIEW

Types of studies

We included randomised clinical trials irrespective of blinding, language, year of publication, and publication status. We excluded

studies using quasi-randomisation (for example, allocation by date of birth). Since uncommon adverse events are rarely captured in randomised clinical trials, we also evaluate adverse events from non-randomised, controlled studies and observational studies that fulfilled the inclusion criteria of this review.

Types of participants

Patients with primary biliary cirrhosis, ie, patients having at least two of the following: elevated serum activity of alkaline phosphatases (or other markers of intrahepatic cholestasis), a positive result for serum mitochondrial antibody, and liver biopsy findings diagnostic for or compatible with primary biliary cirrhosis.

Types of intervention

Administration of any dose of azathioprine versus placebo or no intervention or other drugs. Co-interventions were allowed as long as the intervention arms of the randomised clinical trial receive the same co-interventions.

Types of outcome measures

Primary outcome measure

- Mortality.
- Mortality or liver transplantation.

Secondary outcome measures

- Pruritus: number of patients with pruritus at one-year intervention.
- Liver biopsy: number of patients who developed cirrhosis.
- Quality of life: broad nature of a concept that includes physical functioning (ability to carry out activities of daily living such as self-care and walking around), psychological functioning (emotional and mental well-being), social functioning (social relationships and participation in social activities), and perception of health, pain, and overall satisfaction with life.
- Adverse events (excluding mortality and liver transplantation): the adverse event is defined as any untoward medical occurrence in a patient in either of the two arms of the included randomised clinical trials, which did not necessarily have a causal relationship with the treatment, but did, however, result in a dose reduction, discontinuation of treatment, or registration of the advent as an adverse event/side effect in accordance with the ICH-GCP guidelines (ICH-GCP 1997).

SEARCH METHODS FOR IDENTIFICATION OF STUDIES

See: Cochrane Hepato-Biliary Group methods used in reviews.

We identified randomised clinical trials in *The Cochrane Hepato-Biliary Group Controlled Trials Register*, which involves hand searches of major hepatology journals and conference proceedings, the *Cochrane Central Register of Controlled Trials* (*CENTRAL*) in *The Cochrane Library*, *MEDLINE*, *EMBASE*, *Science Citation Index Expanded*, *The Chinese Biomedical Database*, and *LILACS* (Royle 2003). We have given the search strategies in Table 01 with the time span of the searches.

We identified further trials by reading the reference lists of the identified studies. We wrote to the principal investigators to enquire about additional randomised clinical trials they might know of. We also contacted the pharmaceutical company (Salix Pharmaceuticals, Inc) producing azathioprine to obtain any unidentified or unpublished randomised clinical trials.

METHODS OF THE REVIEW

We performed the meta-analysis following our protocol (Gong 2006) and the recommendations given by the *Cochrane Handbook for Systematic Reviews of Interventions* (Higgins 2006) and the *Cochrane Hepato-Biliary Group Module* (Gluud 2007).

Data extraction

Two authors (YG and EC) independently evaluated whether the identified trials fulfil the inclusion criteria. We listed the excluded trials in 'Characteristics of excluded studies' with the reasons for exclusion. YG extracted data and EC validated the data extraction. Disagreements were resolved by discussion with CG.

Assessment of methodological quality

We assessed the methodological quality of the randomised clinical trials using four components (Schulz 1995; Moher 1998; Kjaergard 2001):

Generation of the allocation sequence

- Adequate, if the allocation sequence was generated by a computer or random number table. Drawing of lots, tossing of a coin, shuffling of cards, or throwing dice are considered as adequate if a person who was not otherwise involved in the recruitment of participants performed the procedure;
- Unclear, if the trial was described as randomised, but the method used for the allocation sequence generation was not described.

Allocation concealment

- Adequate, if the allocation of patients involved a central independent unit, on-site locked computer, identically appearing numbered drug bottles or containers prepared by an independent pharmacist or investigator, or sealed envelopes;
- Unclear, if the trial was described as randomised, but the method used to conceal the allocation was not described;
- Inadequate, if the allocation sequence was known to the investigators who assigned participants.

Blinding (or masking)

- Adequate, if the trial was described as double blind and the method of blinding involved identical placebo or active drug;
- Unclear, if the trial was described as double blind, but the method of blinding was not described;
- Not performed, if the trial was not double blind.

Follow-up

- Adequate, if the numbers and reasons for dropouts and withdrawals in all intervention groups were described or if it was specified that there were no dropouts or withdrawals;
- Unclear, if the report gave the impression that there had been no dropouts or withdrawals, but this was not specifically stated;
- Inadequate, if the number or reasons for dropouts and withdrawals were not described.

We *post hoc* classified trials with at least two out of the four quality components, ie, adequate generation of the allocation sequence, adequate allocation concealment, and adequate blinding, as trials with low-risk bias.

Characteristics of patients
Number of patients randomised; mean (or median) age; sex ratio; histological stage; number of patients lost to follow-up.

Characteristics of interventions
Type, dose, and form of azathioprine intervention; type of intervention in the control group and collateral interventions; duration of treatment and follow-up.

Characteristics of outcomes
According to the protocol, outcomes were extracted from each included trial.

Statistical methods
We used the statistical package RevMan Analyses 1.0.2 (RevMan 2003) provided by The Cochrane Collaboration. We presented dichotomous data as relative risk (RR) with 95% confidence interval (CI) and continuous outcome measures by weighted mean differences (WMD) with 95% CI.

We examined intervention effects by using both a random-effects model (DerSimonian 1986) and a fixed-effect model (Mantel 1959) with the significant level set at $P < 0.05$. If the results of the two analyses led to the same conclusion, we presented only the results of the fixed-effect model. In case of discrepancies of the two models, we reported the results of both models. We explored the presence of statistical heterogeneity by chi-squared test with significance set at $P < 0.10$ and measured the quantity of heterogeneity by I^2 (Higgins 2002) .

Sensitivity analyses
For primary outcome measures, we used a method to pool uncertainty intervals, which incorporates both sampling error and the potential impact of missing data (Gamble 2005).

For secondary outcomes, we adopted 'available case analysis' at maximum reported follow-up. Therefore, in the review, the number of patients in the denominator changed according to the secondary outcomes investigated.

Bias exploration
Funnel plot was used to provide a visual assessment of whether treatment estimates were associated with study size. The performance of the available methods of detecting publication bias and other biases (Begg 1994; Egger 1997; Macaskill 2001) vary with the magnitude of the treatment effect, the distribution of study size, and whether a one- or two-tailed test is used (Macaskill 2001). Therefore, we intended to use the most appropriate method having good trade-off in the sensitivity and specificity, based on characteristics of the trials included in this review.

DESCRIPTION OF STUDIES

We identified a total of 190 references through electronic searches of *The Cochrane Hepato-Biliary Group Controlled Trials Register* (n = 16), the *Cochrane Central Register of Controlled Trials* in *The Cochrane Library* (n = 46), *MEDLINE* (n = 27), *EMBASE* (n = 79), and *Science Citation Index Expanded* (n = 22). We excluded 184 duplicates or clearly irrelevant references through reading abstracts. Accordingly, six references were retrieved for further assessment. Of these, we excluded one, which is listed under 'Characteristic of excluded studies' with reasons for exclusion. Accordingly, five references referring to two randomised clinical trials fulfilled our inclusion criteria and were included.

The two randomised clinical trials were parallel group trials published as full articles. All the included trials reported random allocation of 293 patients with primary biliary cirrhosis to azathioprine versus no intervention (Heathcote 1976) and azathioprine versus placebo (Christensen 1985).

The mean age of patients in the included trials was 53 years and 90% of the patients were women. Half patients had histological stage III or IV in Christensen 1985. The entry and exclusion criteria varied across trials, but were generally well-defined, making it highly likely that all patients did have primary biliary cirrhosis. The dosage of azathioprine used in the Heathcote 1976 trial was around 30% higher than the one used in the Christensen 1985 trial. The trial duration (treatment plus follow-up) was 5 years in the Heathcote 1976 and 11 years in the Christensen 1985 trials. Details are listed in the table of 'Characteristics of included studies'.

METHODOLOGICAL QUALITY

Generation of the allocation sequence was adequate in the Christensen 1985 trial. Allocation concealment was adequate in both trials, which used the sealed envelope technique. Heathcote 1976 used no intervention as a control group; Christensen 1985 treated

the control group with identically looking placebo. The follow-up was adequately described in both trials: six patients were lost to follow-up and one patient withdrew from the Heathcote 1976 trial; 63 patients were lost to follow-up and 30 patients withdrew from the Christensen 1985 trial. We regard Christensen 1985 trial as a low-bias risk trial.

RESULTS

Mortality

Seventeen patients died in the Heathcote 1976 trial, whereas 119 patients died in the Christensen 1985 trial. We did not see a significant difference in mortality between azathioprine and control when only data on available patients were included (RR 0.88, 95 CI 0.74, 1.06) (Comparison 01-01). Considering the impact of missing data (eg, lost to follow-up or patients withdrawn), azathioprine did not significantly reduce the mortality risk (RR 0.80, 95% CI 0.49 to 1.31, pooled uncertainty intervals) (Comparison 01-02).

Mortality or liver transplantation

No patients were liver transplanted so this composite outcome could not be assessed.

Pruritus at one-year intervention

At the start of the Heathcote 1976 trial, 18 of the 19 azathioprine-treated and 16 of the 20 control patients complained of pruritus. At one-year follow-up, 5 of the 18 treated and 7 of the 18 control patients complained of pruritus (RR = 0.71, 95% CI 0.28 to 1.84). Pruritus data were not extractable in the Christensen 1985 trial.

Hepatic histology

Ten of the 16 treated patients and 7 of the 12 control patients had developed cirrhosis after entry to the Heathcote 1976 trial (RR 1.07, 95 % CI 0.58 to 1.97).

Quality of life

Nine patients in each group remained fit and able to work for the whole period of the Heathcote 1976 trial. After combining data from the two trials, it seems that azathioprine did not improve the state of well-being among patients with primary biliary cirrhosis (RR 0.74, 95% CI 0.50 to 1.08).

Adverse events

Both trials reported adverse events. The pooled data showed that patients given azathioprine had experienced more adverse events than patients given nothing or placebo (RR 2.44, 95% CI 1.14 to 5.20). The common adverse events were rash, diarrhoea, and marrow depression.

We were unable to extract data on fatigue, liver complication, and liver biochemistry.

DISCUSSION

The results of our systematic review do not support azathioprine for patients with primary biliary cirrhosis. Patients given azathioprine experienced more adverse events than patients given nothing or placebo, though not all adverse events were severe.

To our knowledge, only two trials have been conducted to evaluate the role of azathioprine in primary biliary cirrhosis. Therefore, this systematic review has a major limitation: a small number of trials included (Ioannidis 2001). The Heathcote 1976 trial included only 45 patients, did not use blinding, and only lasted five years. This is shorter than the estimated median survival of the disease, 10 to 15 years (Prince 2003).

Patients given azathioprine did not have significantly lower risk of death than patients given nothing or placebo. The rate of missing data was 16% in the Heathcote 1976 trial and 38% in the Christensen 1985 trial. It is important to take account of the influence of these missing data. We, therefore, performed a sensitivity analysis using the pooled uncertainty intervals to incorporate the potential impact of missing data and sampling error, which resulted in a RR 0.80 with 95% CI 0.49 to 1.31. This result gives consistent finding with the result conditional on data of available patients. During the five-year follow-up in the Heathcote 1976 trial, 17 patients died, and no difference in survival between the two groups was observed. Standard survival analysis in the Christensen 1985 trial revealed no significant difference between the two groups. However, when adjustment for imbalances between the two groups (primarily serum bilirubin) was made, there was a slight, but statistically significant difference in survival favouring azathioprine ($P < 0.01$). The first trial was clearly too small to have a reasonable chance of demonstrating or excluding any benefit of intervention. In contrast, the Christensen 1985 trial contained more patients with severe disease who were followed long enough to potentially showing an improvement in survival. However, even in this ideal clinical setting, demonstrating improvement in patient survival would still require statistical adjustment for baseline prognostic variables; this sort of analysis needs to be interpreted cautiously (Pocock 2002; Deeks 2003).

One of the most important subjective aspects of primary biliary cirrhosis is pruritus. Azathioprine did not improve the degree of pruritus, though there was a consistently smaller number of patients complaining of pruritus in the treated group than in the control group (not statistically significant) (Heathcote 1976). It was reported, in the Christensen 1985 trial that there was no significant difference in the number of patients requiring cholestyramine treatment for pruritus between the two groups. The Heathcote 1976 trial and the Christensen 1985 trial reported the results on liver biochemistry tests. However, we could not extract these data. There were no significant differences in the two groups in regard to the levels of serum bilirubin, alkaline phosphatases, aspartate transaminase, cholesterol, serum albumin, and serum im-

munoglobulin M in the Heathcote 1976 trial. The effect of azathioprine did not reach statistical significance for any biochemical variable in the Christensen 1985 trial. Ten of the 16 treated versus 7 of the 12 control patients had developed cirrhosis (RR 1.07, 95% CI 0.58 to 1.97) in the Heathcote 1976 trial. There were no significant differences in the two groups in regard to intralobular inflammation, peripheral cholestasis, piecemeal necrosis, granulomas, fibrosis (without cirrhosis), and histologic stages in the Christensen 1985 trial.

Another important aspect is quality of life. Both Heathcote 1976 and Christensen 1985 measured this outcome in a similar manner. The Heathcote 1976 trial did not find a beneficial effect of azathioprine, whereas the Christensen 1985 trial claimed a marginal benefit (RR 0.63, P = 0.05). By pooling these two trials, we could not find a significant difference favouring azathioprine on quality of life (RR 0.74, 95% CI 0.50 to 1.08).

The pooled data showed that patients given azathioprine had nearly 2.5 times more adverse events than patients given nothing or placebo (RR 2.44, 95% CI 1.14 to 5.20). Most of the patients who were withdrawn complained of rash, gastrointestinal disorder, and bone marrow depression. In addition, the immunosuppressive action of azathioprine depends on its conversion to active 6-mercaptopurine by thiopurine S-methyl-transferase (Lennard 1992). Patients who inherit a thiopurine S-methyl-transferase deficiency accumulate excessive concentrations of the active thioguanine nucleotides in blood cells. This can lead to severe and potentially life-threatening problems with the formation of blood cells among patients taking azathioprine.

AUTHORS' CONCLUSIONS

Implications for practice

There is insufficient evidence to support azathioprine for patients with primary biliary cirrhosis. Patients taking azathioprine have higher risk of adverse events. Since patients with a thiopurine S-methyl-transferase deficiency would potentially have life-threatening haematopoietic toxicity, prescription of azathioprine should be monitored by laboratory tests, including full blood count and liver function.

Implications for research

Although the reviewed results of the two trials do not offer much optimism for a beneficial effect of azathioprine, researchers should recognize that only 293 patients have been randomised and the number of deaths were 136. These numbers may be considered too few, and investigators may wish to conduct further trials. If such trials are considered, then they ought to be closely monitored for both beneficial and adverse events. Any future trial should contain enough patients in concert with the sample size estimation, and patients should be followed long enough to allow observing potential improvement in survival. Any future trial ought to be reported according to the Consort Statement (www.consort-statement.org).

POTENTIAL CONFLICT OF INTEREST

None known.

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Warnes 1987
Warnes TW, Smith A, Lee FI, Haboubi NY, Johnson PJ, Hunt L. A controlled trial of colchicine in primary biliary cirrhosis: trial design and preliminary report. *Jounal of Hepatology* 1987;**5**(1):1–7. [MedLine: 88008957].

Wiesner 1990
Wiesner RH, Ludwig J, Lindor KD, Jorgensen RA, Baldus WP, Homburger HA, et al. A controlled trial of cyclosporine in the treatment of primary biliary cirrhosis. *New England Journal of Medicine* 1990; **322**(20):1419–24. [MedLine: 90231366].

Yamada 1986
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* *Indicates the major publication for the study*

TABLES

Characteristics of included studies

Study	Christensen 1985
Methods	Generation of allocation sequence: adequate, table of random numbers. Allocation concealment: adequate, sealed envelope technique. Blinding: identically looking placebo, no description of the taste and smell. Follow-up: adequately reported, 29 in the azathioprine group and 34 in the control group were lost to follow-up; 20 in the azathioprine group and 10 in the control group were withdrawn.
Participants	Country: UK, Denmark, Spain, USA, Australia, Belgium, and France. Mean age: 54.7 years in the azathioprine group, 54.9 years in the control group. Female/Male: 222/26. PBC stage status Azathioprine group: 18, 56, 19, and 34 in stage I to IV, respectively. Placebo group: 15, 52, 18, and 36 in stage I to IV, respectively.
Interventions	Azathioprine: 300 to 700 mg/week (n = 127). Placebo: (n = 121). Treatment and follow-up: 11 years.
Outcomes	(1) Mortality. (2) Clinical outcomes and liver biochemical variables. (3) Adverse events. (4) Quality of life.
Notes	
Allocation concealment	A – Adequate

Study	Heathcote 1976
Methods	Generation of allocation sequence: unclear. Allocation concealment: adequate, sealed envelope technique. Blinding: no blinding. Follow-up: adequately reported, 3 in the azathioprine group and 3 in the control group were withdrawn; 1 patient in the control group was lost to follow-up.
Participants	Country: UK. Mean age: 50.6 years in the azathioprine group, 51.7 years in the control group. Female/Male: 42/3. PBC stage status: pre-cirrhotic patients.
Interventions	Azathioprine: 2 mg/kg/day (n = 22). No intervention (control group): (n = 23). Treatment and follow-up: 5 years.
Outcomes	(1) Mortality. (2) Clinical outcomes, liver biochemical, and hepatic histological variables. (3) Adverse events.

	(4) Quality of life.
Notes	
Allocation concealment	A – Adequate

Characteristics of excluded studies

Study	Reason for exclusion
Wolfhagen 1998	A randomised trial comparing ursodeoxycholic acid, prednisone, and azathioprine versus ursodeoxycholic and placebo.

ADDITIONAL TABLES

Table 01. Search strategies

Database	Period	Search term
The Cochrane Hepato-Biliary Group Controlled Trials Register	September 2005.	'primary biliary cirrhosis' and 'azathioprine'
Cochrane Central Register of Controlled Trials (CENTRAL) in The Cochrane Library	Issue 3, 2005.	#1 = LIVER CIRRHOSIS BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = pbc #5 = #1 or #2 or #3 or #4 #6 = AZATHIOPRINE: MESH #7 = IMMUNOSUPPRESSIVE AGENTS: MESH #8 = azathioprine #9 = #6 or #7 or #8 #10 = #5 and #9
MEDLINE	January 1966 to September 2005.	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = PBC #5 = #1 or #2 or #3 or #4 #6 = AZATHIOPRINE: MESH #7 = IMMUNOSUPPRESSIVE AGENTS: MESH #8 = azathioprin* #9 = immunosuppressive agent* #10 = #6 or #7 or #8 or #9 #11 = #5 and #10 #12 = random* or placebo* or blind* or meta-analysis #13 = #11 and #12
EMBASE	January 1980 to September 2005.	#1 = PRIMARY-BILIARY-CIRRHOSIS: MESH #2 = BILIARY-CIRRHOSIS: MESH #3 = primary and biliary and cirrhosis

Table 01. Search strategies (*Continued*)

Database	Period	Search term
		#4 = primary biliary cirrhosis #5 = PBC #6 = #1 or #2 or #3 or #4 or #5 #7 = AZATHIOPRINE: MESH #8 = IMMUNOSUPPRESSIVE AGENTS: MESH #9 = azathioprin* #10 = immunosuppressive agent* #11 = #7 or #8 or #9 or #10 #12 = #6 and #11 #13 = random* or placebo* or blind* or meta-analysis #14 = #12 and #13
Science Citation Index Expanded (http://portal.isiknowledge.com/portal.cgi?DestApp=WOS&Func=Frame)	1945 to September 2005.	#1 = TS=(primary biliary cirrhosis OR PBC) #2 = TS=(azathioprine OR azathioprin*) #3 = #2 AND #1 #4 = TS=(random* OR blind* OR placebo* OR meta-analysis) #5 = #4 AND #3
LILACS	1982 to September 2005.	#1 = (primary and biliary and cirrhosis) or (primary biliary cirrhosis) #2 = primary biliary cirrhosis #3 = azathioprine #4 = (#1 OR #2) AND # 3
Chinese Biochemical CD Database	January 1979 to September 2005.	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = PBC #5 = #1 or #2 or #3 or #4 #6 = AZATHIOPRINE: MESH #7 = IMMUNOSUPPRESSIVE AGENTS: MESH #8 = azathioprin* #9 = immunosuppressive agent* #10 = #6 or #7 or #8 or #9 #11 = #5 and #10 #12 = random* or placebo* or blind* or meta-analysis #13 = #11 and #12

ANALYSES

Comparison 01. Azathioprine versus placebo or no intervention

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Mortality - complete patient's course analysis and best/worst case scenario			Relative Risk (Fixed) 95% CI	Subtotals only
02 Mortality - uncertainty method	2		Relative risk (Fixed) 95% CI	0.80 [0.49, 1.31]
03 Pruritus at one-year intervention	1	36	Relative Risk (Fixed) 95% CI	0.71 [0.28, 1.84]
04 Cirrhosis development	1	28	Relative Risk (Fixed) 95% CI	1.07 [0.58, 1.97]
05 Quality of life	2		Relatvie risks (Fixed) 95% CI	0.74 [0.50, 1.08]
06 Adverse events	2	287	Relative Risk (Fixed) 95% CI	2.44 [1.14, 5.20]

COVER SHEET

Title	Azathioprine for primary biliary cirrhosis
Authors	Gong Y, Christensen E, Gluud C
Contribution of author(s)	YG performed the searches, selected trials for inclusion, wrote to authors, performed data extraction and data analyses with EC, and drafted the protocol and the systematic review. CG formulated the idea of this review and revised the protocol, selected trials for inclusion, validated, solved discrepancy of data extraction between YG and EC, and revised the review.
Issue protocol first published	2006/2
Review first published	2007/3
Date of most recent amendment	21 May 2007
Date of most recent SUBSTANTIVE amendment	21 May 2007
What's New	Information not supplied by author
Date new studies sought but none found	26 October 2005
Date new studies found but not yet included/excluded	Information not supplied by author
Date new studies found and included/excluded	Information not supplied by author
Date authors' conclusions section amended	Information not supplied by author
Contact address	Dr Yan Gong Cochrane Hepato-Biliary Group Copenhagen Trial Unit, Centre for Clinical Intervention Research Rigshospitalet, Dept. 3344 Blegdamsvej 9 Copenhagen DK-2100 DENMARK

E-mail: ygong@ctu.rh.dk
Tel: +45 3545 7161
Fax: +45 3545 7101

DOI 10.1002/14651858.CD006000.pub2

Cochrane Library number CD006000

Editorial group Cochrane Hepato-Biliary Group

Editorial group code HM-LIVER

GRAPHS AND OTHER TABLES

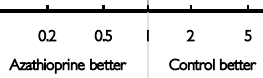
Analysis 01.01. Comparison 01 Azathioprine versus placebo or no intervention, Outcome 01 Mortality - complete patient's course analysis and best/worst case scenario

Review: Azathioprine for primary biliary cirrhosis

Comparison: 01 Azathioprine versus placebo or no intervention

Outcome: 01 Mortality - complete patient's course analysis and best/worst case scenario

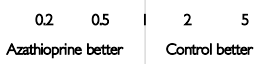
Study	Azathioprine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
01 Complete patient's course analysis					
Christensen 1985	57/78	62/77		86.5	0.91 [0.76, 1.08]
Heathcote 1976	7/19	10/20		13.5	0.74 [0.35, 1.54]
Subtotal (95% CI)	97	97		100.0	0.88 [0.74, 1.06]
Total events: 64 (Azathioprine), 72 (Control)					
Test for heterogeneity chi-square=0.32 df=1 p=0.57 I^2 =0.0%					
Test for overall effect z=1.36 p=0.2					
02 Assuming good outcme					
Christensen 1985	57/127	62/121		86.7	0.88 [0.68, 1.14]
Heathcote 1976	7/22	10/23		13.3	0.73 [0.34, 1.58]
Subtotal (95% CI)	149	144		100.0	0.86 [0.67, 1.10]
Total events: 64 (Azathioprine), 72 (Control)					
Test for heterogeneity chi-square=0.19 df=1 p=0.66 I^2 =0.0%					
Test for overall effect z=1.23 p=0.2					
03 Assuming poor outcome					
Christensen 1985	106/127	106/121		88.8	0.95 [0.86, 1.06]
Heathcote 1976	10/22	14/23		11.2	0.75 [0.43, 1.31]
Subtotal (95% CI)	149	144		100.0	0.93 [0.83, 1.04]
Total events: 116 (Azathioprine), 120 (Control)					
Test for heterogeneity chi-square=0.80 df=1 p=0.37 I^2 =0.0%					
Test for overall effect z=1.32 p=0.2					



(*Continued . . .*)

(. . . *Continued*)

Study	Azathioprine n/N	Control n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
04 Extreme case favouring azathioprine					
Christensen 1985	57/127	106/121		88.8	0.51 [0.42, 0.63]
Heathcote 1976	7/22	14/23		11.2	0.52 [0.26, 1.05]
Subtotal (95% CI)	149	144		100.0	0.51 [0.42, 0.63]
Total events: 64 (Azathioprine), 120 (Control)					
Test for heterogeneity chi-square=0.00 df=1 p=0.96 I^2 =0.0%					
Test for overall effect z=6.62 p<0.00001					
05 Extreme case favouring control					
Christensen 1985	106/127	62/121		86.7	1.63 [1.35, 1.97]
Heathcote 1976	10/22	10/23		13.3	1.05 [0.54, 2.01]
Subtotal (95% CI)	149	144		100.0	1.55 [1.29, 1.86]
Total events: 116 (Azathioprine), 72 (Control)					
Test for heterogeneity chi-square=1.66 df=1 p=0.20 I^2 =39.6%					
Test for overall effect z=4.69 p<0.00001					



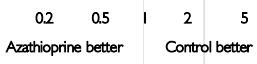
Analysis 01.02. Comparison 01 Azathioprine versus placebo or no intervention, Outcome 02 Mortality - uncertainty method

Review: Azathioprine for primary biliary cirrhosis

Comparison: 01 Azathioprine versus placebo or no intervention

Outcome: 02 Mortality - uncertainty method

Study	log [Relative risk] (SE)	Relative risk (Fixed) 95% CI	Weight (%)	Relative risk (Fixed) 95% CI
Christensen 1985	-0.09 (0.39)		39.6	0.91 [0.42, 1.97]
Heathcote 1976	-0.30 (0.32)		60.4	0.74 [0.40, 1.38]
Total (95% CI)			100.0	0.80 [0.49, 1.31]
Test for heterogeneity chi-square=0.17 df=1 p=0.68 I^2 =0.0%				
Test for overall effect z=0.88 p=0.4				

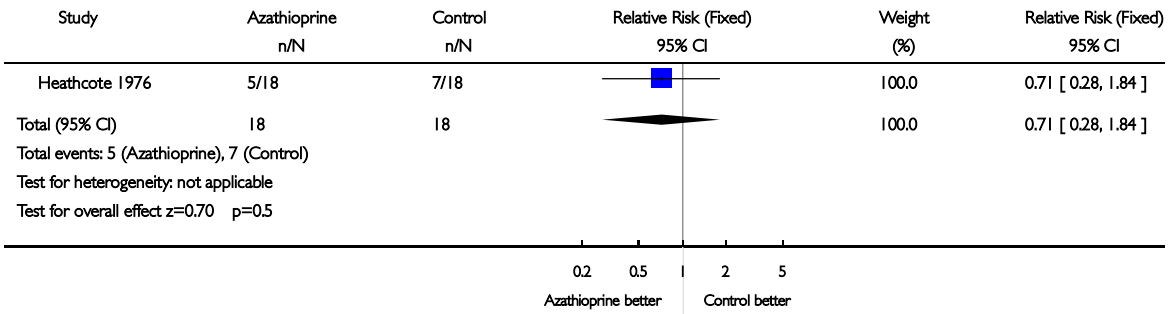


Analysis 01.03. Comparison 01 Azathioprine versus placebo or no intervention, Outcome 03 Pruritus at one-year intervention

Review: Azathioprine for primary biliary cirrhosis

Comparison: 01 Azathioprine versus placebo or no intervention

Outcome: 03 Pruritus at one-year intervention

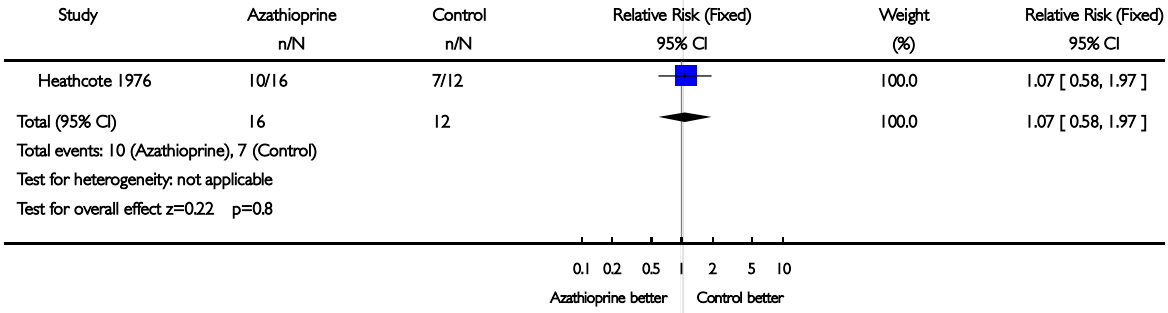


Analysis 01.04. Comparison 01 Azathioprine versus placebo or no intervention, Outcome 04 Cirrhosis development

Review: Azathioprine for primary biliary cirrhosis

Comparison: 01 Azathioprine versus placebo or no intervention

Outcome: 04 Cirrhosis development

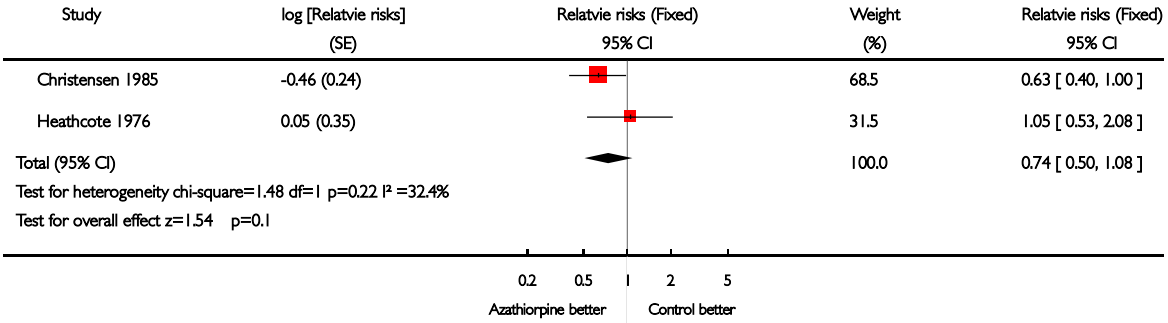


Analysis 01.05. Comparison 01 Azathioprine versus placebo or no intervention, Outcome 05 Quality of life

Review: Azathioprine for primary biliary cirrhosis

Comparison: 01 Azathioprine versus placebo or no intervention

Outcome: 05 Quality of life

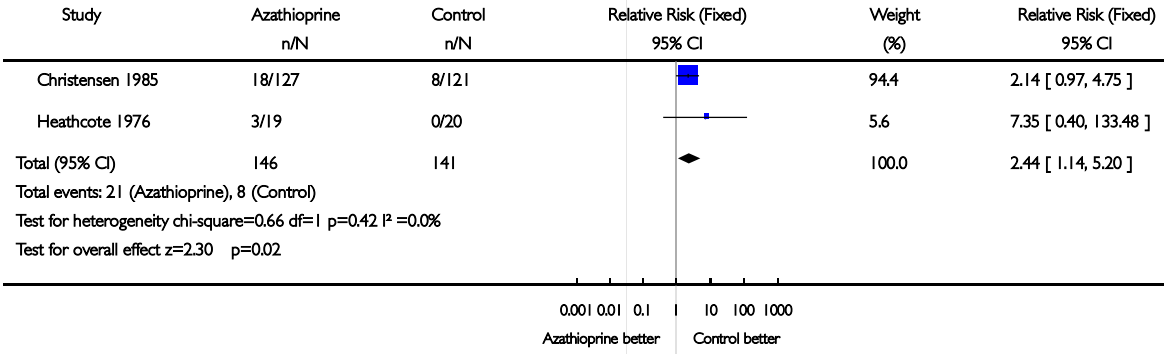


Analysis 01.06. Comparison 01 Azathioprine versus placebo or no intervention, Outcome 06 Adverse events

Review: Azathioprine for primary biliary cirrhosis

Comparison: 01 Azathioprine versus placebo or no intervention

Outcome: 06 Adverse events



Appendix 6

Cyclosporin A for primary biliary cirrhosis (Review)

Gong Y, Christensen E, Gluud C

This is a reprint of a Cochrane review, prepared and maintained by The Cochrane Collaboration and published in *The Cochrane Library* 2007, Issue 3

http://www.thecochranelibrary.com

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Cyclosporin A for primary biliary cirrhosis (Review)

Gong Y, Christensen E, Gluud C

Status: *New*

This record should be cited as:
Gong Y, Christensen E, Gluud C. Cyclosporin A for primary biliary cirrhosis. *Cochrane Database of Systematic Reviews* 2007, Issue 3. Art. No.: CD005526. DOI: 10.1002/14651858.CD005526.pub2.

This version first published online: 18 July 2007 in Issue 3, 2007.
Date of most recent substantive amendment: 21 May 2007

ABSTRACT

Background
Cyclosporin A has been used for patients with primary biliary cirrhosis, but the therapeutic responses in randomised clinical trials have been heterogeneous.

Objectives
To assess the beneficial and harmful effects of cyclosporin A for patients with primary biliary cirrhosis.

Search strategy
Relevant randomised clinical trials were identified by searching *The Cochrane Hepato-Biliary Group Controlled Trials Register,* the *Cochrane Central Register of Controlled Trials (CENTRAL)* in *The Cochrane Library, MEDLINE, EMBASE, Science Citation Index Expanded, The Chinese Biomedical Database,* and *LILACS,* and manual searches of bibliographies to June 2006. We contacted authors of trials and the company producing cyclosporin A.

Selection criteria
Randomised clinical trials comparing cyclosporin A with placebo, no intervention, or another drug were included irrespective of blinding, language, year of publication, and publication status.

Data collection and analysis
Our primary outcomes were mortality, and mortality or liver transplantation. Dichotomous outcomes were reported as relative risk (RR) and if appropriate, Peto odds ratio with 95% confidence interval (CI). Continuous outcomes were reported as weighted mean difference (WMD) or standardised mean difference (SMD). We examined intervention effects by random-effects and fixed-effect models.

Main results
We identified three trials with 390 patients that compared cyclosporin A versus placebo. Two of them were assessed methodologically adequate with low-bias risk. Cyclosporin A did not significantly reduce mortality risk (RR 0.92, 95% CI 0.59 to 1.45), and mortality or liver transplantation (RR 0.85, 95% CI 0.60 to 1.20). Cyclosporin A significantly improved pruritus (SMD -0.38, 95% CI -0.63 to -0.14), but not fatigue. Cyclosporin A significantly reduced alanine aminotransferase (WMD -41 U/L, 95% CI -63 to -18) and increased serum albumin level (WMD 1.66 g/L, 95% CI 0.26 to 3.05). Significantly more patients experienced adverse events in the cyclosporin A group than in the placebo group, especially renal dysfunction (Peto odds ratio 5.56, 95% CI 2.52 to 12.27) and hypertension (SMD 0.88, 95% CI 0.27 to 1.48).

Authors' conclusions
We found no evidence supporting or refuting that cyclosporin A may delay death, death or liver transplantation, or progression of primary biliary cirrhosis. Cyclosporin A caused more adverse events than placebo, like renal dysfunction and hypertension. We do not recommend the use of cyclosporin A outside randomised clinical trials.

PLAIN LANGUAGE SUMMARY

Cyclosporin A was without significant effects on mortality, liver transplantation, or progression of primary biliary cirrhosis, and patients given cyclosporin A experienced more adverse events

Primary biliary cirrhosis (PBC) is a chronic disease of the liver that is characterised by destruction of bile ducts. Estimates of annual incidence range from 2 to 24 people per million population, and estimates of prevalence range from 19 to 240 people per million population. PBC primarily affects middle-aged women. The forecast for the symptomatic patient after diagnosis is between 10 and 15 years. The cause of PBC is unknown, but the dynamics of the disease resemble the group 'autoimmune disease'. Therefore, one might expect a noticeable effect of administering an immune repressing drug (immunosuppressant). This review evaluates all clinical data on the immunosuppressant cyclosporin A for PBC.

The findings in this review are based on three clinical trials with 390 patients. The drug cyclosporin A was tested against placebo. The primary findings of the review are that cyclosporin A has no effect on survival or progression of the disease (cirrhosis development). Patients given cyclosporin A experienced more adverse events than patients given placebo, especially renal dysfunction and hypertension. There was significant improvement in itching (pruritus) and liver biochemistry, which were secondary outcome measures.

We cannot recommend the use of cyclosporin A outside randomised clinical trials.

BACKGROUND

Primary biliary cirrhosis is a chronic liver disease of unknown aetiology. Ninety per cent of patients with primary biliary cirrhosis are females and the majority are diagnosed after the age of 40 years (James 1981). Over the past 30 years, substantial increases in the prevalence of primary biliary cirrhosis has been observed (Kim 2000). Primary biliary cirrhosis is now a frequent cause of liver morbidity, and patients with primary biliary cirrhosis are significant users of health resources, including liver transplantation (Prince 2003). Primary biliary cirrhosis is diagnosed on the basis of the triad: antimitochondrial antibodies, found in over 95% of patients with primary biliary cirrhosis (Fregeau 1989; Lacerda 1995; Invernizzi 1997; Turchany 1997; Mattalia 1998); abnormal liver function tests that are typically cholestatic (with raised activity of alkaline phosphatases being the most frequently seen abnormality); and characteristic liver histological changes (Scheuer 1967) in the absence of extrahepatic biliary obstruction (Kaplan 1996).

Patients with primary biliary cirrhosis have been subjected to many drugs. Ursodeoxycholic acid (a bile acid) is the most extensively used drug in these patients (Verma 1999). Other drugs have been immunomodulatory and other agents, such as colchicine (Warnes 1987; Vuoristo 1995; Poupon 1996; Gong 2005b), prednisolone (Mitchison 1992; Prince 2005), chlorambucil (Hoofnagle 1986), azathioprine (Heathcote 1976; Christensen 1985), D-penicillamine (Dickson 1985; Neuberger 1985; Gong 2004), methotrexate (Kaplan 1991; Lindor 1995; Gong 2005a), or cyclosporin A (Minuk 1988; Wiesner 1990; Gong 2005c).

Cyclosporin A has proved effective in preventing immune-mediated rejection of a variety of transplanted human allografts (Cohen 1984) and has been shown to produce clinical improvement in a number of autoimmune conditions (Tugwell 1990). Cyclosporin A is a cyclic endecapeptide of fungal origin. It alters lymphokine production so that the T-helper-inducer subpopulations are attenuated, T-cell help required for B-cell activation is blocked, cytotoxic T-cell generation is attenuated, and T-suppressor cell subpopulations are expanded (Harris 1987). Thus, cyclosporin A would appear a potential ideal agent to modify the immunologic irregularities in primary biliary cirrhosis (James 1983). Since 1980, when Routhier showed beneficial effects of cyclosporin A on serum aspartate transaminase and alkaline phosphatases in six patients with primary biliary cirrhosis (Routhier 1980), several randomised clinical trials have been carried out with different results (Minuk 1988; Wiesner 1990). We could not identify any meta-analyses or systematic reviews on the beneficial and harmful effects of cyclosporin A in primary biliary cirrhosis.

OBJECTIVES

To systematically assess the beneficial and harmful effects of cyclosporin A for patients with primary biliary cirrhosis.

CRITERIA FOR CONSIDERING STUDIES FOR THIS REVIEW

Types of studies

We included randomised clinical trials irrespective of blinding, language, year of publication, and publication status. We excluded studies using quasi-randomisation (for example, allocation by date of birth).

Types of participants

Patients with primary biliary cirrhosis, ie, patients having at least two of the following: elevated serum activity of alkaline phosphatases (or other markers of intrahepatic cholestasis), and/or a positive result for serum mitochondrial antibody, and/or liver biopsy findings diagnostic for or compatible with primary biliary cirrhosis.

Types of intervention

Administration of any dose of cyclosporin A versus placebo or no intervention or other drugs. Co-interventions were allowed as long as the intervention arms of the randomised clinical trial received similar co-interventions.

Types of outcome measures

Primary outcome measures

- Mortality.
- Mortality or liver transplantation.

Secondary outcome measures

- Pruritus: number of patients without improvement of pruritus or pruritus score.
- Fatigue: number of patients without improvement of fatigue or fatigue score.
- Incidence of liver complications: number of patients developing variceal bleeding, ascites, hepatic encephalopathy, jaundice, or hepato-renal syndrome.
- Liver biochemistry: serum (s-)bilirubin; s-alkaline phosphatases; s-gamma-glutamyltransferase; s-aspartate aminotransferase; s-alanine aminotransferase; s-albumin; s-cholesterol (total); plasma immunoglobulins.
- Liver biopsy: worsening of liver histological stage or score.
- Quality of life: physical functioning (ability to carry out activities of daily living such as self-care and walking around), psychological functioning (emotional and mental well-being), social functioning (social relationships and participation in social activities), and perception of health, pain, and overall satisfaction with life.
- Adverse events (excluding mortality and liver transplantation). The adverse event is defined as any untoward medical occurrence in a patient in either of the two arms of the included randomised clinical trials, which did not necessarily have a causal relationship with the treatment, but did, however, result in a dose reduction, discontinuation of treatment, or registration of the advent as an adverse event/side effect (ICH-GCP 1997).
- Cost-effectiveness: the estimated costs connected with the interventions were to be weighed against any possible health gains.

SEARCH METHODS FOR IDENTIFICATION OF STUDIES

See: Cochrane Hepato-Biliary Group methods used in reviews.

We identified relevant randomised clinical trials by searching *The Cochrane Hepato-Biliary Group Controlled Trials Register,* which involves hand searches of major hepatology journals and conference proceedings, the *Cochrane Central Register of Controlled Trials (CENTRAL)* in *The Cochrane Library, MEDLINE,EMBASE, Science Citation Index Expanded, The Chinese Biomedical Database,* and *LILACS* (Royle 2003). The search strategies are given in Table 01 with the time span of the searches.

We tried to identify further trials by reading the reference lists of the identified publications. We wrote to the principal authors of the identified trials and to the researchers active in the field to inquire about additional randomised clinical trials they might know of. We also contacted the pharmaceutical company, Novartis, producer of cyclosporin A, to obtain any unidentified or unpublished randomised clinical trials.

METHODS OF THE REVIEW

We performed a meta-analysis following the protocol (Gong 2005c) and the recommendations given by the *Cochrane Handbook for Systematic Reviews of Interventions* (Higgins 2006) and the *Cochrane Hepato-Biliary Group Module* (Gluud 2007).

Data extraction

Two authors (YG and EC) independently evaluated whether the identified trials fulfilled the inclusion criteria. We listed the excluded trials in 'Characteristics of excluded studies' with the reasons for exclusion. YG extracted data and EC validated the data extraction. Disagreements were resolved by discussion with CG. We wrote to the authors of the included trials and asked them to specify the data of interest, if they had not been reported clearly in the publications.

Assessment of methodological quality of included trials

We assessed the methodological quality of the randomised clinical trials using four components (Schulz 1995; Moher 1998; Kjaergard 2001). High-quality trials, ie, trials with low-bias risk, were considered adequate on two out of the first three components.

Generation of the allocation sequence

- Adequate, if the allocation sequence was generated by a computer or random number table. Drawing of lots, tossing of a coin, shuffling of cards, or throwing dice was considered as adequate if a person who was not otherwise involved in the recruitment of participants performed the procedure;
- Unclear, if the trial was described as randomised, but the method used for the allocation sequence generation was not described.

Allocation concealment

- Adequate, if the allocation of patients involved a central independent unit, on-site locked computer, identically appearing numbered drug bottles or containers prepared by an independent pharmacist or investigator, or sealed envelopes;
- Unclear, if the trial was described as randomised, but the method used to conceal the allocation was not described;
- Inadequate, if the allocation sequence was known to the investigators who assigned participants.

Blinding (or masking)

- Adequate, if the trial was described as double blind and the method of blinding involved identical placebo or active drug;
- Unclear, if the trial was described as double blind, but the method of blinding was not described;
- Not performed, if the trial was not double blind.

Follow-up

- Adequate, if the numbers and reasons for dropouts and withdrawals in all intervention groups were described or if it was specified that there were no dropouts or withdrawals;
- Unclear, if the report gave the impression that there had been no dropouts or withdrawals, but this was not specifically stated;
- Inadequate, if the number or reasons for dropouts and withdrawals were not described.

Characteristics of patients

Number of patients randomised; patient inclusion and exclusion criteria; mean (or median) age; sex ratio; histological stage; number of patients lost to follow-up.

Characteristics of interventions

Type, dose, and form of cyclosporin A intervention; type of intervention in the control group and collateral interventions (if any); duration of treatment and follow-up.

Characteristics of outcomes

All outcomes were extracted from each included trial. We analysed the outcome measures at maximum follow-up.

Statistical methods

We used the statistical package RevMan Analyses 1.0 (RevMan 2003) provided by The Cochrane Collaboration. We presented dichotomous data as relative risk (RR) with 95% confidence interval (CI). Peto odds ratio (OR) was used to combine rare event data (less than 5%). We presented continuous outcome measures by weighted mean differences (WMD) with 95% CI. We used standardised mean differences (SMD) to combine dichotomous data and continuous data on pruritus, fatigue, and blood pressure (Higgins 2006).

We examined intervention effects by using both a random-effects model (DerSimonian 1986) and a fixed-effect model (Mantel 1959) with the significant level set at $P < 0.05$. If the results of the two analyses concurred, we presented only the results of the fixed-effect model. In case of discrepancies of the two models, we reported the results of both models. We explored the presence of statistical heterogeneity by chi-squared test with significance set at $P < 0.10$ and measured the quantities of heterogeneity by I^2 (Higgins 2002) .

Due to small number of trials included, we did not perform subgroup analysis, sensitivity analysis, and statistical tests to explore publication bias and other biases, which were planned in the protocol (Gong 2005c).

DESCRIPTION OF STUDIES

We identified a total of 269 references through electronic searches of *The Cochrane Hepato-Biliary Group Controlled Trials Register* (n = 61), the *Cochrane Central Register of Controlled Trials* (*CENTRAL*) in *The Cochrane Library* (n = 54), *MEDLINE* (n = 31), *EMBASE* (n = 45), *Science Citation Index Expanded* (n = 35), *The Chinese Biomedical CD Database* (n = 43), and *LILACS* (n = 0). We excluded 254 duplicates and clearly irrelevant references by reading abstracts. Accordingly, 15 references were retrieved for further assessment. Of these, we excluded nine because they were non-randomised clinical studies or observational studies. The remaining six references referred to three randomised clinical trials involving 390 patients with primary biliary cirrhosis, which fulfilled our inclusion criteria. The publication year of the trials ranged from year 1988 to 1993. All trials were published as full papers.

All the trials compared cyclosporin A versus placebo. The formulation included was the original one, not microemulsion and topical emulsion. The mean age of the patients was about 52 years. The majority of the patients were women (women/men: 338/52). Slightly more patients had stage III or IV than stage I or II (178/154). The dose of cyclosporin A was 2.5, 3, or 4 mg/kg/day. The duration of treatment and follow-up varied from one to three years (See 'Characteristics of included studies').

METHODOLOGICAL QUALITY

None of the trials, except Lombard 1993, had adequate generation of the allocation sequence. Allocation concealment was adequate in two trials (Minuk 1988; Lombard 1993) and unclear in Wiesner 1990. Blinding was adequate in all trials. Follow-up was adequately reported in all the trials. In total, 74 patients (19%) were lost to follow-up: 46 (23%) patients in the cyclosporin A group and 28 (15%) in the placebo group. None of the trials reported a sample size estimate. Lombard 1993 reported that they used intention-

to-treat analyses. Overall, two trials were regarded as low-bias risk trials (Minuk 1988; Lombard 1993).

RESULTS

Mortality

Three trials with 390 patients provided data to estimate the risk of mortality of cyclosporin A versus placebo (Comparison 01-01). Compared with placebo, cyclosporine A did not significantly affect mortality (15% versus 17%). The relative risk was 0.92 (95% CI 0.59 to 1.45).

Mortality or liver transplantation

Compared with placebo, cyclosporine A did not significantly affect mortality or liver transplantation (22% versus 27%) (Comparison 01-02). The relative risk of mortality or liver transplantation was 0.85 (95% CI 0.60 to 1.20).

Pruritus, fatigue, and liver complications

Cyclosporin A significantly improved pruritus (SMD -0.38, 95% CI -0.63 to -0.14), but did not significantly have an affect on fatigue (SMD -0.35, 95% CI -1.16 to 0.46). We were not able to locate data on liver complications because of poor reporting.

Liver biochemical and histological outcomes

Regarding liver biochemistry (Comparison 01-105 to 01-10), cyclosporin A appeared to decrease the levels of s-bilirubin, s-alanine aminotransferase, and s-alkaline phosphatases except for the levels of immunoglobulin M. Cyclosporin A also increased s-albumin compared to the placebo group. Lombard et al used log transformed data on serum bilirubin, alkaline phosphatases, and aminotransferase for comparisons which prevented us from combining the data from all the three trials (Lombard 1993). Wiesner et al reported data on liver biopsy: histologic progression to at least one more stage and increased or unaltered portal inflammation (Wiesner 1990). There was no significant difference between cyclosporin A and placebo (Comparison 01-10).

Adverse events

In the largest trial (Lombard 1993), 34 out of 176 patients given cyclosporin A had adverse events that led to permanent discontinuation of the treatment versus 18 out of 173 patient given placebo (RR 1.86, 95% CI 1.09 to 3.16). All the three trials reported on other adverse events not necessitating permanent discontinuation of treatment (RR 1.41, 95% CI 1.15 to 1.73). The risks of such adverse events were significantly increased in the cyclosporin A treated patients. Among the adverse events, cyclosporin A significantly increased the risk of renal dysfunction (Peto OR 5.56, 95% CI 2.52 to 12.27). Cyclosporine significantly increased the blood pressure (SMD 0.88, 95% CI 0.27 to 1.48) as defined by a rise in the diastolic pressure above 5 mmHg since the previous visit (Lombard 1993) or an increase of $\geq$ 25 mmHg in the systolic pressure or $\geq$ 12 mmHg in the diastolic pressure (Wiesner 1990).

Quality of life and cost-effectiveness

None of the trials examined specific quality-of-life scales or cost-effectiveness.

Regarding the subgroup and sensitivity analyses, they were not done because of the limited number of trials.

DISCUSSION

Cyclosporin A did not significantly influence the risk of mortality or liver transplantation in patients with primary biliary cirrhosis, nor did it delay liver histological progression. Cyclosporin A seemed to ameliorate the patients' pruritus, but not fatigue. Cyclosporin A appeared to decrease the concentration of serum bilirubin and the activities of alanine aminotransferase and alkaline phosphatases. Patients given cyclosporin A experienced significantly more adverse events, especially renal dysfunction and hypertension.

To our knowledge, only three trials have been conducted to evaluate the effects of cyclosporin A for patients with primary biliary cirrhosis. Therefore, this systematic review has a major limitation: the small number of trials included (Ioannidis 2001). Furthermore, all the trials had shorter follow-up than the estimated median survival of primary biliary cirrhosis, ie, 10 years to 15 years (Prince 2003). Therefore, it is difficult to detect a significant difference on mortality or liver transplantation.

Patients given cyclosporin A had not significantly lower risk of death and liver transplantation. Since two of the trials had a short trial duration (Minuk 1988; Wiesner 1990), few patients died during the period. In the largest trial by Lombard et al, patients were treated and followed up to six years. A total of 30 patients in the cyclosporin A group died and an additional 14 patients required liver transplantation, compared with 31 deaths and 15 transplants in the placebo group (Lombard 1993). When we combined the data, we found no significant difference on deaths and/or liver transplantations between the two groups. The heterogeneity was moderate (I^2 = 41.4%) in spite of the disparity on trial duration. Lombard et al found a survival benefit (including death or liver transplantation) only after adjustment for a seemly imbalance in pretreatment variables (Lombard 1993). However, they did not find the same beneficial effect when adjustment was not applied (logrank P = 0.63). Furthermore, they did not confirm a beneficial effect in reducing the risk of death only - neither without nor with the adjustment (logrank P = 0.87; Cox model P = 0.14). Therefore, we are not convinced of a beneficial effect of cyclosporin A on patients' survival and liver transplantation.

It seems that cyclosporin A improved the symptom of pruritus, which is one of the major complaints of the disease. But this finding should be interpreted with great caution. First of all, the pooling method here is based on an assumption that the underlying distribution of the pruritus score in each treatment group follows a logistic distribution, which might not be the case. Secondly, since

pruritus is a subjective assessment, depending on patient's threshold and physician's experience, the potential improvement caused by cyclosporin A needs to be further investigated. We cannot exclude that blinding might have been broken in the trials because of, eg, occurrence of adverse events. This actually happened in the Wiesner 1990 trial. Such unblinding might have biased the assessment of pruritus (Schulz 1995; Kjaergard 2001).

Cyclosporin A seems to have beneficial effect in reducing the activity of alanine aminotransferase and in increasing serum albumin level. The variety of reporting did not allow us to integrate the data on serum bilirubin and alkaline phosphatases, which were found to be improved in Wiesner 1990 and Lombard 1993 trials. None of the three trials have found that cyclosporin A delayed the histological progression (including the assessment of inflammation or fibrosis).

Our review shows a benefit from treatment with cyclosporin A on pruritus and liver biochemistry and poses the question as to whether the shown benefits statistically outweigh the adverse events. Lombard et al reported that more patients in the cyclosporin A group experienced adverse events warranting discontinuation and that the proportion of patients with discontinuation was significantly higher than in the placebo group (Lombard 1993). Most of the adverse events were renal impairment, hypertension, and infective episodes. All the three trials reported adverse events not necessitating permanent discontinuation of treatment. Patients given cyclosporin A experienced significantly more adverse events with the majority being hirsutism, increased blood pressure, and a slight increase in viral or bacterial infection occurrence.

For cyclosporin A, nephrotoxicity and hypertension are adverse events of major concerns. We have, therefore, also extracted the data on these adverse events. Our analyses show that significantly more patients given cyclosporin A had renal dysfunction as defined by creatinine persistently above 141 μmol/L (Wiesner 1990; Lombard 1993). In a majority of the patients, reducing the dose or discontinuing cyclosporin A temporarily was associated with the resolution of the adverse events. On the other hand, no dynamic renal function tests were undertaken in the trials, and it must be conceded that serum creatinine elevation probably underestimates the incidence of nephrotoxicity. Our result demonstrates that cyclosporin A treated patients significantly increased blood pressure. In general, hypertension was easily controlled with medical therapy when indicated (Wiesner 1990).

AUTHORS' CONCLUSIONS

Implications for practice

Despite improvements in pruritus and liver biochemical variables, cyclosporin A did not delay the progression to death or liver transplantation, or to an advanced histological stage. In addition, patients given cyclosporin A experienced more adverse events, especially renal dysfunction and hypertension. We do not recommend the use of cyclosporin A outside randomised clinical trials.

Implications for research

Further randomised clinical trials need to investigate the short-term and long-term effects of cyclosporin A on progression of the disease, need for liver transplantation, and survival. The potential benefits in pruritus and liver biochemistry also need to be further investigated. Future trials need to be closely monitored because of the adverse events, especially renal dysfunction and hypertension. Future trials ought to be reported according to the recommendations of the CONSORT Group (http://www.consort-statement.org/).

POTENTIAL CONFLICT OF INTEREST

None known.

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* *Indicates the major publication for the study*

TABLES

Characteristics of included studies

Study	**Lombard 1993**
Methods	Generation of the allocation sequence: a schedule of block randomisation - considered adequate. Allocation concealment: a 'blinded' investigator - considered adequate. Blinding: patients and investigators - considered adequate. Follow-up: 40 in cyclosporin A group and 25 in placebo group were lost to follow-up - considered adequate.
Participants	Country: UK. Mean age: 53.9 years in cyclosporin A group, 54.2 years in placebo group. Female/Male: 298/51. PBC stage status: stage I/II: 62 in cyclosporin A group, 71 in placebo group; stage III/IV: 87 in cyclosporin A group, 71 in placebo group.
Interventions	Cyclosporin A: 3 mg/kg/day (n = 176); Placebo (n = 173). Median follow-up: 928 days (range 6 to 2146 days).
Outcomes	(1) Mortality and liver transplantation. (2) Clinical outcomes and liver biochemical variables. (3) Adverse events.

Characteristics of included studies (*Continued*)

Notes	(1) Two types of analysis were presented: the first one was on death (the end point) and liver transplantation censored at time of transplantation; the second one combined death and liver transplantation. (2) Correspondence sent to the author on 8 June 2005. No reply was received.
Allocation concealment	A – Adequate

Study	**Minuk 1988**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: sealed envelopes - considered adequate. Blinding: patients - considered adequate. Follow-up: no one lost to follow-up - considered adequate.
Participants	Country: Canada. Mean age: 50.7 years in cyclosporin A group, 58.6 years in placebo group. Female/Male: 11/1 PBC stage status: stage I/II: 3 in cyclosporin A group, 2 in placebo group; stage III/IV: 3 in cyclosporin A group, 4 in placebo group.
Interventions	Cyclosporin A: 2.5 mg/kg/day (n = 6); Placebo (n = 6). Treatment: one year Posttreatment follow-up: 6 months.
Outcomes	(1) Mortality and liver transplantation. (2) Clinical outcomes and liver biochemical variables. (3) Histological assessment. (4) Adverse events.
Notes	(1) Correspondence sent to the author on 8 June 2005. His email with information on methodological quality was received on the same day.
Allocation concealment	D – Not used

Study	**Wiesner 1990**
Methods	Generation of the allocation sequence: unclear. Allocation concealment: unclear. Blinding: patients and investigators were planned to be 'blinded'. However, the assessment of the 'blinding' effectiveness revealed that a considerable unblinding did occur, so we considered it inadequate. Follow-up: 6 in cyclosporin A group and 3 in placebo group were lost to follow-up - we considered it adequate.
Participants	Country: US. Mean age: 45.5 years in cyclosporin A group, 48.0 years in placebo group. Female/Male: 29/0 PBC stage status: stage I/II: 11 in cyclosporin A group, 5 in placebo group; stage III/IV: 8 in cyclosporin A group, 5 in placebo group.
Interventions	Cyclosporin A: 4 mg/kg/day (n = 19); Placebo (n = 10). Median follow-up: 2.7 years.
Outcomes	(1) Mortality and liver transplantation. (2) Clinical outcomes and liver biochemical variables. (3) Histological assessment. (4) Adverse events.

Notes	(1) This trial only included precirrhotic patients with primary biliary cirrhosis. (2) It was a preliminary report of first 29 patients out of 59. (3) Correspondence sent to the author on 8 June 2005. No reply was received.
Allocation concealment	B – Unclear

Characteristics of excluded studies

Study	Reason for exclusion
Chau 2001	The authors described the histological patterns of rejection in liver transplant recipients using induction therapies with cyclosporin and tacrolimus monotherapy compared with standard triple therapy as historical control.
Dmitrewski 1996	The authors have examined the liver allograft biopsies taken at 1 and 2 years after transplantation from patients receiving either FK-506 or cyclosporin as part of a multi-centre trial. The objective was to study the recurrence of primary biliary cirrhosis in the liver allograft.
McMichael 1993	A randomised concentration-controlled clinical trial was performed to discover important concentration response relationships of FK-506, a potent immunosuppressive agent for prevention and treatment of graft rejection.
McMichael 1996	This is a computer-guided randomised concentration-controlled trials of tacrolimus in autoimmunity: multiple sclerosis and primary biliary cirrhosis.
Mueller 1995	In the present study, 121 patients, 61 randomly assigned to FK-506- and 60 assigned to cyclosporin A-based immunosuppression, were analysed according to the primary diagnosis for liver transplantation.
Robert 2003	A clinical review article to discuss the specific treatment to primary biliary cirrhosis.
Sanchez 2003	Data were obtained from prospectively maintained liver-transplant database and evaluated statistically to determine the recurrence of primary biliary cirrhosis.
Slitzky 1990	A clinical review article discussing the approaches to the treatment of primary biliary cirrhosis.
von Graffenried 1985	In this paper, the authors reported the presently available experience with regard to renal function in patients with autoimmune diseases treated with cyclosporin A.

ADDITIONAL TABLES

Table 01. Search strategies

Database	Time span	Search strategy
The Cochrane Hepato-Biliary Group Controlled Trials Register	June 2006.	'primary biliary cirrhosis' and 'cyclosporin A'
Cochrane Central Register of Controlled Trials (CENTRAL) in The Cochrane Library	Issue 2, 2006.	#1 = LIVER CIRRHOSIS BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = pbc #5 = #1 or #2 or #3 or #4 #6 = CYCLOSPORIN A: MESH #7 = IMMUNOSUPPRESSIVE AGENTS: MESH #8 = cyclosporins #9 = #6 or #7 or #8 #10 = #5 and #9

Table 01. Search strategies (*Continued*)

Database	Time span	Search strategy
MEDLINE	1966 to June 2006.	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = PBC #5 = #1 or #2 or #3 or #4 #6 = CYCLOSPORIN A: MESH #7 = IMMUNOSUPPRESSIVE AGENTS: MESH #8 = cyclosporin* #9 = immunosuppressive agent* #10 = #6 or #7 or #8 or #9 #11 = #5 and #10 #12 = random* or placebo* or blind* or meta-analysis #13 = #11 and #12
EMBASE	1980 to June 2006.	#1 = PRIMARY-BILIARY-CIRRHOSIS: MESH #2 = BILIARY-CIRRHOSIS: MESH #3 = primary and biliary and cirrhosis #4 = primary biliary cirrhosis #5 = PBC #6 = #1 or #2 or #3 or #4 or #5 #7 = CYCLOSPORIN A: MESH #8 = IMMUNOSUPPRESIVE AGENTS: MESH #9 = cyclosporin* #10 = immunosuppressive agent* #11 = #7 or #8 or #9 or #10 #12 = #6 and #11 #13 = random* or placebo* or blind* or meta-analysis #14 = #12 and #13
Science Citation Index Expanded (http://portal.isiknowledge.com/portal.cgi?DestApp=WOS&Func=Frame)	1945 to June 2006.	#1 = TS=(primary biliary cirrhosis OR PBC) #2 = TS=(cyclosporine OR cyclosporin*) #3 = #2 AND #1 #4 = TS=(random* OR blind* OR placebo* OR meta-analysis) #5 = #4 AND #3
LILACS	1982 to June 2006.	#1 = (primary and biliary and cirrhosis) or (primary biliary cirrhosis) #2 = primary biliary cirrhosis #3 = cyclosporin A #4 = (#1 OR #2) AND # 3
Chinese Biochemical CD Database	1979 to June 2006.	#1 = LIVER-CIRRHOSIS-BILIARY: MESH #2 = primary and biliary and cirrhosis #3 = primary biliary cirrhosis #4 = PBC #5 = #1 or #2 or #3 or #4 #6 = CYCLOSPORIN A: MESH #7 = IMMUNOSUPPRESSIVE AGENTS: MESH #8 = cyclosproin* #9 = immunosuppressive agent*

Table 01. Search strategies (*Continued*)

Database	Time span	Search strategy
		#10 = #6 or #7 or #8 or #9 #11 = #5 and #10 #12 = random* or placebo* or blind* or meta-analysis #13 = #11 and #12

ANALYSES

Comparison 01. Cyclosporin A versus placebo

Outcome title	No. of studies	No. of participants	Statistical method	Effect size
01 Mortality	3	390	Relative Risk (Fixed) 95% CI	0.92 [0.59, 1.45]
02 Mortality and/or liver transplantation	3	390	Relative Risk (Fixed) 95% CI	0.85 [0.60, 1.20]
03 Pruritus score and number of patients with the improvements	3		SMD (Fixed) 95% CI	-0.38 [-0.63, -0.14]
04 Fatigue score and number of patients with the improvements	2		SMD (Fixed) 95% CI	-0.35 [-1.16, 0.46]
05 Bilirubin (µmol/L)			Weighted Mean Difference (Fixed) 95% CI	Subtotals only
06 Alanine aminotransferase (U/L)	2	39	Weighted Mean Difference (Fixed) 95% CI	-40.55 [-63.38, -17.71]
07 Alkaline phosphatases (U/L) (change from baseline)			Weighted Mean Difference (Fixed) 95% CI	Subtotals only
08 Immunoglobulin M (g/L)	2	39	Weighted Mean Difference (Fixed) 95% CI	-1.05 [-2.71, 0.62]
09 Serum albumin (g/L)	3	388	Weighted Mean Difference (Fixed) 95% CI	1.66 [0.26, 3.05]
10 Histologic assessment			Relative Risk (Fixed) 95% CI	Subtotals only
11 Adverse event	4	739	Relative Risk (Fixed) 95% CI	1.49 [1.23, 1.81]
12 Renal dysfunction	2	378	Peto Odds Ratio 95% CI	5.56 [2.52, 12.27]
13 Increased blood pressure	3		SMD (Fixed) 95% CI	0.88 [0.27, 1.48]

COVER SHEET

Title	Cyclosporin A for primary biliary cirrhosis
Authors	Gong Y, Christensen E, Gluud C
Contribution of author(s)	YG performed the searches, selected trials for inclusion, wrote to authors and pharmaceutical companies, performed data extraction and data analyses, and drafted the protocol and the review. EC validated data extraction and revised the protocol and the review. CG formulated the idea of this review and revised the protocol, arbitrated disagreements on data extraction, validated data analyses, and revised the protocol and the review.
Issue protocol first published	2005/4
Review first published	2007/3
Date of most recent amendment	22 May 2007
Date of most recent SUBSTANTIVE amendment	21 May 2007
What's New	Information not supplied by author

Date new studies sought but none found	30 June 2005
Date new studies found but not yet included/excluded	Information not supplied by author
Date new studies found and included/excluded	09 April 2005
Date authors' conclusions section amended	04 July 2005
Contact address	Dr Yan Gong Cochrane Hepato-Biliary Group Copenhagen Trial Unit, Centre for Clinical Intervention Research Rigshospitalet, Dept. 3344 Blegdamsvej 9 Copenhagen DK-2100 DENMARK E-mail: ygong@ctu.rh.dk Tel: +45 3545 7161 Fax: +45 3545 7101
DOI	10.1002/14651858.CD005526.pub2
Cochrane Library number	CD005526
Editorial group	Cochrane Hepato-Biliary Group
Editorial group code	HM-LIVER

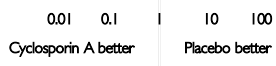
GRAPHS AND OTHER TABLES

Analysis 01.01. Comparison 01 Cyclosporin A versus placebo, Outcome 01 Mortality

Review: Cyclosporin A for primary biliary cirrhosis
Comparison: 01 Cyclosporin A versus placebo
Outcome: 01 Mortality

Study	Cyclosporin A n/N	Placebo n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Lombard 1993	30/176	31/173		95.4	0.95 [0.60, 1.50]
Minuk 1988	0/6	1/6		4.6	0.33 [0.02, 6.86]
x Wiesner 1990	0/19	0/10		0.0	Not estimable
Total (95% CI)	201	189		100.0	0.92 [0.59, 1.45]

Total events: 30 (Cyclosporin A), 32 (Placebo)
Test for heterogeneity chi-square=0.45 df=1 p=0.50 I^2 =0.0%
Test for overall effect z=0.35 p=0.7



Analysis 01.02. Comparison 01 Cyclosporin A versus placebo, Outcome 02 Mortality and/or liver transplantation

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 02 Mortality and/or liver transplantation

Study	Cyclosprin A n/N	Placebo n/N	Relative Risk (Fixed) 95% CI	Weight (%)	Relative Risk (Fixed) 95% CI
Lombard 1993	44/176	46/173		87.8	0.94 [0.66, 1.34]
Minuk 1988	0/6	2/6		4.7	0.20 [0.01, 3.46]
Wiesner 1990	1/19	3/10		7.4	0.18 [0.02, 1.48]
Total (95% CI)	201	189		100.0	0.85 [0.60, 1.20]

Total events: 45 (Cyclosprin A), 51 (Placebo)

Test for heterogeneity chi-square=3.41 df=2 p=0.18 I^2 =41.4%

Test for overall effect z=0.94 p=0.3

Analysis 01.03. Comparison 01 Cyclosporin A versus placebo, Outcome 03 Pruritus score and number of patients with the improvements

Review: Cyclosporin A for primary biliary cirrhosis

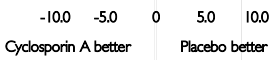
Comparison: 01 Cyclosporin A versus placebo

Outcome: 03 Pruritus score and number of patients with the improvements

Study	SMD (SE)	SMD (Fixed) 95% CI	Weight (%)	SMD (Fixed) 95% CI
Lombard 1993	-0.37 (0.13)		95.2	-0.37 [-0.62, -0.12]
Minuk 1988	-0.28 (0.65)		3.6	-0.28 [-1.55, 0.99]
Wiesner 1990	-1.77 (1.17)		1.1	-1.77 [-4.06, 0.51]
Total (95% CI)			100.0	-0.38 [-0.63, -0.14]

Test for heterogeneity chi-square=1.46 df=2 p=0.48 I^2 =0.0%

Test for overall effect z=3.09 p=0.002

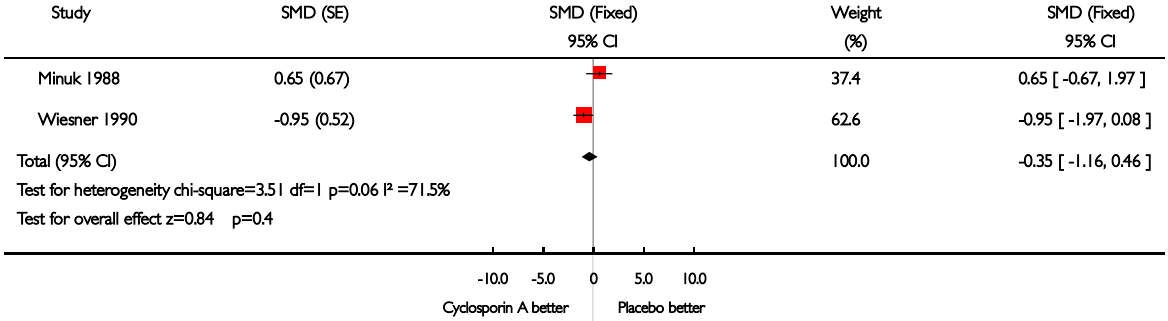


Analysis 01.04. Comparison 01 Cyclosporin A versus placebo, Outcome 04 Fatigue score and number of patients with the improvements

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 04 Fatigue score and number of patients with the improvements

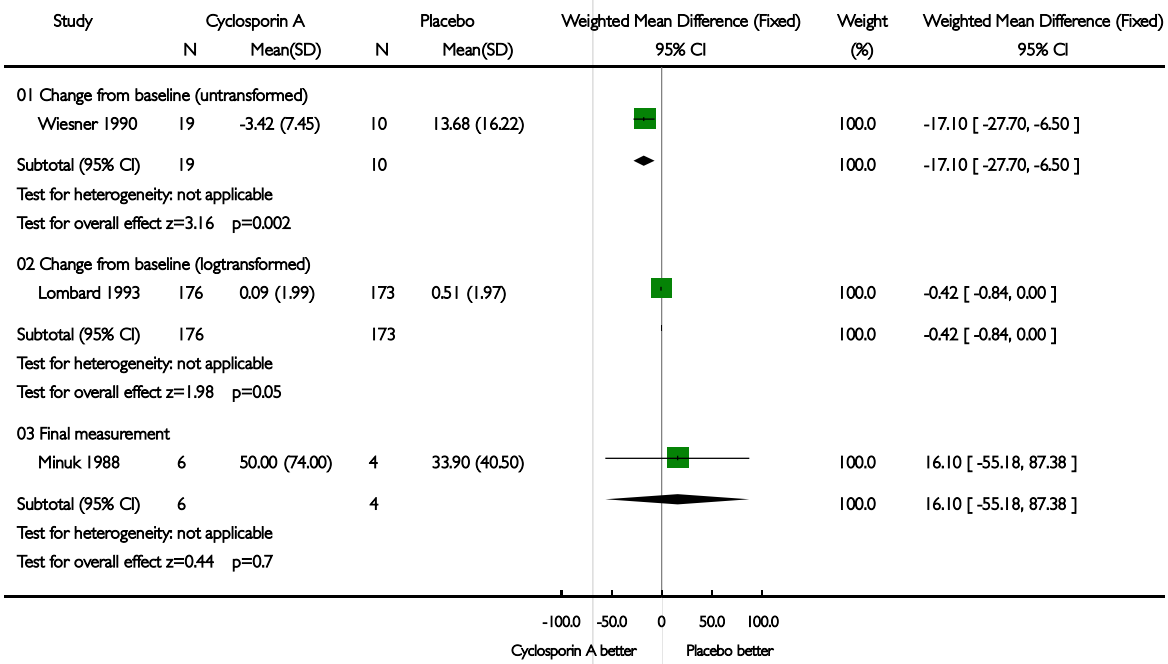


Analysis 01.05. Comparison 01 Cyclosporin A versus placebo, Outcome 05 Bilirubin (µmol/L)

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 05 Bilirubin (µmol/L)

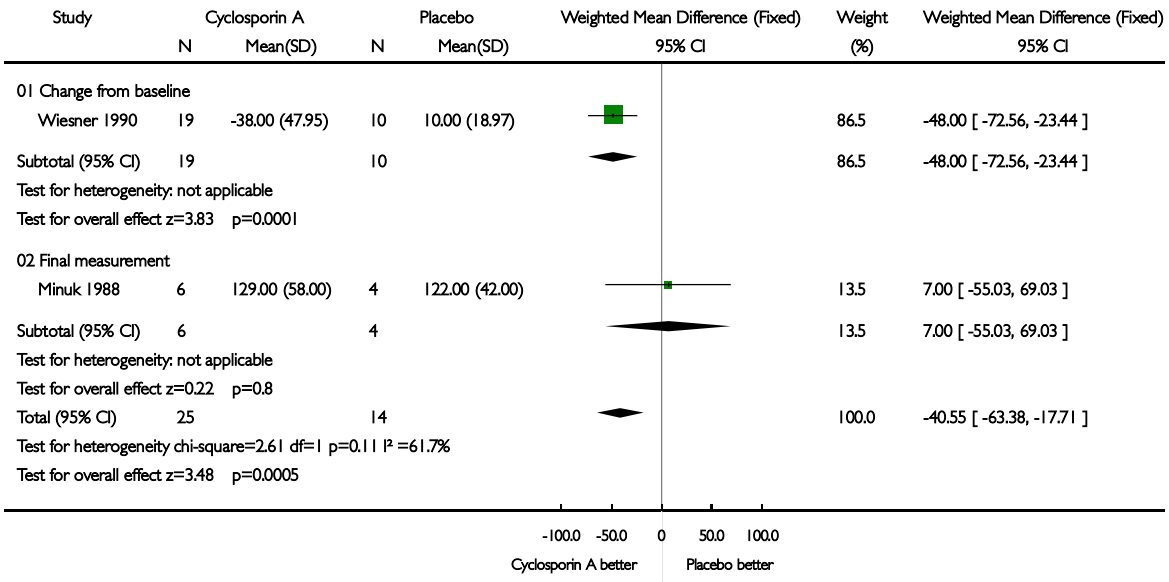


Analysis 01.06. Comparison 01 Cyclosporin A versus placebo, Outcome 06 Alanine aminotransferase (U/L)

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 06 Alanine aminotransferase (U/L)

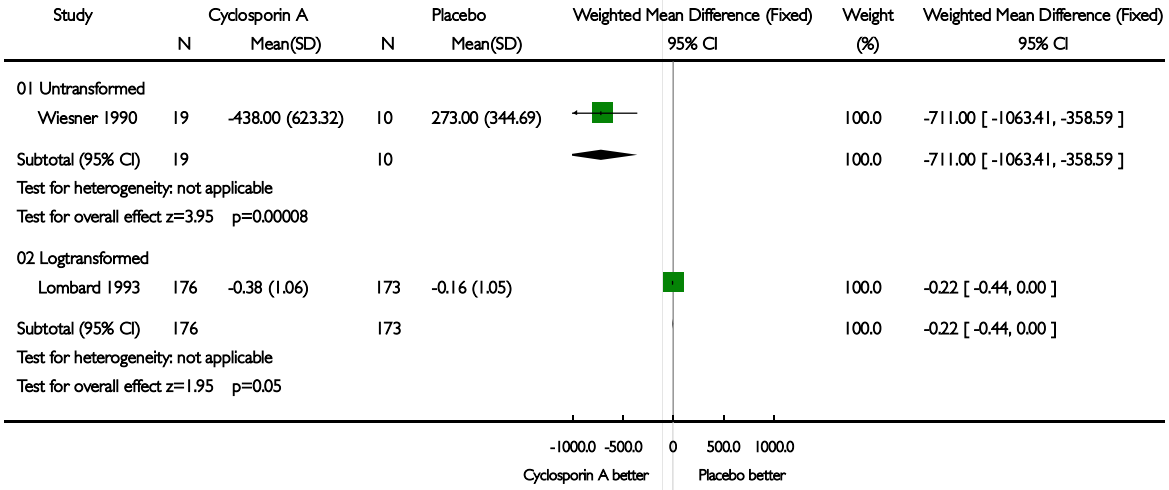


Analysis 01.07. Comparison 01 Cyclosporin A versus placebo, Outcome 07 Alkaline phosphatases (U/L) (change from baseline)

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 07 Alkaline phosphatases (U/L) (change from baseline)

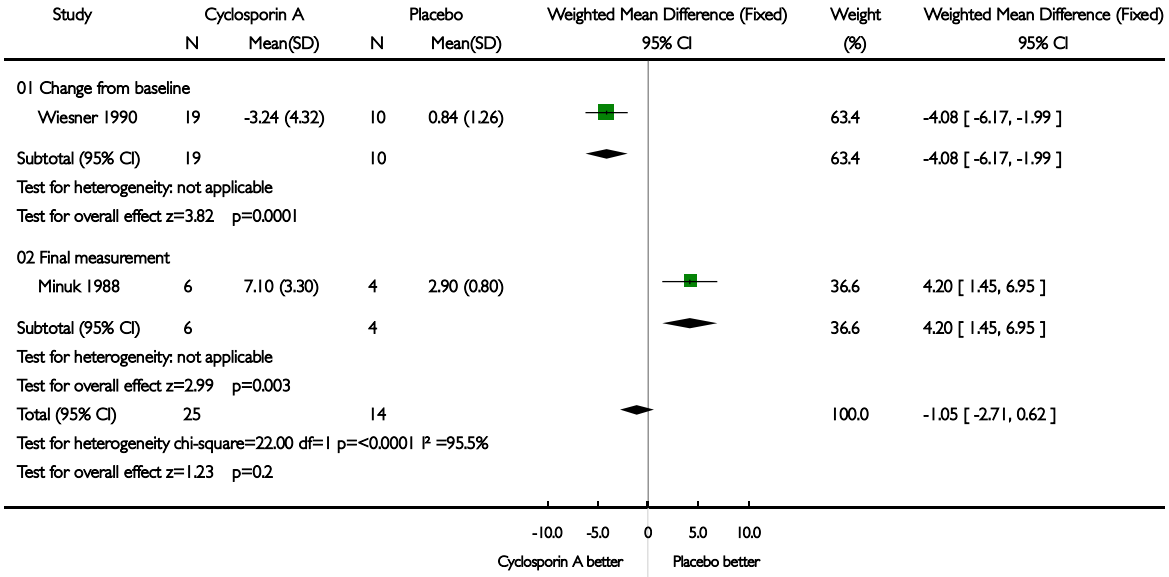


Analysis 01.08. Comparison 01 Cyclosporin A versus placebo, Outcome 08 Immunoglobulin M (g/L)

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 08 Immunoglobulin M (g/L)

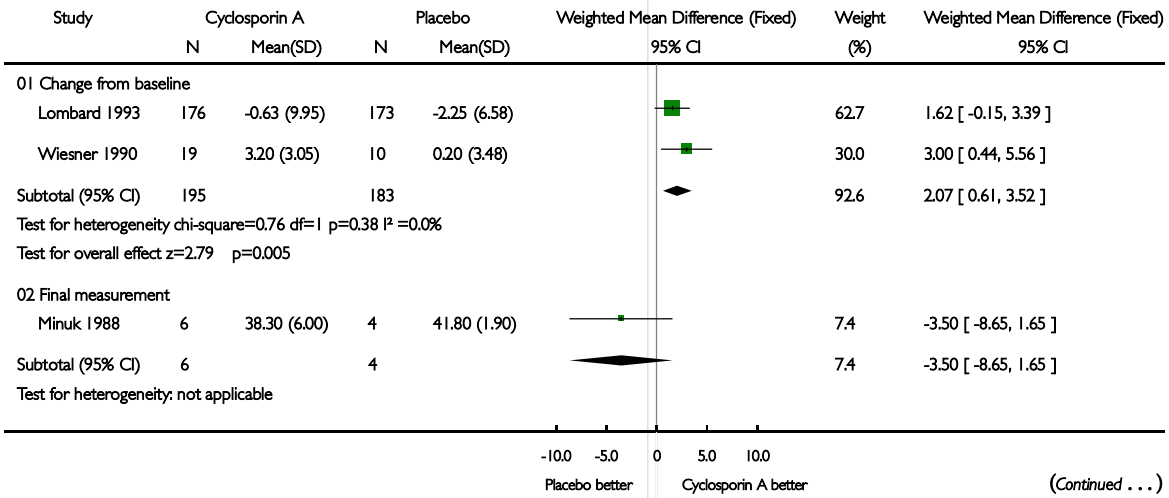


Analysis 01.09. Comparison 01 Cyclosporin A versus placebo, Outcome 09 Serum albumin (g/L)

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 09 Serum albumin (g/L)



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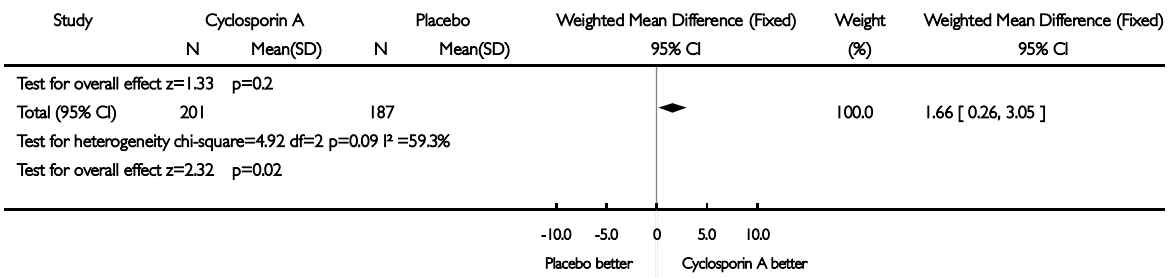
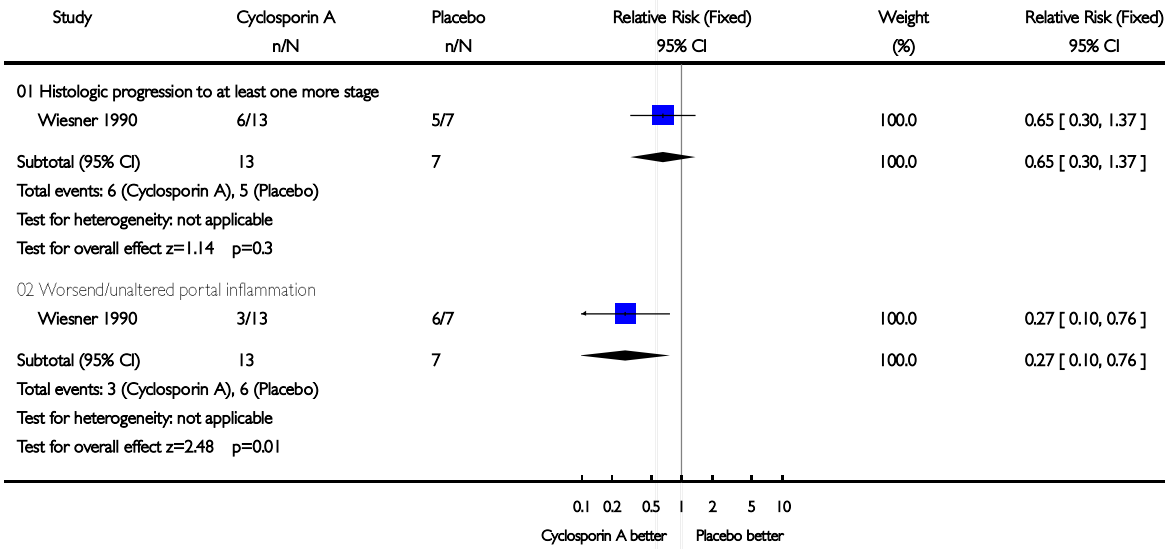


Analysis 01.10. Comparison 01 Cyclosporin A versus placebo, Outcome 10 Histologic assessment

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 10 Histologic assessment

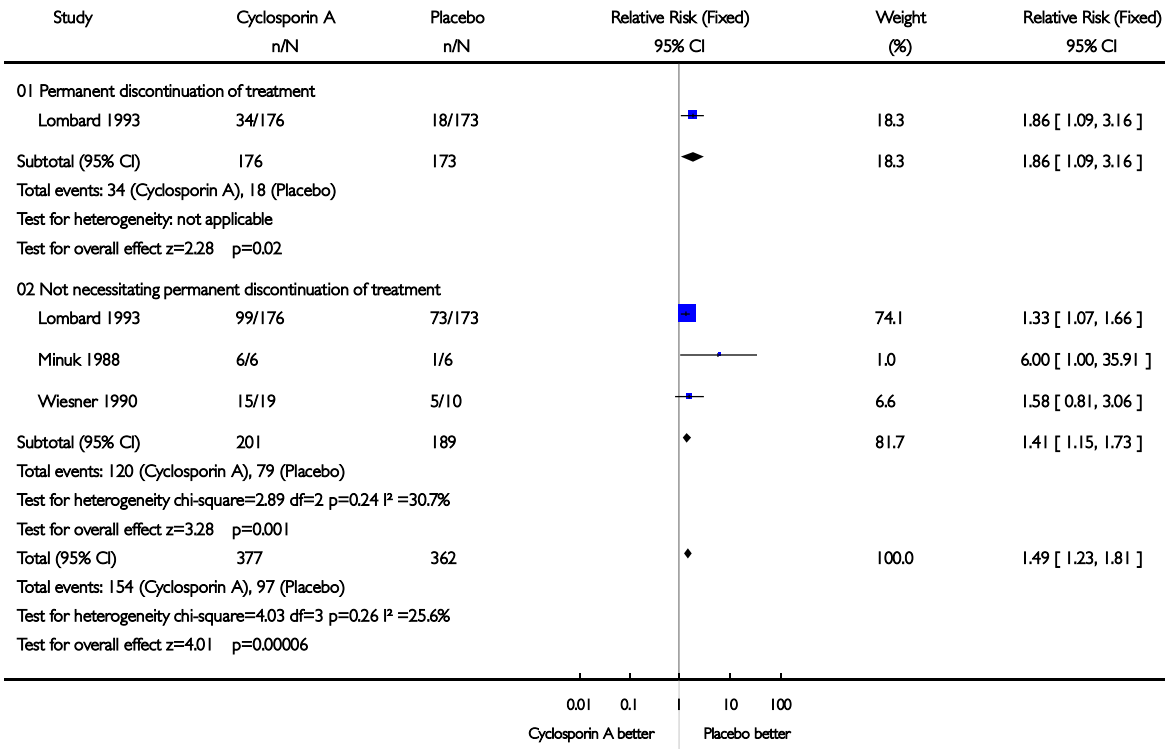


Analysis 01.11. Comparison 01 Cyclosporin A versus placebo, Outcome 11 Adverse event

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 11 Adverse event

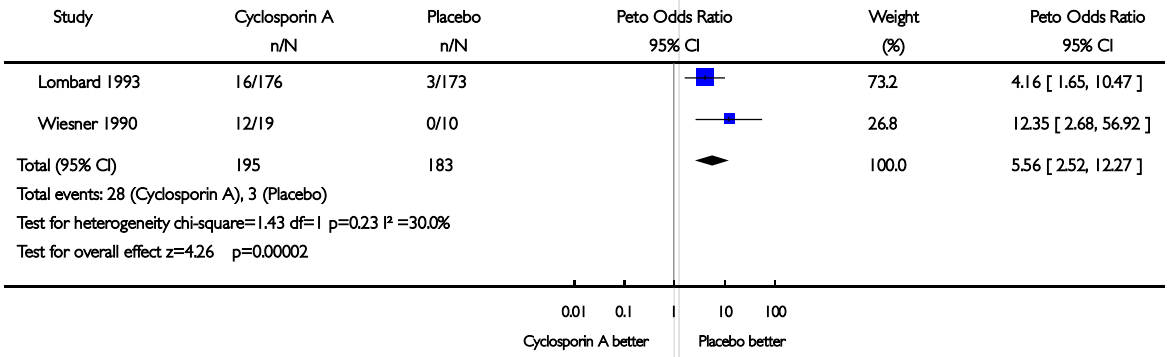


Analysis 01.12. Comparison 01 Cyclosporin A versus placebo, Outcome 12 Renal dysfunction

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 12 Renal dysfunction



Analysis 01.13. Comparison 01 Cyclosporin A versus placebo, Outcome 13 Increased blood pressure

Review: Cyclosporin A for primary biliary cirrhosis

Comparison: 01 Cyclosporin A versus placebo

Outcome: 13 Increased blood pressure