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Evaluation of Peginterferon alfa-2a Response Rate in HBe-Negative Chronic Hepatitis B: A Systematic Review and Meta-Analysis

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ABSTRACT

Selecting the best medications for ALT and HBV-DNA control in HBeAg negative chronic hepatitis patients is necessary as they are vulnerable to developing cirrhosis and hepatocellular carcinoma. The present study was aimed to evaluate the response rate of treatment by Peginterferon alfa-2a. In this systematic review and meta-analysis, a review of articles evaluating response rate to Peginterferon alfa-2a was done through the databases of Scopus, Google Scholar, Pubmed (inception - 2019). Fourteen studies reporting the response rate of HBeAg negative chronic patients' response to Peginterferon alfa-2a were included in the study. Cochran test was used to determine the homogeneity of the samples (based on Q test and I^2 index). The prevalence was estimated based on the Random Effect size model in Revman software (version 3.5). The main result of this study showed that, in the 14 studies that PEG interferon was prescribed for patients with Hbe Ag negative chronic hepatitis B, the pooled incidence of treatment responders in comparison to non-responders was significantly lower (OR = 0.78, CI (0.7-0.87), p <0.0001). High heterogeneity was observed in this analysis (I^2 = 95%, p <0.0001). to investigate factors affecting heterogeneity, a subgroup analysis was done based on the HBV genotype. 5 Studies in which most patients (>90%) had C genotype were compared with 3 studies with D genotype dominant study population. Again a high heterogeneity was observed in both genotype C and D dominant (I^2=92% and 95%, respectively). However, higher pooled responders to non-responders rate were seen in C dominant populations versus D genotype (OR=0.72, CI (0.69-0.75), p<0.0001). Using Peginterferon alfa-2a alone is not the best choice of treatment due to the low rate of response to treatment; while considering the patient's HBV genotype is important in response rate.

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Hbe Ag negative chronic hepatitis B, HBV, DNA, ALT, Peginterferon alfa-2a.

INTRODUCTION

Hepatitis B is one of viral hepatitis that has more than 300 million carriers worldwide and about one million people die each year due to its complications. Hepatitis B virus is one of the causes of acute and chronic hepatitis, liver cirrhosis and hepatocellular carcinoma (Asrani *et al*., 2019). Clinically important viral proteins concerning hepatitis B disease course include HBsAg, HBcAg, HBeAg, which HBsAg indicates infection with hepatitis B and its anti-HBs antibodies indicate improvement and immunity against the disease (Cherre *et al*., 2019). Clinical signs and symptoms of this infection vary, resulting in acute, chronic, no-clinical symptoms, and even fulminant infections. Acute infection is somewhat self-limited and is associated with inflammation and necrosis of the liver cells. The mortality rate in acute infection is about 0.5 to 1% (Malani, 2010). HbsAg in the blood or serum for more than six months indicates chronic infection. Upon HBV entry into the body, the disease gets chronic in 95% of children and 3 to 5% of adults (Nicolini *et al*., 2019).

Initial evaluation of a person with chronic HBV infection requires measurement of HBeAg and HBeAb levels and serum HBV DNA levels (Wilkins *et al*., 2019). Newborns born of mothers with chronic hepatitis often have HBe Ag virus, HBV positive, normal enzymes, high virus titers and mild disease in the biopsy. This is called the immune tolerant phase (Lee, 1997). This stage may last for the second and third decades of life. After this stage, liver enzymes are elevated in some patients, while still positive for HBe Ag and most liver biopsies show significant and severe liver disease. This phase is called HBe Ag positive chronic hepatitis (Mahoney, 1999). Prolonged liver fibrosis or cirrhosis may develop if this stage gets prolonged. After this stage, spontaneously or with treatment, HBeAb seroconversion occurs as HBeAg becomes negative and HBeAb becomes positive. In the majority of patients, after a course of hepatic failure, hepatic enzymes become normal and the severity of liver disease decreases. These patients are called inactive HBV carriers (Fujisawa *et al*., 2000). These patients have a good prognosis and may live for decades and die due to reasons other than liver disease. However, some of these patients, with HBeAg negative, have high viral titers and elevated liver enzymes and

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show significant liver disease in liver biopsy. This group of patients is called HBeAg negative chronic hepatitis individuals. Such patients may experience cirrhosis and hepatocellular carcinoma. Most HBV-negative cirrhosis patients have HBe Ag (Chu *et al*., 2002). Chronic hepatitis B is difficult to be treated as many patients require long-term treatment with expensive medications and the possibility of drug resistance, so treatment should only be initiated in patients who really need treatment and the best medical regimen should be prescribed for good treatment response (Raimondo and Pollicino, 2016).

In patients with HBeAg-positive chronic hepatitis, the goal of treatment is to achieve HBeAg-negative status. In the treatment of patients with HBeAg-negative chronic hepatitis, the goal is to normalize ALT and suppress the virus continuously, so that HBV DNA should get negative or negative less than 2000 IU/ml (Sundaram and Kowdley, 2015; Mohanty *et al*., 2006).

To date, seven drugs have been approved for the treatment of chronic hepatitis, including interferon-alpha, PEG interferon, lamivudine, adenovir adipovir dipivocal, entektavir, telbivodine teno firavir interferon (Yuen *et al*., 2011).

PEGylated recombinant human interferon-alpha 2A is an inducer of an innate antiviral immune response. One of the benefits of using PEG Interferon is the specific duration of treatment with this drug and the lack of drug resistance. The purpose of this research was to assess Peginterferon alfa-2a's response rate to therapy (Fung *et al*., 2016).

METHODS

This study is a meta-analysis of the ratio of response to treatment with PEG interferon-alpha in patients with HBeAg-negative chronic hepatitis in comparison of non-responding patients, in a variety of studies, using literature review and meta-analysis of existing sources between 1900 and 2019. Articles published in the Pub Med, ISI Web of Science, SCOPUS and Google scholar databases have been used to find relevant studies. The search strategy was mainly done using the systematic search with all possible combinations of the following keywords: Hbe Ag negative chronic hepatitis B, HBV, DNA, ALT, Peginterferon alfa-2a.

Criteria for selection and evaluation of articles quality

Initially, a list of the titles and abstracts of all the articles in the databases were provided by the researcher and independently reviewed to determine and select the relevant titles. Then the related articles were independently entered into the study process. The main criterion for the various articles in this study is: (i) studies reporting responder and non-responder patients in Hbe Ag negative chronic hepatitis B patients receiving PEG interferon, and (ii) unrelated studies to the treatment of HBeAg-negative chronic hepatitis with PEG interferon alpha have been excluded.

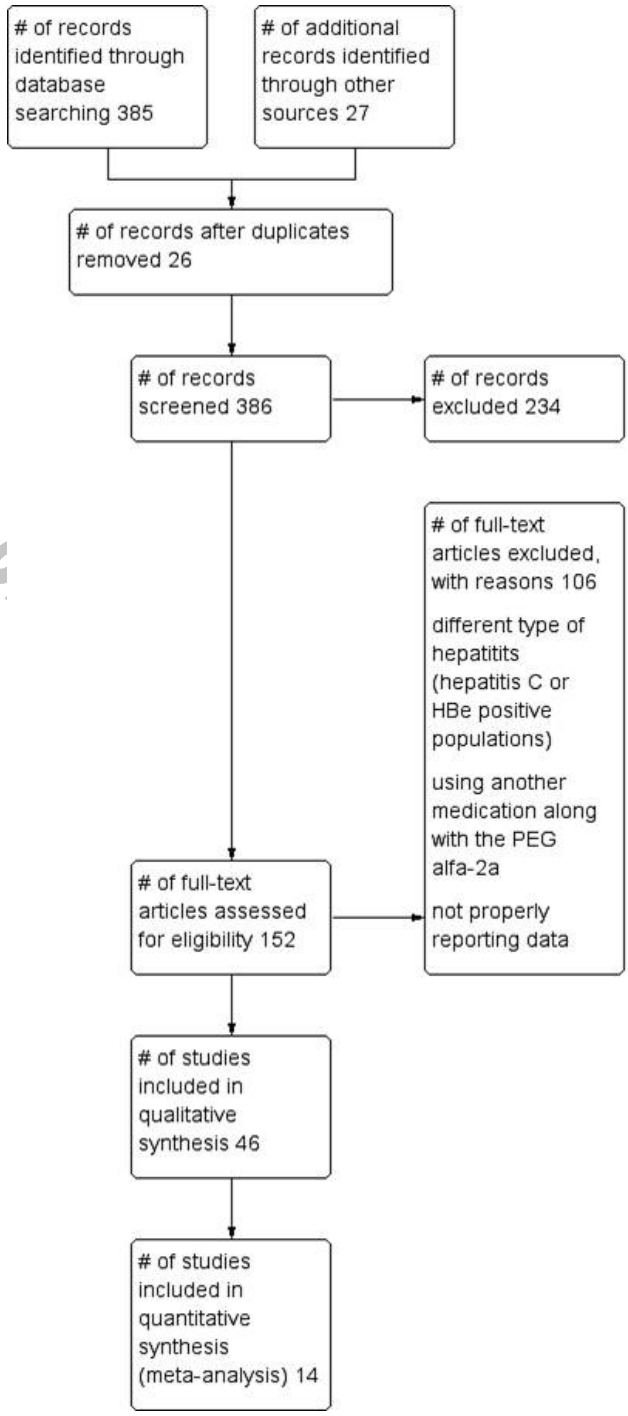

Fig. 1. Study flow.

Secondly, after identifying relevant reviews in terms of titles, the abstract of the various papers selected by the researcher using the STROBE (The Strengthening the Reporting of Observational Studies in Epidemiology) checklist which is an internationally renowned standard and quality statement for studies' quality assessment. The checklist consists of 43 different sections and includes various aspects of the methodology including sampling methods, variables measurement, and statistical analysis, adjusting for confounders, mentioning the validity and reliability of the tools used and the objectives of the study. A minimum score of 40 was deemed necessary in this checklist for inclusion of study. There was no need to assess Publication Bias and Funnel Plot, depending on the use of the quality control checklist; however study bias was shown in Figure 1. Based on the explanations given in the first step, 386 articles were selected. After reviewing the titles, 152 articles were identified to be related to the topic and entered the next phase, and 98 articles were removed due to being not relevant to the research topic. Related articles were identified and entered the second phase of the qualitative evaluation of abstracts. At the end of this phase, 32 articles that did not report our desired factors were excluded from the study. Finally, 14 studies were included in statistical analysis and their information is presented in Table I. The flow diagram of study selection based on PRISMA is shown in Figure 1.

Statistical analysis

The ratio of the number of responders to nonresponders in each study was determined first. The ratio standard error the true proportion (SE) was calculated using the following formula:

$$SE = \sqrt{[P \times (1-P)]/_N}$$

Where, P is proportion and N is the number of all subjects.

The weight of each study was calculated based on the Fix Effect Model (Hackshaw, 2009). Then, taking into account the weight of each study, the relative amounts obtained were combined to determine heterogeneity (Random Effect size model) and the overall proportion was calculated. Finally, the heterogeneity index with a heterogeneity test between studies was determined using Cochran Q tests. After confirming that studies are heterogeneous, the best estimate was calculated based on the Random Effect model. The subtotal analysis was performed based on the type of HBV genotype. All analyses were conducted using Review Manager Version 5.3 (Nordic Cochrane Centre, Copenhagen, Denmark). The studies bias is shown in Figure 2.

Table I.- Characteristics of the studies included in the meta-analysis study.

Reference	Design	Country	Sample size	Mean age	Most frequently observed HBV genotype		Male gender (%)	
					Responders	Non-responder	Responders	Non-responder
Bonino *et al*. (2007)	Prospective	China	518	39.9	C	83	82	D
Brook *et al*. (1989)	RCT	UK	114	35.8	NA	88.7	100	NA
Buster *et al*. (2009)	Retrospective	Netherlands	721	33.6	C	79.3	70.3	C
Chuaypen *et al*. (2018)	RCT	Thailand	121	40.4	C	68.6	74.3	C
de Niet *et al*. (2016)	Prospective	Amsterdam	28	42	D	86	57	A
Manesis and Hadziyannis (2001)	Retrospective	Greece	200	48.4	NA	91	82	NA
Goulis *et al*. (2015)	Prospective	Greece	95	42	D	70	82	D
Lampertico *et al*. (2003)	Cohort	Italy	101	46	D	93	77	D
Moucari *et al*. (2009)	Not clear	France	48	44	A	83	83	D
Rijckborst *et al*. (2010)	RCT	Poland	107	42	D	73.5	66.7	D
Tamai *et al*. (2017)	Cohort	Japan	23	47	C	54.5	83	C
Tangkijvanich *et al*. (2010)	Retrospective	Thailand	30	37	C	80	70	C
Tatsukawa *et al*. (2018)	RCT	Japan	21	53	C	69	100	C
Zhang *et al*. (2016)	Prospective	China	46	34.7	NA	70.6	66.7	NA

RESULTS

In this meta-analysis study, 14 articles with a sample size of 1655 were included. The highest sample size (721 cases) was reported in the study of Buster *et al*. (2009) and the lowest (23 cases) in the study of Tamai *et al*. (2017). The general characteristics and information of the selected studies are presented in Table I.

Based on Figure 3, in the 14 studies that PEG interferon was prescribed for patients with Hbe Ag negative chronic hepatitis B, the pooled incidence of treatment responders in comparison to non-responders was significantly lower (OR=0.78, CI (0.7-0.87), p<0.0001). High heterogeneity was observed in this analysis (I2=95%, p<0.0001).

A subgroup analysis was done based on the HBV genotype. 5 studies in which most patients (>90%) had C genotype were compared with 3 studies with D genotype dominant study population. Again a high heterogeneity was observed in both genotype C and D dominant (I2=92% and 95%, respectively). However, higher pooled responders to non-responders rate was seen in C dominant populations versus D genotype (45.2% vs. 54.8%. overall OR=0.72, CI (0.69-0.75), p<0.0001), as shown in Figure 4.

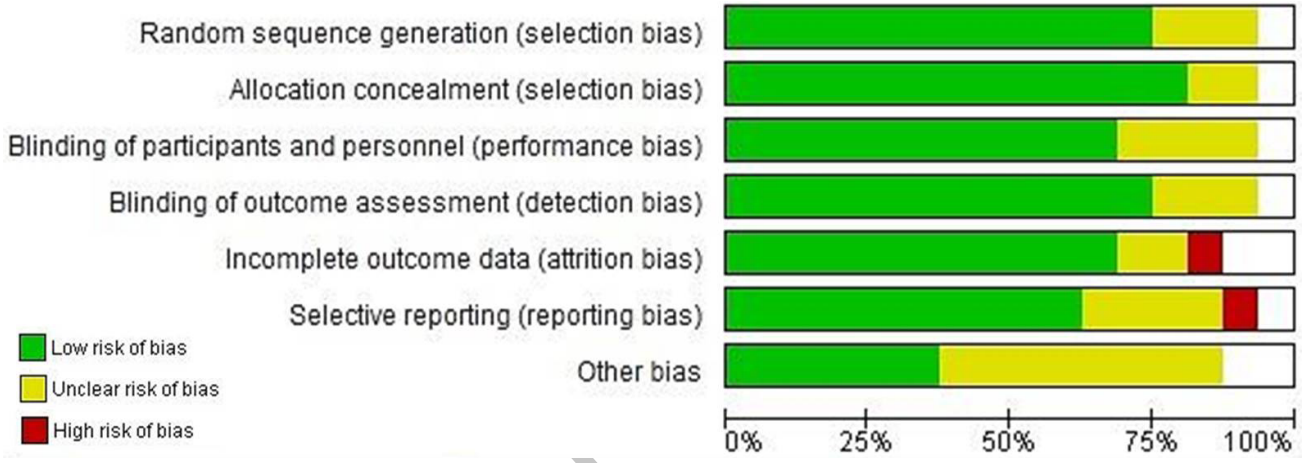

Fig. 2. Study bias summary.

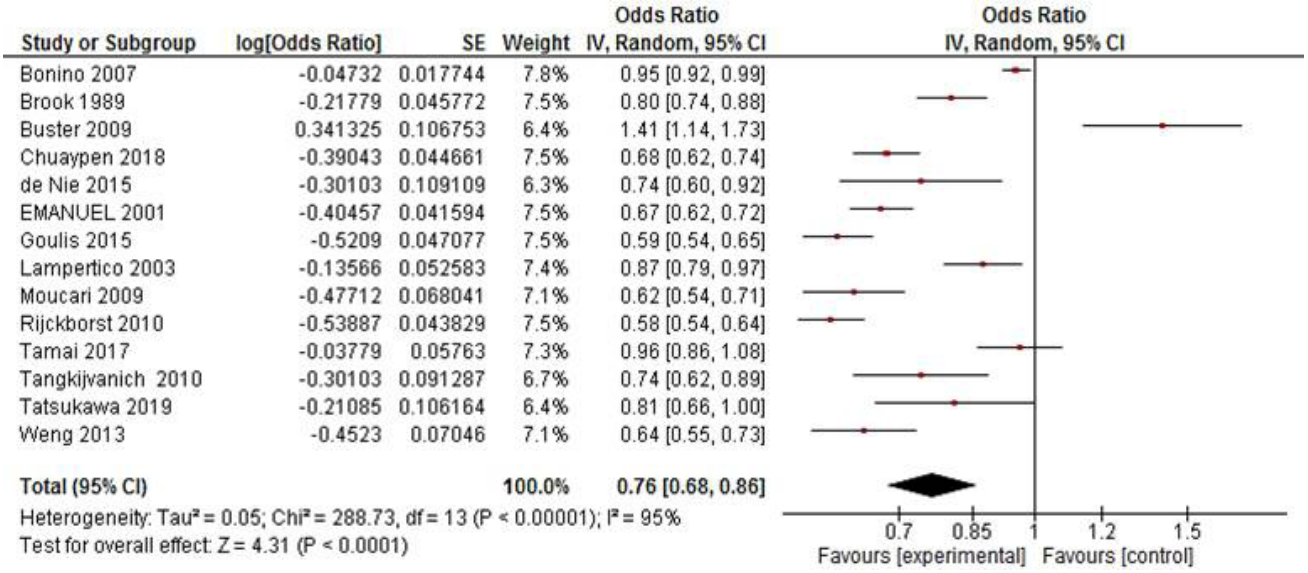

Fig. 3. Distribution of studies based on the ratio of the number of responders to non-responders. The 95% confidence interval for each study is plotted in horizontal lines around the baseline ratio. The rhombic mark is the result of a combination of studies with a 95% confidence interval.

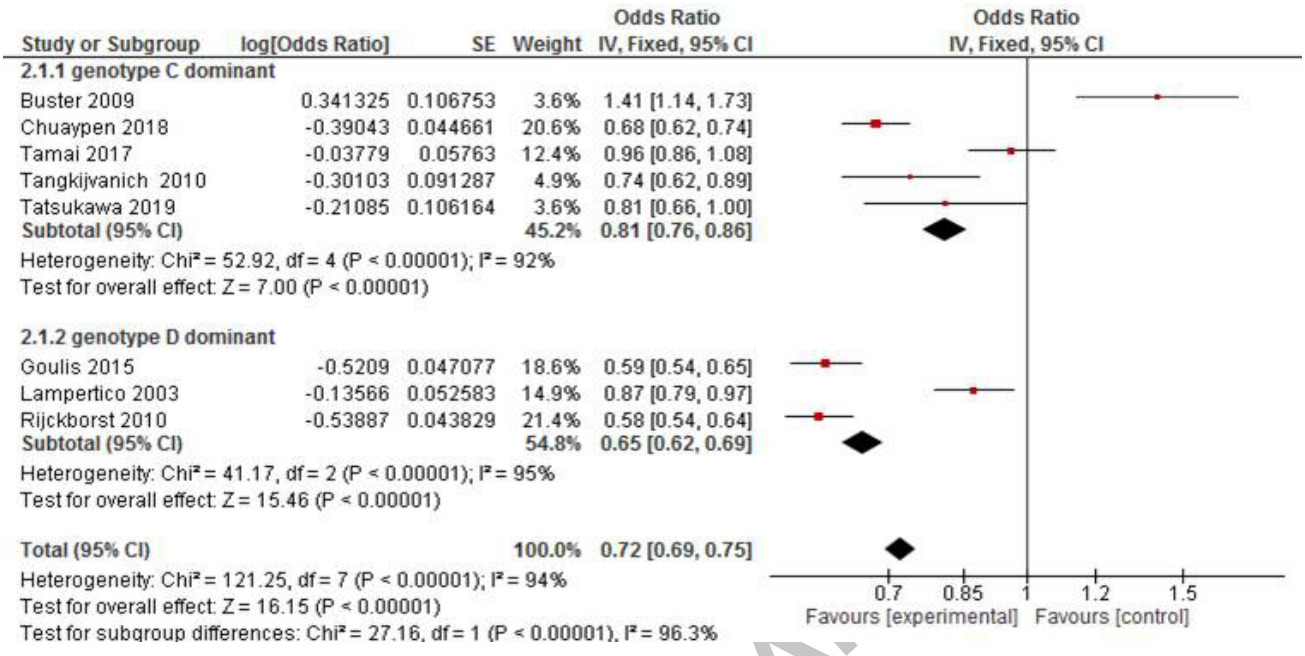

Fig. 4. Distribution of studies having genotype C or D dominant population based on the ratio of the number of responders to non-responders. The 95% confidence interval for each study is plotted in horizontal lines around the baseline ratio. The rhombic mark is the result of a combination of studies with a 95% confidence interval.

DISCUSSION

The biological activity of PEG interferon is related to its interferon fraction. PEG Interferon-alpha 2A binds to the type 1 interferon receptor and dimerizes it. Receptor dimerization activates several intracellular signal transduction pathways, mainly through the JAK/STAT pathway (Sarasin-Filipowicz *et al*., 2009; Grace *et al*., 2005).

This process enhances the phagocytic activity of macrophages and cytotoxic activity of lymphocytes for target cells. The polyethylene glycol portion of the drug protects the drug molecule from proteolytic degradation, thereby extending its half-life and reducing the likelihood of an immune response by physically covering the protein portion of the drug and reducing access to it (Feld and Hoofnagle, 2005; Perry and Jarvis, 2001).

The efficacy of 40 kDa of IGF-interferon was compared with conventional interferon in 194 patients. In that study, PEG interferon decreased HBV compared to conventional interferon in HBeAg negative chronic hepatitis treatment, by DNA and ALT naturalization. The overall conclusions drawn from the effects of the drug on HBV-DNA depletion, HBeAg negative, and liver enzymes normalization were 24% when used with peg-interferon and 12% with conventional interferon (Cooksley *et al*., 2003).

However our study evaluated the response to PEG interferon therapy and revealed that in 1655 subjects with negative Hbe Ag chronic hepatitis, response to treatment happens in a lesser number of patients. While there was high heterogeneity in our analysis, we separated studies based on the most frequent genotype. Results showed a better response in populations with C genotype. Ten genotypes (A-J) and several sub-genotypes have been reported for HBV DNA (Kao *et al*., 2000).

These genotypes are geographically and ethnically dispersed throughout the world, as Genotypes A (serotype adw) and D (serotype ayw) are predominant in Europe and the US, while Genotype B (serotype adw) and C (serotype adr) are widespread in China and Southeast Asia (Schaefer, 2007).

These genotypes cause different courses, the severity of clinical symptoms, response to treatment, resistance to medications like PEG interferon-alpha and resistance to HBe A seroconversion (Liaw *et al*., 2011; Reijnders *et al*., 2011).

Spontaneous HBeAg seroconversion is much higher in patients with genotype B than in patients with genotype C. Virus genotype not only predicts clinical outcomes but also correlates with response to PEG interferon treatment. Genotype A is an independent risk factor for the progression of acute infection to chronic infection. Acute infection in Genotypes A and D patients may be more vulnerable to develop a chronic infection than Genotypes B and C patients. Spontaneous HBeAg seroconversion was

less likely in genotypes C and D than in genotype A and B. Genotype C and D are most commonly associated with severe liver diseases including cirrhosis and hepatocellular carcinoma. The response to interferon treatment, especially in Genotypes A and B, is seen to be better than Genotypes C and D. While our study made a comparison between C and D genotype dominant populations in response to PEG interferon and revealed better response in genotype C dominant populations. We defined subgroups based on the most frequent genotype seen in the population as most studies hadn't separated responders and non-responders based on each genotype.

CONCLUSION AND RECOMMENDATION

According to the results of the present study, it was shown that the proportion of responders to nonresponder individuals treated with PEG-Inf alone was less than one, meaning that those who responded to treatment were less than those who didn't. so PEG interferon may not be the best choice alone. However, in our study, there was heterogeneity. To investigate the source of heterogeneity, as there was no considerable difference in gender and age in included studies (most of the population was male between 30-50 years old), Genotypes and their effects were examined, which genotype C dominant studies had a better response than those with genotype D, but this response was still insufficient.

Statement of conflict of interest

The authors declare no conflict of interest.

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