Interferon and lamivudine combination therapy versus lamivudine monotherapy for hepatitis B e antigen-negative hepatitis B treatment: a meta-analysis of randomized controlled trials

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BACKGROUND: **It has been demonstrated that only a minority of patients with hepatitis B e antigen (HBeAg)-negative chronic hepatitis B (CHB) obtain a sustained response after either interferon (IFN) or nucleos(t)ide analogue monotherapy. Therefore, combination therapy of drugs with synergistic antiviral effects was proposed to have a sustained response in these patients. We compared the effect and safety of lamivudine monotherapy and its combination with IFN including conventional interferon (CON-IFN) and pegylated interferon (PEG-IFN) for HBeAg-negative CHB patients.**

DATA SOURCES: **A group of three independent reviewers identified 9 eligible randomized controlled trials through electronic searches (MEDLINE, OVID, EMBASE, the Cochrane Library Clinical Trials Registry, and the Chinese Medical Database), manual searches, and contact with experts. Sustained virological and biochemical responses were defined as primary efficacy measures. We performed quantitative meta-analyses to assess differences between CON-IFN plus lamivudine combination and lamivudine monotherapy groups.**

RESULTS: **No greater sustained virological and biochemical rates were found in patients receiving CON-IFN/lamivudine combination therapy [29.1% vs. 26.7%, odds ratio (OR)=0.98, 95% confidence interval (CI) 0.65-1.50, *P*=0.94, and 41.8% vs. 40.3%, OR=1.13, 95% CI 0.78-1.65, *P*=0.51, respectively], though a reduced YMDD mutation rate was achieved in the combination group [8.39% vs. 30.0%, OR=0.16, 95% CI 0.076-0.33, *P*<0.001]. However, data from one PEG-IFN trial showed greater sustained virological and biochemical rates in patients receiving combination therapy [response rate 19.5% vs. 6.6%, OR=3.42, 95% CI 1.71-6.84, *P*<0.001 and 60.0% vs. 44.2%, OR=1.88, 95% CI 1.23-2.85, *P*=0.003, respectively].**

CONCLUSIONS: **Addition of CON-IFN to lamivudine did not improve treatment efficacy but suppressed YMDD mutation by lamivudine. Combination of PEG-IFN and lamivudine might increase the sustained response, and further clinical trials are needed for confirmation.**

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KEY WORDS: **lamivudine; interferon-alpha; combination therapy; monotherapy; HBeAg-negative; chronic hepatitis B**

Introduction

Hepatitis B is a major health burden with more than 400 million people chronically infected worldwide.[1, 2] Based on the status of hepatitis B e antigen (HBeAg), chronic hepatitis B (CHB) can be categorized into two clinically distinctive patterns: HBeAg-positive and HBeAg-negative.[3, 4] HBeAg-positive CHB is common among patients acquiring infection perinatally and is characterized by high levels of HBV DNA replication.[5, 6] Seroconversion of HBeAg, which is frequently accompanied by a durable response of viral suppression and clinical improvement,[7-9] marks a treatment end-point for HBeAg-positive CHB.[10-12] HBeAg-negative CHB, with frequent mutation in the precore or core promoter region of HBV, precluding the expression of HBeAg,[13, 14] is associated with

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progressive liver damage and a lower level of HBV replication than HBeAg-positive CHB.[15-17] Though the majority of patients with HBeAg-negative CHB initially respond well to both nucleos(t)ide analogues and interferon-alpha (IFN-α),[3, 18] there is no definite indication for discontinuation of antiviral therapy like the seroconversion in HBeAg-positive CHB[19] and most patients undergo a relapse after treatment cessation.[20-23] In recent years, the notion of combination therapy has been proposed and several randomized controlled trials have evaluated the therapeutic effect and safety of such therapies for HBeAg-negative CHB.[24-26] However, the results from different trials are controversial.[25-27] In the present study, we performed a systematic review and meta-analysis of eligible clinical trials to compare the effect of lamivudine monotherapy with lamivudine plus IFN combination therapy in HBeAg-negative patients.

Methods

Literature search and eligibility criteria

A group of three independent researchers conducted the literature search; trial selection and data extraction and disagreements were resolved by consensus. We identified eligible trials by searching the electronic databases MEDLINE, OVID, EMBASE, the Cochrane Library Clinical Trials Registry, and the Chinese Medical Database. Included terms were "chronic hepatitis B", "lamivudine", "interferon", "drug combination", "combination therapy", and "sequential therapy". Searching with both MeSH terms and free keywords was conducted. We also performed manual searches of the bibliographies of relevant articles and conference proceedings. We included randomized controlled trials comparing lamivudine monotherapy with IFN plus lamivudine combination therapy in adult HBeAg-negative CHB patients, irrespective of publication status or language. In addition, if multiple trials were derived from the same or partly overlapping study populations, only the largest or most recent eligible trial was included. The searches of the entire databases were conducted by September 2009.

Data extraction and efficacy measure definitions

For each trial, we gathered data on the following characteristics: location where trials were conducted, inclusion and exclusion criteria, regimen design (including type and dose of IFN, drug dose, and administration method), efficacy measures, duration of treatment and follow-up, losses to follow-up, and trial quality. Disagreements were resolved through discussion among reviewers. Incomplete data were supplemented by contact with primary investigators.

We used end-of-follow-up (sustained) virological and biochemical response rates as primary efficacy measures. End-of-treatment virological and biochemical response rates, histological response, incidence of YMDD (tyrosine, methionine, and aspartate) motif mutations, liver-related mortality, and treatment safety were used as secondary efficacy measures. Virological response was defined as suppression of HBV DNA below the lower detection limit as determined by polymerase chain reaction (PCR). Biochemical response was defined as alanine aminotransferase (ALT) normalization. Histological response was defined as at least a two-point reduction in the Knodell score[28] for pre- and post-treatment liver histopathology studies. HBsAg seroconversion was defined by the loss of HBsAg and the presence of anti-HBsAg antibody. Treatment safety was defined as the occurrence rate of adverse effects causing withdrawal from therapy.

Assessment of methodological quality and statistical analysis

We assessed trial quality using the Jadad quality scale.[29] Each study was evaluated by examining the allocation sequence generation, allocation concealment, blinding of outcome assessors, and reporting of patient withdrawal and dropout. Studies with scores more than 4 were defined as high-quality.

Quantitative meta-analysis was conducted using STATA version 10.0 (STATA Corp., College Station, Texas, USA). We pooled conventional interferon (CON-IFN) plus lamivudine combination therapy and lamivudine monotherapy as an overall effect and performed separate meta-analyses examining the defined efficacy measures. Subgroup analysis based on treatment duration (1 year and 2 years) or sensitivity analysis excluding trials with a treatment duration of 2 years was performed. The effect measures of differences between the two groups were odds ratios (OR) and the corresponding 95% confidence intervals (CI). A P value of less than 0.05 was considered to indicate a statistically significant difference. Heterogeneity was assessed for each analysis by means of Cochrane's Q test. A P value less than 0.10 indicated heterogeneity. The fixed effect model was used if no heterogeneity existed and the random effect model was used if heterogeneity was detected. The potential risk of publication bias was examined by the Egger test. Publication bias was indicated if the P value was less than 0.10. Intention to treat analysis was used in the study except for histological response rate analysis because the reporting rate was low.

Results

Patient selection and characteristics

We identified 3131 references and 1779 duplicates were deleted. Then after title, abstract and full-text screening, we finally included nine remaining trials involving 942 patients[24-27, 30-34] (Fig. 1). CON-IFN was used in eight trials (n=579)[24-27, 30-33] and one used pegylated (PEG) IFN-α (n=360).[34] Two trials (n=240) included only antiviral treatment-naive patients,[26, 30] three studies (n=162) exclusively studied IFN non-responders,[24, 25, 33] and the others (n=540) included both IFN treatment-naive and previously treated patients.[27, 31, 32, 34] Only one study used sequential therapy (n=162)[26] and the others (n=780)[24, 25, 27, 30-34] used simultaneous therapy. Three trials (n=535) comprised 48 weeks of treatment followed by 24 weeks of follow-up,[26, 33, 34] whereas patients in three trials (n=162) were treated for 96 weeks,[25, 27, 32] 3 had a longer follow-up (n=187)[24, 30, 31] and one (n=58) had no follow-up data (the trial was on-going when published).[27] Three trials (n=492) were of high methodological quality (Jadad scores ⩾3) [30, 32, 34] and the others (n=450) were not (Jadad scores <3);[24-27, 31, 33] however, none of the included studies were double-blinded. All studies were published in English as full publications (Tables 1 and 2).

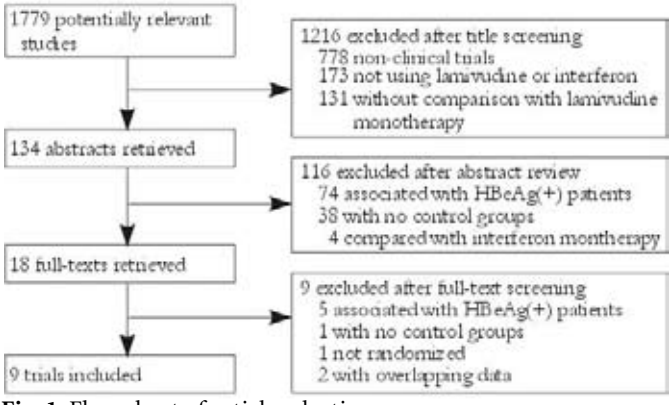

Fig. 1. Flow chart of article selection.

End-of-treatment virological response

Eight trials reported the end-of-treatment virological response rate.[24-27, 30-33] No significant difference in this rate was found between patients in combination and monotherapy groups [78.0% vs. 70.3%, OR=1.37, 95% CI 0.92-2.05, P=0.12]. The fixed effects model was used because no substantial heterogeneity existed (χ^2=8.15, df=7, P=0.32). Subgroup analysis showed no greater response in patients receiving either 1-year [77.9% vs. 70.2%, OR=1.33, 95% CI 0.82-2.16, P=0.25] or 2-year treatment [78.2% vs. 70.7%, OR=1.47, 95% CI 0.73-2.98, P=0.28]. No publication bias was detected (P=0.60, Egger test) (Fig. 2).

Table 1. Characteristics of included randomized controlled trials

Study	Location	Study design	Sample size (Com/Mono)	Regimen	Therapy period (mon)	Follow-up period (mon)	Jadad scores
Yurdaydin 2005	Turkey	RCT	39/39	Lamivudine 100 mg/d with or without INF-α-2a 9 MU 3/w	12	27 (include 6 months follow-up data)	3
Shi 2006	China	RCT	64/98	Lamivudine 100 mg/d ×20 w then lamivudine 100 mg/d plus INF-α-2b 5 MU 3/w ×4 w then INF-α-2b 5 MU 3/w ×24 w vs. lamivudine 100 mg/d ×48 w	12	6	1
Santantonio 2002	Italy	RCT	24/26	Lamivudine 100 mg/d with or without INF-α-2b 5 MU 3/w	12	6-13	2
Marcellin 2004	Asia and Europe	RCT	179/181	Lamivudine 100 mg/d with or without PEG-INF-α-2a 180 μg 1/w	12	6	3
Jaboli 2003	Italy	RCT	34/24	Lamivudine 100 mg/d ×4 w then lamivudine 100 mg/d plus INF-α-2b 5 MU 3/w ×12 mon then INF-α 6 MU 3/w ×16 mon vs. lamivudine 100 mg/d ×12 mon	12	6	2
Economou 2005	Greece	RCT	24/26	Lamivudine 100 mg/d with or without INF-α-2b 5 MU 3/w	24	6	3
Akyuz 2007	Turkey	RCT	21/24	Lamivudine 100 mg/d with or without INF-α-2b 10 MU 3/w	24	6	2
Scotto 2006	Italy	RCT	21/20/18	Lamivudine 100 mg/d with or without INF-α-2b 6 MU 3/w	Roughly 12	Roughly 12	2
Akarca 2004	Turkey	RCT	40/40	Lamivudine 150 mg/d with or without INF-α-2b 9-10 MU 3/w ×24 w	24	Missing	2

RCT: randomized controlled trial; IFN: interferon; PEG-IFN: pegylated interferon; Com: conventional interferon with lamivudine combination therapy; Mono: lamivudine monotherapy; w: week(s); mon: month(s); MU: million units.

Table 2. Selection criteria of included trials in the study

Study	Inclusion criteria	Exclusion criteria
Yurdaydin 2005	1. HBeAg(-), anti-HBe(+) and HBsAg(+) for at least 6 months 2. Presence of HBV DNA 3. Evidence of inflammation on biopsy within 1 year of study entry 4. 1.3 NL<ALT<10.0 NL	1. Previous treatment with nucleoside analogue 2. Co-infection of HIV, hepatitis C or D 3. Albumin below 3.5 g, bilirubin above 2 mg/dl, an increased prothrombin time of more than 3 seconds above the normal value 4. White blood cell or platelet counts of less than 3000 and 100 000 mm 5. Any significant disease which might have interfered with the conduct of the study
Shi 2006	1. Age>16 years 2. HBeAg(-), anti-HBe(+) and HBsAg(+) for at least 6 months 3. HBV DNA >100 000 copies/ml 4. 1.3 NL<ALT<10.0 NL	1. Co-infection of HIV, hepatitis C, D or E 2. Decompensated liver diseases or HCC 3. History of alcohol or drug abuse within 1 year before entry 4. Other possible causes of chronic liver damage 5. Previous treatment of CHB
Santantonio 2002	1. HBeAg(-), anti-HBe(+) and HBsAg(+) for at least 6 months 2. Evidence of inflammation on biopsy within 1 year of study entry 3. Presence of HBV DNA	1. Co-infection of HIV, hepatitis C or D 2. Decompensated liver cirrhosis or evidence of other liver disease
Marcellin 2004	1. HBeAg(-), anti-HBe(+) and HBsAg(+) for at least 6 months 2. HBV DNA >100 000 copies/ml 3. 1 NL<ALT<10.0 NL 4. Evidence of inflammation on biopsy within 1 year of study entry	1. Co-infection of HIV, hepatitis C or D 2. Decompensated liver disease 3. Coexisting serious medical or psychiatric illness 4. Neutrophil count <1500/mm^3 or Plt <90 000/mm^3 or Cr >1.5 NL 5. History of alcohol or drug abuse within one year before entry 6. Treatment for CHB within the previous 6 months
Jaboli 2003	1. Age between 18 and 70 years 2. HBeAg(-), anti-HBe(+) and HBsAg(+) for at least 6 months 3. HBV DNA >5 pg/ml 4. ALT>1 NL	1. Co-infection of HIV, hepatitis C or D 2. Decompensated liver disease or HCC 3. Other causes of chronic liver damage 4. Assumption of immunosuppressive or antiviral therapy within 6 months before study
Economou 2005	1. HBeAg(-), anti-HBe(+) and HBsAg(+) for at least 6 months 2. Serum HBV DNA >100 000 copies/ml 3. ALT>1.5 NL 4. Evidence of inflammation on biopsy within 1 year before study entry	1. Co-infection of HIV, hepatitis C or D 2. Decompensated liver disease (Child-Pugh score >8) or previous liver transplantation 3. Active alcohol consumption (>50 g/d) 4. Suspected HCC 5. Previous antiviral treatment other than IFN or immunosuppressive therapy within 6 months before study entry
Akyuz 2007	1. HBeAg(-), anti-HBe(+) and HBsAg(+) for at least 18 months 2. Serum HBV DNA >4 pg/ml 3. ALT>1.3 NL 4. Evidence of inflammation on biopsy	1. Co-infection of HIV, hepatitis C or D 2. Decompensated liver disease
Scotto 2006	1. HBeAg(-), anti-HBe(+) and HBsAg(+) for at least 6 months 2. Presence of HBV DNA 3. ALT>2 NL 4. Evidence of inflammation on biopsy	1. Co-infection of HIV, hepatitis C or D 2. Episodes of hepatic failure 3. Co-existing causes of liver diseases (e.g. autoimmune diseases, Wilson disease, alcoholism)
Akarca 2004	1. HBeAg(-), anti-HBe(+) and HBsAg(+) for at least 6 months 2. Presence of HBV DNA 3. ALT >1.5 NL 4. Evidence of inflammation on biopsy	1. Co-infection of HIV, hepatitis C or D 2. Decompensated liver disease 3. White blood cell or platelet counts of less than 4000 and 100 000/mm^3 or Cr >2.5 mg/dl 4. ANA >1/160 or AFP >20 ng/ml 5. History of hereditary diseases such as Wilson's disease, hematochromatosis, alpha-antitrypsin deficiency 6. Presence of uncontrolled diabetes and cardiac disease 7. History of psychiatric disease 8. Active alcohol consumption (>20 g/d) 9. Previous nucleoside analogue treatment or immunosuppressive therapy or interferon therapy within 1 month before study entry

NL: normal limit.

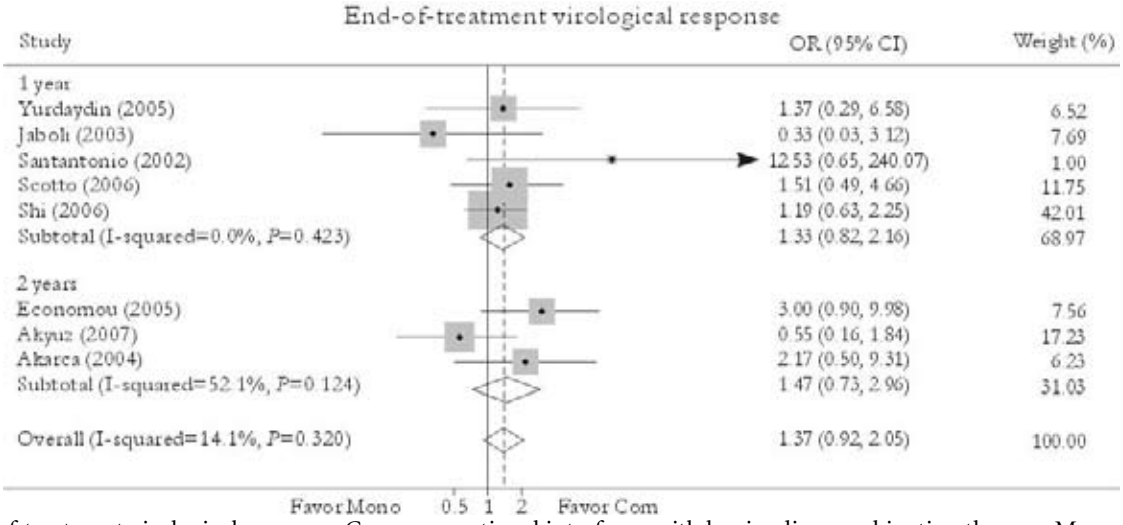

Fig. 2. End-of-treatment virological response. Com: conventional interferon with lamivudine combination therapy; Mono: lamivudine monotherapy.

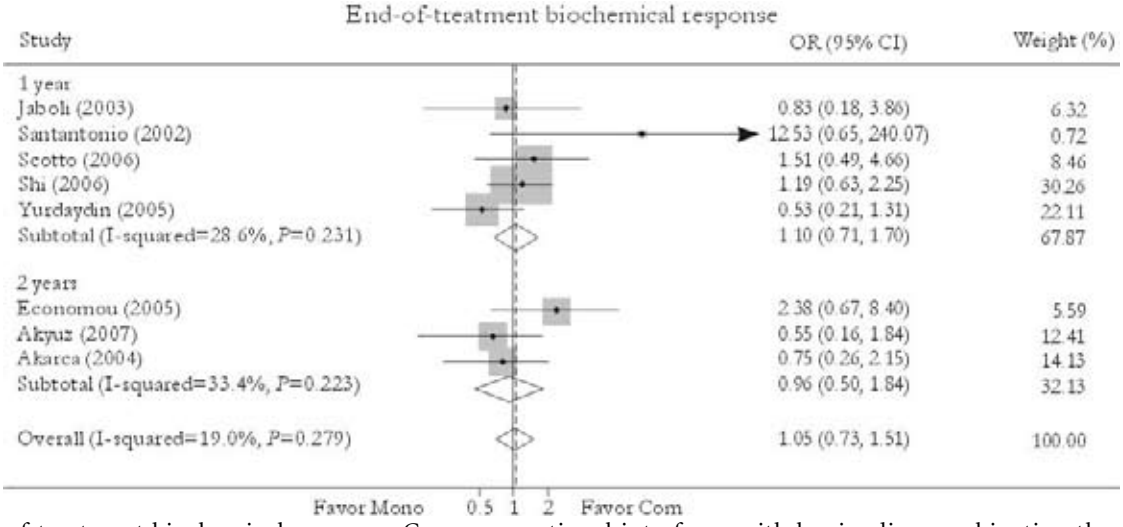

Fig. 3. End-of-treatment biochemical response. Com: conventional interferon with lamivudine combination therapy; Mono: lamivudine monotherapy.

End-of-treatment biochemical response

Eight trials reported the end-of-treatment biochemical response rate.[24-27, 30-33] No significant difference in this rate was found between combination and monotherapy groups [69.9% vs. 67.0%, OR=1.05, 95% CI 0.73-1.51, P=0.77]. The fixed effects model was used because of substantial heterogeneity (χ^2=8.65, df=7, P=0.28). Subgroup analysis showed no greater response in patients receiving either 1-year [69.3% vs. 65.0%, OR=1.10, 95% CI 0.71-1.70, P=0.66] or 2-year treatment [71.3% vs. 71.7%, OR=0.96, 95% CI 0.50-1.84, P=0.89]. No publication bias was detected (P=0.40, Egger test) (Fig. 3).

Sustained virological response

Seven trials reported the sustained virological response rate.[24-26, 30-33] No significant difference in this was found between combination and monotherapy groups [29.1% vs. 26.7%, OR=0.98, 95% CI 0.65-1.50, P=0.94]. No substantial heterogeneity was found (χ^2=3.07, df=6, P=0.80) and the fixed effects model was used. Subgroup analysis showed no greater response in patients receiving either 1-year [31.1% vs. 28.4%, OR=0.98, 95% CI 0.62-1.55, P=0.93] or 2-year treatment [20.0% vs. 20.0%, OR=1.01, 95% CI 0.37-2.75, P=0.99]. No publication bias was detected (P=0.92, Egger test) (Fig. 4).

Sustained biochemical response

Seven trials reported the sustained biochemical

response rate.[24-26, 30-33] Compared with patients in the monotherapy group, a greater rate was found in patients receiving combination therapy [41.8% vs. 40.3%, OR=1.13, 95% CI 0.78-1.65, *P*=0.51]. No statistically significant heterogeneity was found (χ^2=3.62, *df*=6, *P*=0.73) and the fixed effects model was used. Subgroup analysis showed no greater response in patients receiving either 1-year [46.2% vs. 44.2%, OR=1.18, 95% CI 0.79-1.78, *P*=0.42] or 2-year treatment [22.2% vs. 24.0%, OR=0.91, 95% CI 0.35-2.35, *P*=0.84]. No publication bias was detected (*P*=0.96, Egger test) (Fig. 5).

Incidence of YMDD mutation during treatment

Seven trials reported the incidence of YMDD mutation at the end of treatment;[24-26, 30-33] however, three were excluded because only patients who did not respond to treatment were tested for YMDD variants.[24, 25, 31] Compared with patients in the monotherapy group, a lower YMDD mutation emergence rate was found in patients receiving combination treatment [8.39% vs. 30.0%, OR=0.16, 95% CI 0.076-0.33, *P*<0.001]. No statistically significant heterogeneity was found (χ^2=1.74, *df*=3, *P*=0.63) and the fixed effects model was used. Sensitivity excluding trials with a 2-year treatment duration did not change the trend [7.63% vs. 27.3%, OR=0.17, 95% CI 0.077-0.39, *P*<0.001]. Substantial publication bias was detected (*P*=0.23, Egger test) (Fig. 6).

Histological response

Three studies reported the histological response

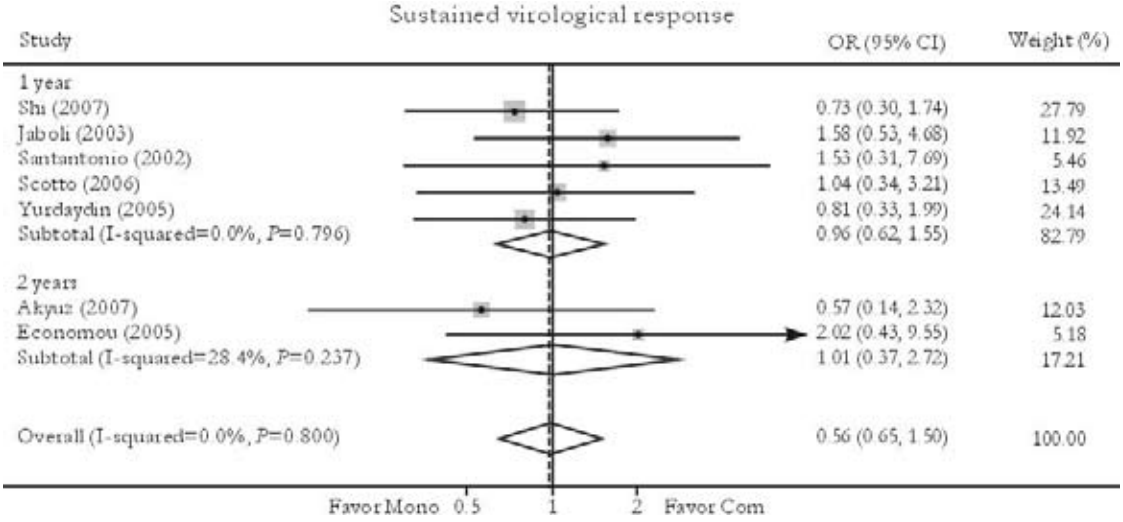

Fig. 4. Sustained virological response. Com: conventional interferon with lamivudine combination therapy; Mono: lamivudine monotherapy.

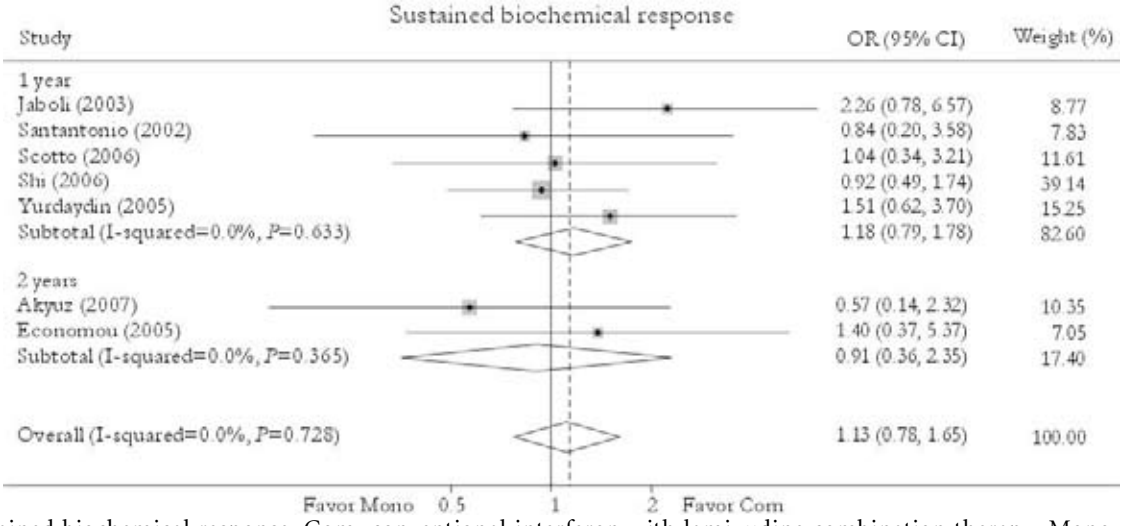

Fig. 5. Sustained biochemical response. Com: conventional interferon with lamivudine combination therapy; Mono: lamivudine monotherapy.

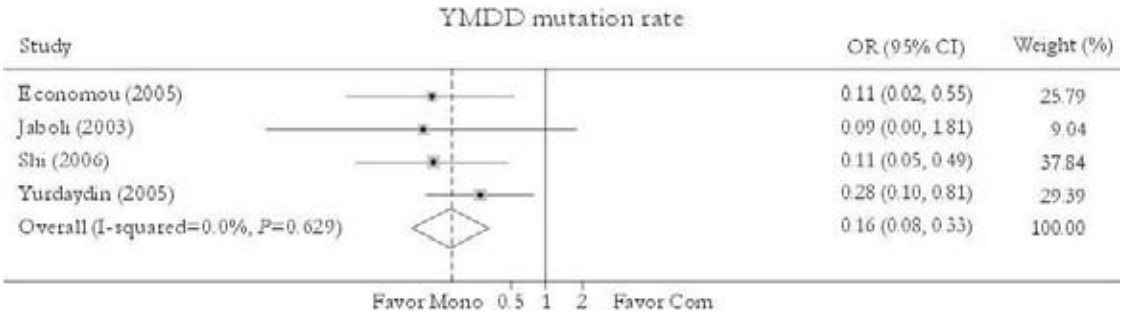

Fig. 6. YMDD mutation rate. Com: conventional interferon with lamivudine combination therapy; Mono: lamivudine monotherapy.

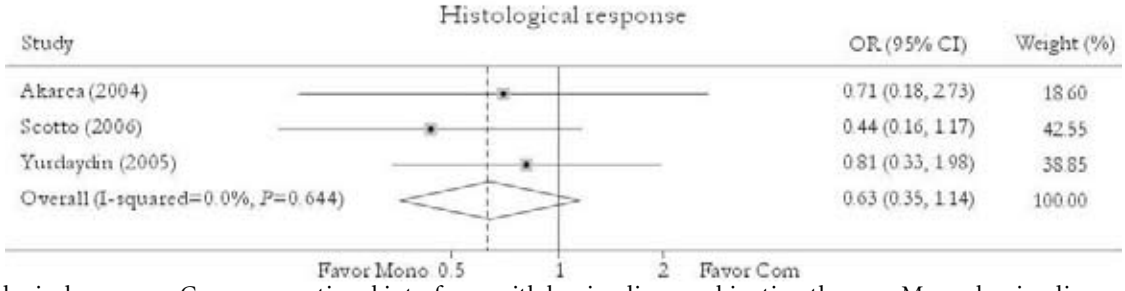

Fig. 7. Histological response. Com: conventional interferon with lamivudine combination therapy; Mono: lamivudine monotherapy.

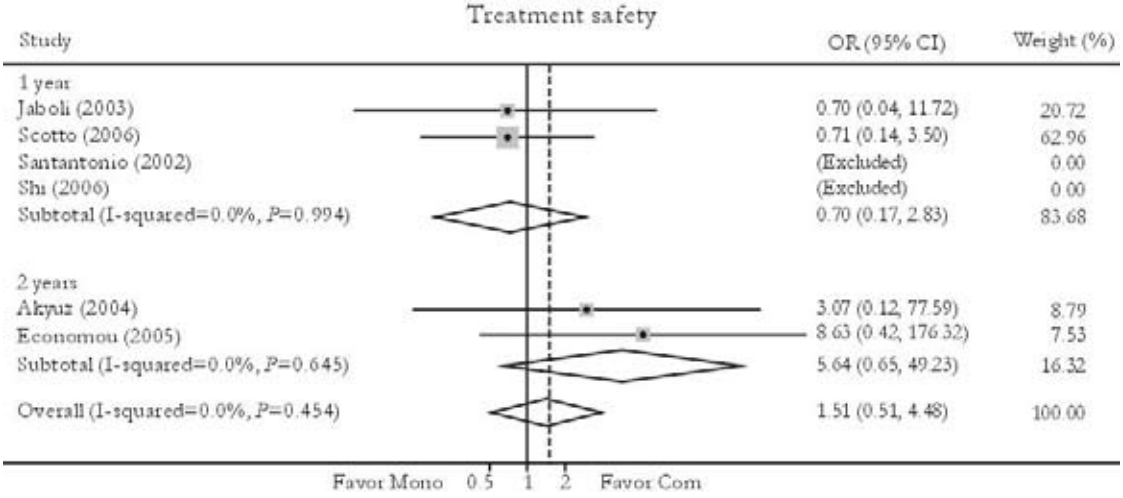

Fig. 8. Treatment safety. Com: conventional interferon with lamivudine combination therapy; Mono: lamivudine monotherapy.

rate.[24, 27, 30] No significant difference in this rate was found between patients in combination and monotherapy group [47.7% vs. 56.0%, OR=0.63, 95% CI 0.35-1.14, P=0.13]. No substantial heterogeneity was found (χ^2=0.88, df=2, P=0.64) and the fixed effects model was used. Sensitivity excluding trials with 2-year treatment duration did not change the trend [36.4% vs. 48.6%, OR=0.61, 95% CI 0.32-1.19, P=0.15]. No publication bias was detected (P=0.95, Egger test) (Fig. 7).

HBsAg loss or seroconversion

Four trials reported the HBsAg loss or seroconversion rate[24, 26, 27] and no such cases were found.

Treatment safety

Six trials reported the treatment safety rate.[24, 26, 27, 31-33] No significant difference in this rate was found between patients in the combination and monotherapy groups [3.98% vs. 1.69%, OR=1.51, 95% CI 0.51-4.48, P=0.46]. No substantial heterogeneity was found (χ^2=2.62, df=3, P=0.45) and the fixed effects model was used. Subgroup analysis also showed no statistically significant difference in the two subgroups [3.12% vs. 2.37%, OR=0.70, 95% CI 0.17-2.83, P=0.62 (1 year); 6.06% vs. 0.00%, OR=5.64, 95% CI 0.64-49.23, P=0.12 (2 years)]. No publication bias was detected (P=0.28, Egger test) (Fig. 8).

Table 3. Comparison of CON-IFN and PEG-IFN combination therapy (1-year results)

Effect measures	CON-IFN group			PEG-IFN group		
	No. of Com/ Mono group	Rate of Com/ Mono group (%)	OR (95% CI) and *P* value	No. of Com/ Mono group	Rate of Com/ Mono group (%)	OR (95% CI) and *P* value
EVR	120/97	85.0/79.4	1.72 (0.82-3.58) *P*=0.15	179/181	87.1/73.5	2.45 (1.41-4.23) *P*=0.001
EBR	199/208	69.3/65.0	1.10 (0.71-1.70) *P*=0.66	179/181	48.6/72.9	0.35 (0.23-0.54) *P*<0.001
SVR	96/71	33.3/25.3	1.22 (0.68-2.16) *P*=0.42	179/181	19.5/6.6	3.42 (1.71-6.84) *P*<0.001
SBR	289/308	46.2/44.2	1.18 (0.79-1.78) *P*=0.42	179/181	60.0/44.2	1.88 (1.23-2.85) *P*=0.003
YMDD mutation	131/154	7.63/27.3	0.17 (0.077-0.39) *P*<0.001	179/181	0.55/17.7	0.027 (0.004-0.20) *P*<0.001
HR	77/70	36.4/48.6	0.61 (0.32-1.19) *P*=0.15	179/181	38.0/40.0	0.93 (0.61-1.42) *P*=0.73
HBsAg loss or seroconversion	102/119	0/0	-	179/181	2.8/0	11.44 (0.63-208.46) *P*=0.10
Treatment safety	160/169	3.12/2.37	0.70 (0.17-2.83) *P*=0.62	179/181	4.0/0	15.78 (0.89-278.44) *P*=0.06

CON-IFN: conventional interferon, PEG-IFN: pegylated interferon, Com: combination therapy, Mono: monotherapy, EVR: end-of-treatment virological response, EBR: end-of-treatment biochemical response, SVR: sustained virological response, SBR: sustained biochemical response, HR: histological response.

Liver-related mortality

No liver-related death was reported in the included trials.

CON-IFN combination therapy versus PEG-IFN combination therapy

Only one trial used PEG-IFN, and the treatment duration was one year. We listed the 1-year pooled results of CON-IFN combination therapy with those of PEG-IFN combination therapy (Table 3). Our findings showed significantly higher end-of-treatment virological response, sustained virological response, and sustained biochemical response in patients receiving combination therapy than in those who received monotherapy in the PEG-IFN group, which was different from the results of the CON-IFN group. The differences in histological response, HBsAg loss or seroconversion rate, and treatment safety remained of no statistical significance in the PEG-IFN group. And the reduced YMDD mutation was also consistent with the results of the CON-IFN group.

Discussion

HBeAg-negative CHB is associated with higher risk of cirrhosis, hepatocyte failure, and HCC than the HBeAg-positive pattern, which prompted long-term viral suppression treatment.[16, 35, 36] However, patients receiving monotherapy with either nucleos(t)ide analogues or IFN-α frequently failed to achieve sustained remission.[37, 38] Therefore, the notion of combination therapy was proposed, aiming to decrease mutagenicity and obtain a synergistic effect.[39-41] This review, comprised 10 randomized controlled trials, compared the effect and safety of lamivudine plus IFN-α combination therapy with lamivudine monotherapy for patients with HBeAg-negative CHB. Our findings demonstrated that though addition of CON-IFN reduced the YMDD mutation emergence rate, it improved neither end-of-treatment nor sustained response rates, and this conclusion was supported by both the 1-year and 2-year results. There were also no statistically significant differences in histological response rate, HBsAg loss or seroconversion rate, and the occurrence of severe adverse events between patients receiving the two therapies.

Based on the data from one multicenter and randomized trial, our analysis suggested that addition of PEG-IFN is superior to lamivudine monotherapy in maintaining the sustained response. However, it was noted that the sustained virologic response in this trial was significantly lower than in the CON-IFN trials. This discrepancy in response rate may be due to the stricter definition of virological response applied in the PEG-IFN trial, defined as suppression of HBV DNA to below 400 copies/ml, while most of the CON-IFN trials used a threshold level of 5 pg/ml. Nonetheless, further randomized controlled trials with large simple sizes are needed to draw a definite conclusion.

We found a YMDD-prevention effect of both CON- and PEG-IFN combination therapies. First, the two drugs inhibit different targets in the HBV DNA replication pathway and therefore provide synergistic antiviral activity.[31, 42, 43] Lamivudine acts primarily as a DNA polymerase inhibitor,[44] whereas IFN suppresses HBV DNA by inducing posttranscriptional degradation of HBV RNA and the expression of antiviral proteins.[45-47] Furthermore, several studies demonstrated that lamivudine treatment reconstitutes

the cytotoxic T lymphocyte-mediated immune response against HBV[48-50] while the immunomodulatory effect of IFN has long been confirmed.[51-53] Second, due to this synergistic antiviral effect, patients with combination therapy tend to achieve a sharper reduction of HBV DNA level than those with monotherapy. Accumulating evidence indicates an inverse correlation between the rapidity and profundity of HBV DNA suppression and the emergence of resistance.[54-57] This may explain the low occurrence rate of YMDD motif mutation in patients receiving IFN-α and lamivudine combination therapy. Considering that the duration of interferon therapy was predefined; however, the cessation of interferon treatment might negate this YMDD-suppressing effect.

We also found that PEG-IFN/lamivudine but not CON-IFN/lamivudine achieved a sustained virological and biochemical response. Peg-IFN-α, produced by covalently attaching a 40-kDa branched-chain polyethylene glycol moiety to IFN-α,[58] has pharmacokinetics superior to conventional IFN-α.[59, 60] Therefore PEG-IFN may elicit a more pronounced immune response in the host against HBV replication and elimination of reservoirs of infected cells compared with conventional IFN. A randomized controlled trial comparing the effect of PEG-IFN and CON-IFN monotherapy in HBeAg-positive patients showed that PEG-IFN-treated patients achieved a greater magnitude of HBV DNA reduction.[61] In addition, three trials in the CON-IFN subgroup exclusively studied IFN non-responders, whereas the only trial included in the PEG-IFN subgroup selected a mixed population of treatment-naive and previously treated patients. This may explain why the PEG-IFN/lamivudine combination is superior to CON-IFN/lamivudine in obtaining a sustained response.

This review is limited in several aspects. First, the heterogeneity of studied population characteristics, quality score, regimen design, and follow-up protocols among the included studies may have led to certain biases in our meta-analysis. However, these concerns may be alleviated by the lack of substantial heterogeneity and publication bias and the low loss rate of subjects included in the trials. Second, 1-year antiviral monotherapy with lamivudine in several studies, which is not consistent with current guidelines, would contribute to the high relapse rate after treatment discontinuation. However, this concern was alleviated by the confirmation of 2-year results. Third, only one trial using PEG-IFN was included and the patients in the monotherapy arm received only 1-year treatment with lamivudine, which would reduce the validity of the evaluation of PEG-IFN. In addition, there was methodological heterogeneity between the PEG-IFN and CON-IFN groups. In particular, the trial evaluating PEG-IFN was of high quality, whereas the CON-IFN subgroup included several studies with small sample sizes and low quality. And we used the 1-year treatment results when we compared the efficacy between the CON-IFN and PEG-IFN groups, which would undermine the conclusion.

In conclusion, combination of CON-IFN and lamivudine added no benefit but reduced the YMDD mutation rate. PEG-IFN combined with lamivudine, however, might improve sustained therapeutic efficacy, which needs further clinical trials with long-term therapy to be confirmed.

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