Int J Clin Exp Med 2015;8(3):3924-3934
www.ijcem.com /ISSN:1940-5901/IJCEM0005570

Original Article

Association between vitamin E and non-alcoholic steatohepatitis: a meta-analysis

Renfan Xu[1], Anyu Tao[1], Shasha Zhang[2], Youbin Deng[1], Guangzhi Chen[2]

[1]Department of Medical Ultrasound, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, People's Republic of China; [2]Department of Internal Medicine and Institute of Hypertension, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, People's Republic of China

Received January 5, 2015; Accepted February 21, 2015; Epub March 15, 2015; Published March 30, 2015

Abstract: Non-alcoholic fatty liver disease (NAFLD) generally has a relatively favorable clinical course; however, non-alcoholic steatohepatitis (NASH) was much more frequently progresses to cirrhosis and hepatocellular carcinoma. We performed a systematic review and meta-analysis of clinical trials to examine the effects of vitamin E supplementation in improving liver histology in NASH. We performed a comprehensive search of the PubMed, Embase and Cochrane databases through October 2014. Weighted mean differences (WMDs) and their respective 95% confidence intervals (CIs) were calculated to assess the efficacy of vitamin E in improving liver histological scores by using fixed effects or random effects. Standard methods were performed to explore statistical heterogeneity and publication bias. Compared with controls, vitamin E supplementation significantly improved all histological parameters, including steatosis (WMD = -0.62, 95% CI: -0.95, -0.77, P = 0.0002), hepatocyte ballooning (WMD = -0.30, 95% CI: -0.56, -0.04, P = 0.03), lobular inflammation (WMD = -0.39, 95% CI: -0.67, -0.11, P = 0.007) and fibrosis (WMD = -0.39, 95% CI: -0.72, -0.06, P = 0.02). Our analysis also indicated the absence of publication bias between NASH and Vitamin E intake. This meta-analysis indicates that vitamin E supplementation had a significant and positive effect in the improvement of steatosis, ballooning degeneration, lobular inflammation and fibrosis in patients with NASH.

Keywords: Non-alcoholic steatohepatitis, vitamin E, histological parameters, meta-analysis

Introduction

Non-alcoholic fatty liver disease (NAFLD) is characterized by fat accumulation in hepatocytes in the absence of significant alcohol intake [1]. NAFLD is the most common liver disease in developed countries, afflicting up to 30% of the general population. It is rapidly spreading in developing countries and has an approximately 10% prevalence [2]. The majority of patients with NAFLD have simple steatosis, which has a benign clinical outcome, whereas approximately 15%-20% of patients will progress to non-alcoholic steatohepatitis (NASH) [3], which includes macrovesicular steatosis, hepatocyte ballooning, lobular inflammation and fibrosis and can develop into cirrhosis or even liver failure [1].

However, the pathogenesis of NASH is not fully understood. The main well-accepted theory is the 'two-hit' hypothesis by Day and James [4]. According to this paradigm, in the 'first hit', insulin resistance promotes the accumulation of triglycerides within the hepatocytes to form steatosis [5, 6]. In the 'second hit', oxidative stress causes lipid peroxidation and activates inflammatory cytokines that induce hepatocyte injury and inflammation in NASH [4]. Oxidative stress appears to result from either excess reactive oxygen species (ROS) production and/or deficient antioxidant capacity [7]. Oxidative stress is observed in most diseases, especially the inflammatory diseases [8]. In addition, many liver diseases have also been associated with high levels of ROS, which is linked to disease progression by influencing the magnitude of oxidative protein and lipid modifications [9]. Thus, it is reasonable to postulate the possibility of using antioxidant therapy for NASH.

Vitamin E (α-tocopherol), a type of lipid-soluble, chain-breaking antioxidant, is an essential micronutrient that is mainly acquired from the

intake of vegetables, fruit, fortified breakfast, fortified beverages, supplements and so on [10]. Vitamin E can protect cellular structural integrity against damage from lipid peroxidation and oxygen-free radicals [10]. Epidemiologic studies have reported that plasma α-tocopherol is lower in NASH patients than healthy controls [11]. In vivo and in vitro studies have demonstrated that vitamin E attenuated NASH via multiple mechanisms, including up-regulation of superoxide dismutase activity and inhibition of genes related to inflammation, fibrosis and hepatocellular necrosis [12-15]. This has led to enthusiasm over the possibility that vitamin E has beneficial effects on the inhibition of NASH progression.

There is no currently approved treatment for NASH. Patients often are advised to engage in physical activity and lose weight, which is difficult to achieve and harder to maintain [16, 17]. In the United States, vitamin E capsules are popular for the adult population, and approximately 12.7% of adults take daily vitamin E supplements [18]. Several randomized controlled trials are currently underway to test whether down-regulation of oxidative stress with high doses of vitamin E supplementation attenuates the progression of NASH [19-23]. However, the results of these trials have been inconsistent, and the sample sizes have been relatively modest. To better understand the effect of vitamin E on NASH progression, we systematically reviewed all randomized controlled trials (RCTs) to evaluate the effect of vitamin E supplementation on the histological parameters of NASH.

Methods

Search strategy

We searched the PubMed, EMBASE and Cochrane library databases from their inception through October 2014 for trials that described the association between vitamin E, assessed as dietary intake, and histological outcomes of NASH. The following search terms were used: (vitamin E OR alpha-tocopherol OR tocopherols OR tocotrienols) AND (non-alcoholic fatty liver disease OR non-alcoholic steatohepatitis OR fatty liver OR steatosis OR NAFLD OR NASH). In addition, we reviewed the references of all retrieved relevant articles and reviews for published randomized controlled trials that met the inclusion criteria. The search was confined to human studies, and no language restriction was applied.

Study selection

Relevant studies were selected by two independent reviewers, and disagreement was settled by consensus. The inclusion criteria for the meta-analysis were as follows: (1) randomized controlled trials with either a parallel or a crossover design, (2) adults with NASH supplied with vitamin E which is currently FDA-approved for at least treatment for 24 weeks, (3) diagnosis of NASH determined by histology and (4) liver biopsy performed at the beginning and end of the clinical trials. As the trials included in this meta-analysis were limited, we also included the articles that combined vitamin E with vitamin C (ascorbic acid) as the treatment group because vitamin C is also an anti-oxidant agent and can enhance regeneration of oxidized vitamin E [24].

The exclusion criteria were as follows: (1) NASH patients were diagnosed by elevated transaminases and/or imaging techniques (CT scan, magnetic resonance imaging, ultrasound) without liver biopsy; (2) studies enrolling patients with co-existing liver disease, including alcoholic liver disease, autoimmune hepatitis, hepatitis B, hepatitis C and so on; and (3) studies that involved drugs, bariatric surgery, environmental toxins or total parenteral nutrition, which may cause secondary NAFLD.

Data extraction

Two independent investigators reviewed all collected trials, and disagreements were resolved by consensus. The following characteristics of the identified studies were recorded: trial characteristics (authors, publication year, sample size, study design); treatment characteristics (type of intervention, dosage, study duration); and patient characteristics (number of participants, mean age, gender). The primary analyzed outcome was the histological features, including the number improved or not improved in histological outcomes and the net changes in histological features, including steatosis grade (0-3), lobular inflammation score (0-3), hepatocyte ballooning score (0-2) and fibrosis stage (0-4) [25].

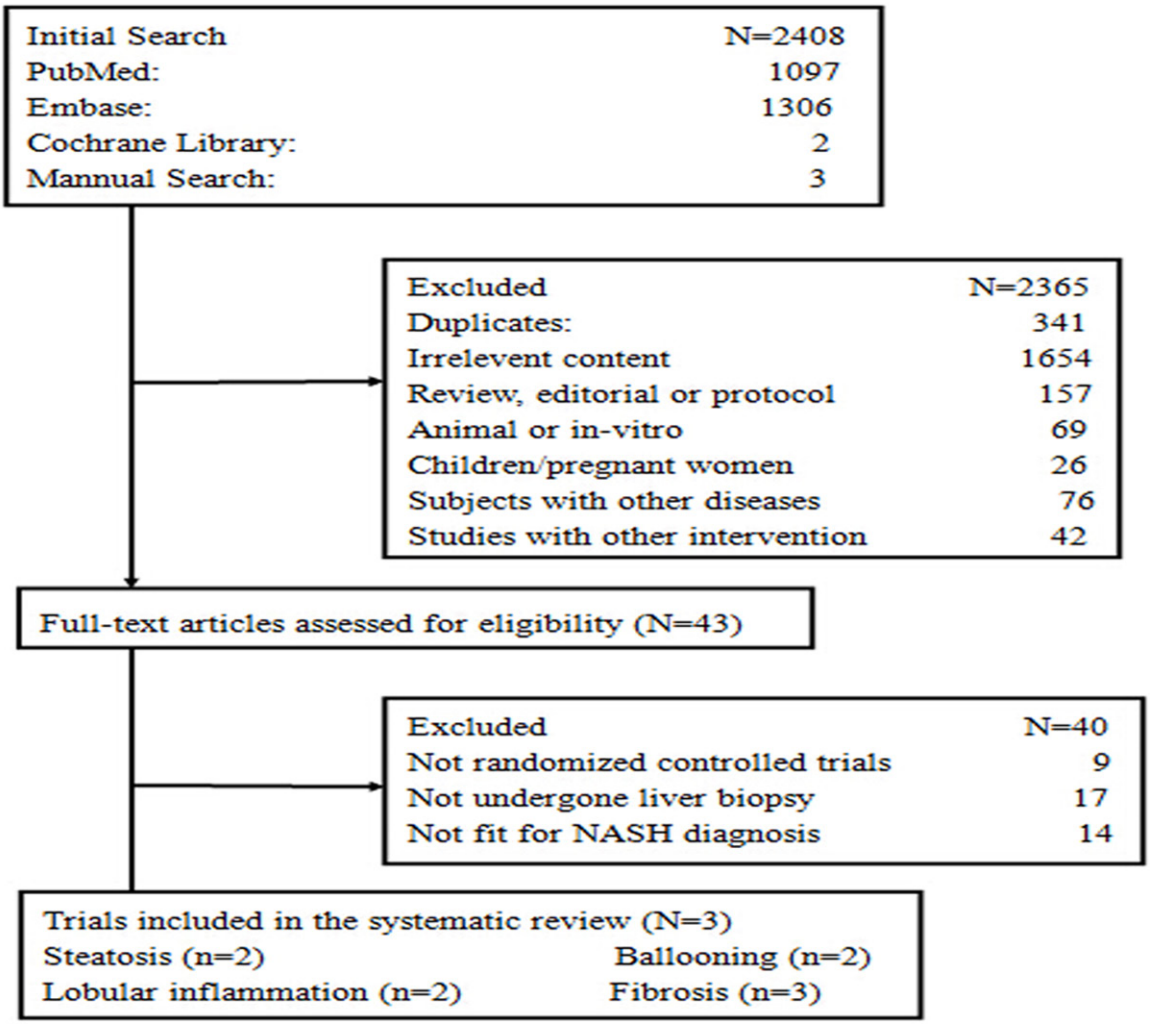

Figure 1. Flow diagram of the process of article selecting eligible publications.

Quality assessment

We evaluated and scored the quality of studies based on the Jadad scoring system [26]. The criteria (1 point each) included reporting of trials, including randomization, allocation concealment, double-blinding, generation of random numbers, description of withdrawals and dropouts. The possible score ranged between 0 and 5. Higher score represented a better quality.

Statistical analysis

We performed statistical analysis using RevMan software version 5 (Cochrane Collaboration). Histological features were assessed as both a continuous variable (mean change in histological scores, expressed as the weighted mean difference (WMDs) and 95% confidence interval (CI)) and a dichotomous outcome (subjects improved compared with subjects unchanged or worsened, expressed as relative risk (RR) and 95% CI). Secondary outcomes were expressed as continuous variables. Fixed-effect or random-effect models were used in this meta-analysis. If the variances for the mean changes were not provided, the paired differences were imputed by assuming a correlation coefficient of 0.5 between variances at baselines and ends of trials according to the method of Follmann et al [27]. We assumed equal variances during the trial and between the intervention and control groups.

Cochran's Q statistic was assessed to evaluate the heterogeneity, and the inconsistency index (I^2) was evaluated to quantify the degree of inconsistency across individual studies. A P value < 0.10 was considered significant, and a

Table 1. Main characteristics of all studies included in the meta-analysis

Trial	Country	Vitamin E group intervention	Control group	Duration (weeks)	Study Size (n)		Paried Biopsies (n)		Mean age (year)		Male (%)		Scoring system
					T	C	T	C	T	C	T	C	
Harrison 2003	USA	α-tocopherol 1000 IU/d + Vit C 1000 mg/d	Placebo	24	23	22	23	22	52.5	50.2	39	50	Brunt
Dufour 2006	Switzerland	d-tocopherol 800 IU/d + UDCA 12-15 mg/kg/d	UDCA	96	15	18	10	11	46	47	67	72	Promrat
Sanyal 2010	USA	RRR-α-tocopherol 800 IU/d	Placebo	96	84	83	80	72	46.6	45.4	38	42	Kleiner

T: treatment; C: control; vit C: vitamin C; UDCA: ursodeoxycholic acid.

Table 3. Subgroup analysis and sensitivity analysis of histological score

Variables	Steatosis			Ballooning			Lobular inflammation			Fibrosis		
	No. of trials	Mean difference (95% CI)	P for heterogeneity	No. of trials	Mean difference (95% CI)	P for heterogeneity	No. of trials	Mean difference (95% CI)	P for heterogeneity	No. of trials	Mean difference (95% CI)	P for heterogeneity
Vit.E dosage												
≤ 800 mg/d	0	NA	NA	0	NA	NA	0	NA	NA	1	-0.25 (-0.72, 0.22)	NA
> 800 mg/d	2	-0.62 (-0.95, -0.29)	0.76	2	-0.30 (-0.56, -0.04)	0.97	2	-0.39 (-0.67, -0.11)	0.80	2	-0.44 (-0.90, 0.02)	0.03
Duration												
≤ 24 week	0	NA	NA	0	NA	NA	0	NA	NA	1	-0.25 (-0.72, 0.22)	NA
> 24 week	2	-0.62 (-0.95, -0.29)	0.76	2	-0.30 (-0.56, -0.04)	0.97	2	-0.39 (-0.67, -0.11)	0.80	2	-0.44 (-0.90, 0.02)	0.03
Quality score												
≥ 4	0	NA	NA	0	NA	NA	0	NA	NA	1	-0.25 (-0.72, 0.22)	NA
< 4	2	-0.62 (-0.95, -0.29)	0.76	2	-0.30 (-0.56, -0.04)	0.97	2	-0.39 (-0.67, -0.11)	0.80	2	-0.44 (-0.90, 0.02)	0.03
Sensitivity analysis												
Trials use vitE only as treatment	1	-0.60 (-0.95, -0.25)	NA	1	-0.30 (-0.57, -0.03)	NA	1	-0.40 (-0.69, -0.11)	NA	1	-0.20 (-0.50, 0.10)	NA

VitE: vitamin E; NA: not applicable.

Table 2. Methodological quality of included trials

Authors	Randomisation	Allocation concealment	Random sequence generation	Blinding	Reporting of withdrawals	Jadad score
Harrison 2003	Y	Y	Y	Y	Y	5
Dufour 2006	Y	U	U	Y	Y	3
Sanyal 2010	Y	U	U	Y	Y	3

Y: yes; U: unclear.

measurement of $I^2 > 50\%$ was taken to indicate substantial heterogeneity [28]. If heterogeneity existed among the studies, the random-effect model (the Dersimonian and Laird method) was used to calculate the pooled RR. Otherwise, a fixed-effect model (the Mantel-Haenszel method) was used for outcomes without obvious heterogeneity [29]. The funnel plots and Egger test were assessed to examine publication bias [30]. To explore whether the results could have been markedly affected by the use of other drugs, sensitivity analyses were performed by excluding studies combine vitamin E and other drugs as treatment group. Subgroup analyses were conducted to assess the possible influence of covariates on net changes of primary outcomes, including vitamin E dosages, vitamin E duration and treatment with vitamin E alone. Except where otherwise specified, A *P* value < 0.05 was considered statistically significant in this meta-analysis.

Results

Literature search

Using primary electronic databases and searching for studies dated up to October 2014, a total of 1084 references on vitamin E for NAFLD patients were identified. Of these, 114 duplicated references and 970 obviously irrelevant references were excluded by review titles and abstracts. Then, 43 references were retrieved and reviewed in full, and a further 40 articles were excluded. Thus, 3 articles were finally selected for this meta-analysis [19-21]. A flow chart presenting the study selection process is shown in **Figure 1**.

Study characteristics

Table 1 shows the characteristics of the 3 included studies. Three published studies with a total of 245 participants, including 122 subjects in the vitamin E group and 123 patients in the control group, were published between 1983 and 2012. All of the studies were case-control trials. Two studies were conducted in the United States, and one study was conducted in Europe. All studies focused on adult NASH patients with liver biopsies. The number of subjects in studies ranged from 45 to 167 cases. All intervention studies declared having a randomized, parallel design. Of note, one trial supplied vitamin E in combination with vitamin C to patients. One had three arms (UDCA + vitamin E, UDCA, placebo), and we chose the arm of UDCA + vitamin E as the treatment group and the UDCA arm as the control group. The dosage of vitamin E supplements ranged from 800 to 1000 IU/d, with an intervention duration ranging from 24 to 96 weeks. The primary outcomes of histological score were examined: steatosis (2 studies), ballooning (2 studies), lobular inflammation (2 studies) and fibrosis (3 studies). Among the studies, Dufour 2006 and Sanyal 2010 analyzed the effects of vitamin E treatment on all four histological features, whereas Harrison 2003 only analyzed the effect of vitamin E treatment on fibrosis improvement. The characteristics of these three trials are described in **Table 1**.

Study quality

All three included trials were randomized and controlled trials. All included trials were double-blinded, and complete outcome data were adequately addressed in these three trials (**Table 2**). One trial reported using the method of sequence generation [21], and 1 reported using allocation concealment [21]. Only 1 trial was classified as high quality (Jadad score of 4 or 5) [21], and 2 studies were low quality (Jadad score ≤ 3) [19, 20].

Effect of vitamin E on histological parameters

As shown in **Figure 2**, vitamin E supplementation had a significant association with histological improvement of NASH, including steatosis, ballooning, lobular inflammation and fibrosis.

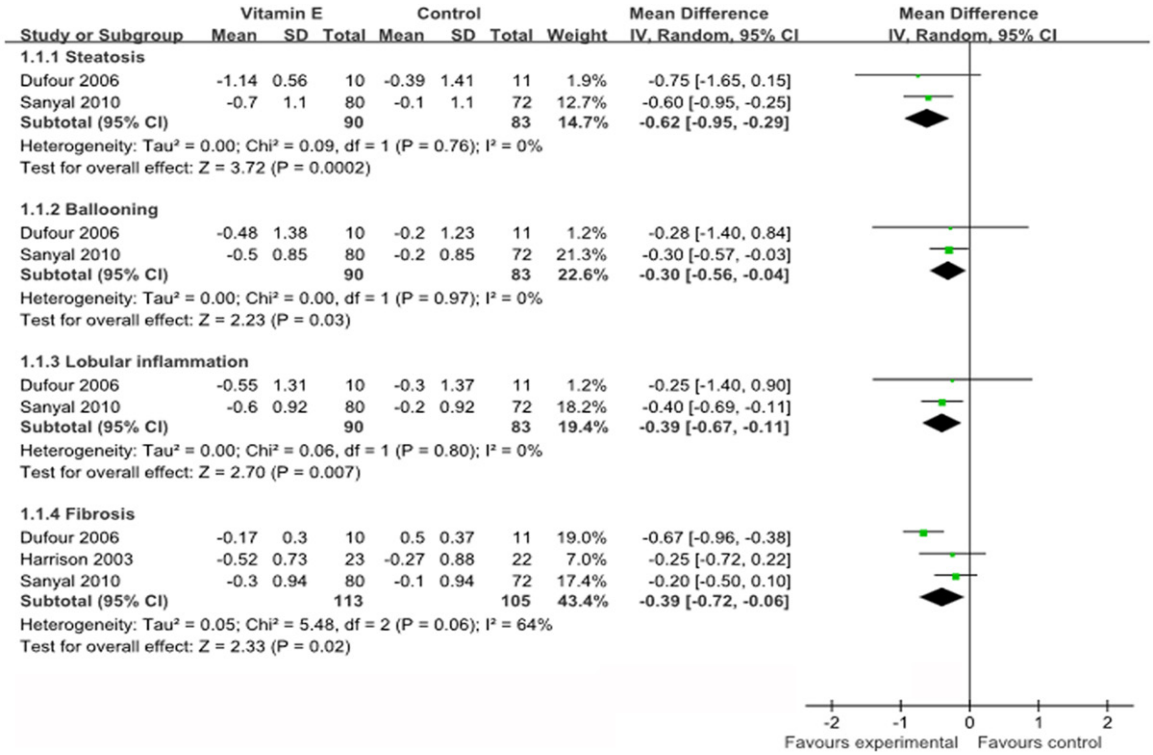

Figure 2. Forest plot of randomised controlled trials investigate the association between vitamin E supplementation and histological score.

Two studies (Dufour et al. and Sanyal et al.) with 173 participants, including 90 subjects in the vitamin E groups and 83 subjects in the placebo groups, reported steatosis, ballooning and lobular inflammation at baseline and follow-up. The mean change in steatosis was more significantly reduced in subjects receiving supplemented with vitamin E (WMD = -0.62, 95% CI: -0.95, -0.29, P = 0.0002) than in controls. Heterogeneity was not demonstrated for this outcome (I^2 = 0%, *P* = 0.76). In addition, a small but significant reduction was observed in hepatic ballooning for subjects supplemented with vitamin E (WMD = -0.30, 95% CI: -0.56, -0.04, *P* = 0.03) compared with control subjects. The results indicated homogeneity (I^2 = 0%, *P* = 0.97). In addition, lobular inflammation was also found to be significantly improved with vitamin E supplementation (WMD = -0.39, 95% CI: -0.67, -0.11, *P* = 0.007) compared with controls, with no evidence of heterogeneity (I^2 = 0%, *P* = 0.80). For all 3 trials that reported data on fibrosis, a significant improvement in fibrosis was observed in subjects of the vitamin E group (WMD = -0.39, 95% CI: -0.72, -0.06, *P* = 0.02) compared with the control group, with significant heterogeneity (I^2 = 64%; *P* = 0.06).

Subgroup analysis

We conducted subgroup analysis to explore the dose-effect relationship, study-duration effects and study quality. The analyses of all subgroups are provided in **Table 3**. Vitamin E consumption was divided into high dosage (> 800 mg/d) or low dosage (≤ 800 mg/d). The study duration was categorized as either the longer-term subgroup (> 24 wk) or shorter-term subgroup (≤ 24 wk). In addition, subgroup analysis indicated that trials have a higher quality (≥ 4) or a lower quality (< 4). Subgroup analysis revealed that significant improvements of steatosis, ballooning and lobular inflammation were not influenced by vitamin E dosage, treatment duration or quality score. Moreover, subgroup analysis indicated that vitamin E had no significant effect on fibrosis in either the high-dosage group or the low-dosage group. In addition, no significant improvement in fibrosis was observed in the longer-term subgroup and shorter-term subgroup. Furthermore, in the higher-quality and lower-quality subgroups, beneficial effects of vitamin E on fibrosis were also not observed.

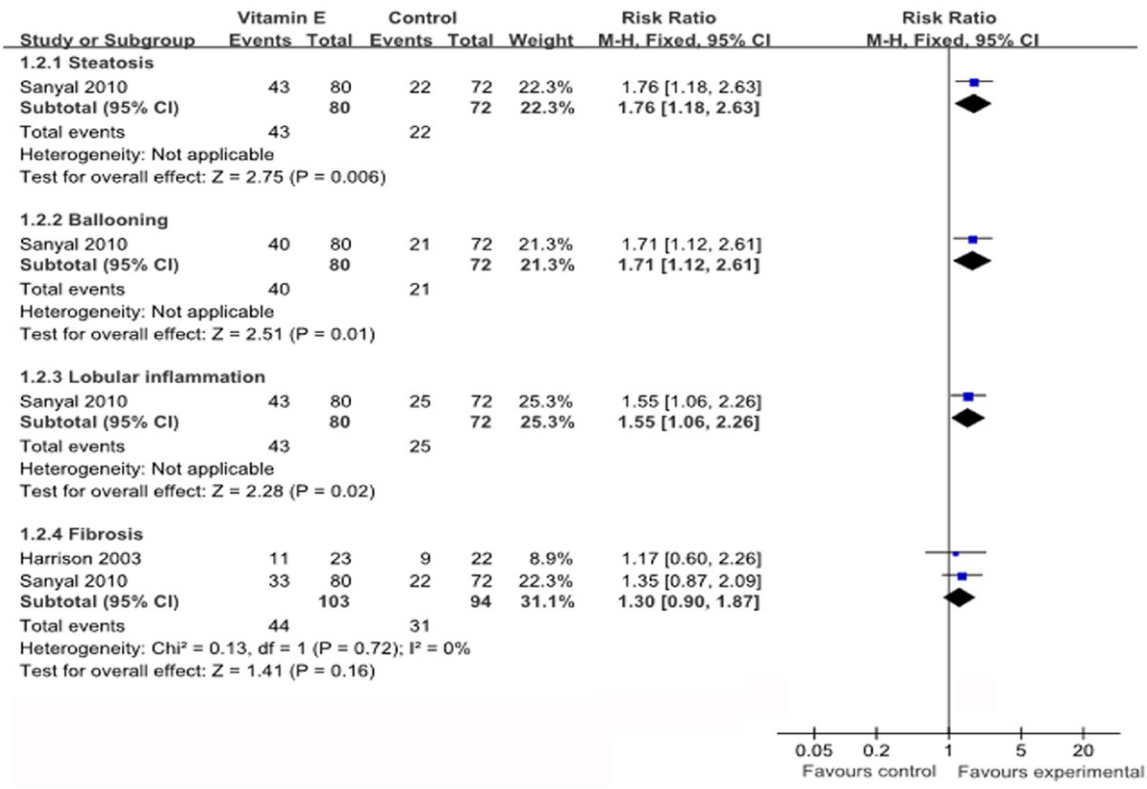

Figure 3. Forest plot of randomised controlled trials investigate the association between vitamin E supplementation and histological improvement.

Table 4. Secondary outcome: biochemical and anthropometric response

	No. of trials	Mean difference (95% CI)	*P* value	I^2 (%)	P for heterogeneity
ALT	3	3.50 (-17.90, 24.90)	0.75	77	0.01
AST	2	-17.64 (-27.29, -7.99)	0.0003	0	0.93
BMI	3	-0.16 (-0.65, 0.33)	0.53	0	0.91

ALT: alanine aminotransferase; AST: aspartate aminotransferase; BMI: body mass index; CI: confidence interval.

A sensitivity analysis was performed by excluding trials (Harrison 2003 and Dufour 2006) using vitamin E and other drugs as the treatment group (**Table 3**). After excluding the trials, the pooled reduction of steatosis, ballooning and lobular inflammation were not significantly influenced, with values of -0.60 (95% CI: -0.95, -0.25; *P* = 0.0008), -0.30 (95% CI: -0.57, -0.03; *P* = 0.03) and -0.40 (95% CI: -0.69, -0.11; *P* = 0.007), respectively. In addition, the exclusion of trials resulted in no obvious reductions in fibrosis (-0.20, 95% CI: -0.50, 0.10; *P* = 0.19), which significantly influenced the results.

Subjects with histological improvement

Two studies reported subjects with fibrosis improvement [19, 21]. The pooled results indicated that vitamin E use (n = 103) did not demonstrate a significant improvement in fibrosis (RR 1.30, 95% CI 0.90-1.87, I^2 = 0%, P = 0.16) compared with the control (n = 94), with a similar result when including only the high-quality trial (RR 1.17, 95% CI 0.60-2.26, *P* = 0.64).

Only one study reported subjects with improvement in steatosis, ballooning and lobular inflammation [19]. Participants treated with vitamin E (n = 80) were more likely to display an improvement in steatosis (RR 1.76, 95% CI 1.18-2.63, *P* = 0.006), ballooning (RR 1.71, 95% CI 1.12-2.61, *P* = 0.01) and lobular inflammation (OR 1.55, 95% CI 1.06-2.26, *P* = 0.02) compared with controls (n = 72) (**Figure 3**).

Liver function and body mass index (BMI)

Changes in ALT and BMI were reported in all three studies, and the change of AST was only available in two studies [19, 20]. Participants

treated with vitamin E displayed a significant improvement in AST compared with those in the control group (SMD -17.64 U/L, 95% CI -27.29, -7.99; P = 0.0003, I^2 = 0%). However, patients treated with vitamin E did not demonstrate a significant improvement in neither ALT (SMD 3.50 U/L, 95% CI -17.90 to 24.90, P = 0.75, I^2 = 77%) or BMI (SMD -0.16, 95% CI -0.65 to 0.33, P = 0.53, I^2 = 0%) compared with the controls (**Table 4**).

Adverse events and publication bias

Vitamin E was well tolerated, and no mortality or harm was reported following supplementation in any of these trials. The funnel plots of the studies were symmetric for fibrosis, steatosis, ballooning and lobular inflammation upon visual examination. Furthermore, the results of the Egger's test did not support the existence of publication bias.

Discussion

In this meta-analysis, we identified three randomized controlled trials with 245 participants that assessed the effects of vitamin E supplementation on improving histological parameters for NASH patients. We found that vitamin E supplementation significantly improved all histological scores, including steatosis, ballooning, lobular inflammation and fibrosis, compared with the control group. However, when we analyzed the subjects with histological improvement, subjects with vitamin E supplementation were only improved in steatosis, ballooning and lobular inflammation vs. the control group and not fibrosis. This may be because only two trials with limited data analyzed the histological improvement in subjects with vitamin E supplementation. Therefore, it is difficult to draw firm conclusions based on the present review.

A number of studies have analyzed the effects of vitamin E on NASH, and further evidence has been provided for the biological plausibility of these findings. Oxidative stress results from an imbalance between excessive generation of reactive oxygen species (ROS) and decreased antioxidant defenses [7]. Oxidative stress, which plays a vital role in the progression of steatosis to NASH, is one of the most popular proposed mechanisms of hepatocellular injury and considered as a critical part of the aforementioned second 'hit' [4]. Vitamin E, a lipid-soluble chain breaking antioxidant, is considered to be the most important natural antioxidant [10]. It stabilizes free radical compounds by complexing with unpaired electrons and protects against lipid peroxidation by acting directly with a variety of oxygen radicals [10]. Thus, vitamin E is a good candidate for investigation of diseases that involve ROS as a main component.

Animal experiments indicate that the improvement in histological parameters and liver function is associated with vitamin E supplementation. In a methionine-choline-deficient (MCD) diet-induced model of steatohepatitis, vitamin E treatment decreased oxidative stress markers, reduced liver enzymes and diminished histological steatosis, necroinflammation and fibrosis [12]. Furthermore, Vitamin E treatment also provided protection against TGF-β1, a cytokine closely associated with fibrosis development in liver [13]. Observational prospective cohorts and case-control studies have been conducted to analyze the effect of vitamin E supplementation on NASH, but the results are conflicting. Erhardt et al. demonstrated that the levels of α-tocopherol were significantly decreased in NASH patients compared with controls [11]. A large multicenter randomized controlled trial of adults revealed that vitamin E supplementation can significantly improve hepatic steatosis, lobular inflammation, and hepatocellular ballooning in NASH patients, whereas a reduction in fibrosis is not obvious [19]. Another large case-control trial demonstrated that vitamin E supplementation exerted beneficial effects on ballooning in children and adolescents patients with NAFLD, but no significant improvement in steatosis, inflammation and fibrosis was observed [22] .

Subgroup and sensitivity analyses indicated that the histological scores of steatosis, ballooning and lobular inflammation were not influenced by the treatment doses of vitamin E, study duration, or quality of the study, whereas for the fibrosis score, no significant improvement was observed in all of the above subgroups. Thus, further trials with more subjects are needed to make a definite conclusion. We also analyzed the effects of vitamin E supplementation on biochemical and metabolic variables in this pooled analysis. The level of AST instead of ALT displayed significant reduction in the vitamin E group compared with controls.

This is not consistent with some previous studies that revealed that vitamin E supplementation significantly improved liver function [31, 32]. This may be because we only included trials with liver biopsy, so not all trials analyzing the effect of vitamin E supplementation on ALT were included in this meta-analysis. Vitamin E supplementation did not result in a significant reduction in BMI compared with the control group.

There are some strengths of this systematic review. First, the trials included in this study were all randomized controlled trials, which increased internal causality with reliable inferences. In addition, the participants included in trials were all histologically proven NASH patients, which minimized the misclassification bias and made it meaningful for histological parameters to be directly associated with the outcome of NASH. Third, the participants included in this meta-analysis were derived from two continents (North America and Europe). The follow-up was adequate with few dropouts in trials.

Some limitations of this meta-analysis should be noted. First, the duration of therapy with a maximum of 96 weeks was not long enough to detect long-term histological change complications. Fibrosis progression into cirrhosis, liver carcinoma or even mortality is slow, and a longer follow-up period is needed [33]. Second, the number of trails included was limited, and the number of participants in the trials was low. It is hard to make definite conclusions using these limited data. Third, the degree of improvement is modest, estimated at approximately half of a grade per year for histological parameter. Though the improvements were significant compared with the controls, it is hard to achieve the expectation of patients. Fourth, the vitamin E formulation and dosage varied across the studies. For example, two studies used α-tocopherol 800 IU, whereas the other study used α-tocopheryl acetate 1000 IU. This led to difficulty in investigating the effect of various dosage regimens. For future investigations, researchers may wish to recognize a safety margin of vitamin E supplementation in the treatment of NASH. Although no serious side effects of vitamin E intake were reported in the included trials, the safety of high-dosage vitamin E supplementation has already been a concern in many trials. A meta-analysis by Miller ER revealed that, in patients with chronic diseases, a vitamin E dosage > 400 IU per day may increase all-cause mortality by antagonizing vitamin K functions in individuals routinely ingesting various drugs and increasing the risk of prolonged bleeding time [34].

In conclusion, this meta-analysis revealed that vitamin E supplementation resulted in significant improvements in histological parameters in NASH patients. There is currently no widely accepted treatment for non-alcoholic steatohepatitis. Over the past several years, studies with vitamin E supplementation in NASH patients have yielded promising results. Vitamin E is well-tolerated, and no morbidity or obvious mortality was reported. However, the results in this conclusion cannot be generalized, as trials were limited, with a short follow-up duration and a small number of participants. Moreover, additional large-scale high-quality studies are needed to investigate the effect of vitamin E supplementation on NASH patients with outcomes (histological parameters, biochemical variables and adverse events) oriented to obtain more comprehensive information on supplementation for clinical use.

Acknowledgements

The present study was supported by the National Natural Science Foundation of China (Nos. 81400369). R.-F. X. and G.-Z.C. conceived the study design; A.-Y. T. and S.-S.Z. searched and selected the articles, and extracted, analysed and interpreted the data; R.-F. X. and G.-Z.C. wrote the manuscript. All authors read and approved the final manuscript.

Disclosure of conflict of interest

None.

Address correspondence to: Dr. Guangzhi Chen, Department of Internal Medicine and Institute of Hypertension, Tongji Hospital, Tongji Medical College, Huazhong University of Science & Technology, 1095 Jiefang Ave., Wuhan 430030, China. Tel: +86-27-8366-3280; Fax: +86-27-8366-3280; E-mail: chengz2003@163.com

References

[1] Chalasani N, Younossi Z, Lavine JE, Diehl AM, Brunt EM, Cusi K, Charlton M, Sanyal AJ; American Gastroenterological Association; Ameri-

can Association for the Study of Liver Diseases; American College of Gastroenterologyh. The diagnosis and management of non-alcoholic fatty liver disease: practice Guideline by the American Association for the Study of Liver Diseases, American College of Gastroenterology, and the American Gastroenterological Association. Hepatology 2012; 55: 2005-23.

[2] Smith BW, Adams LA. Non-alcoholic fatty liver disease. Crit Rev Clin Lab Sci 2011; 48: 97-113.

[3] Bellentani S, Marino M. Epidemiology and natural history of non-alcoholic fatty liver disease (NAFLD). Ann Hepatol 2009; 8 Suppl 1: S4-8.

[4] Day CP, James OF. Steatohepatitis: a tale of two "hits"? Gastroenterology 1998; 114: 842-5.

[5] Cusi K. Role of insulin resistance and lipotoxicity in non-alcoholic steatohepatitis. Clin Liver Dis 2009; 13: 545-63.

[6] Oh MK, Winn J, Poordad F. Review article: diagnosis and treatment of non-alcoholic fatty liver disease. Aliment Pharmacol Ther 2008; 28: 503-22.

[7] Caldwell SH. Mitochondria in nonalcoholic fatty liver disease. Clin Liver Dis 2004; 8: 595-617.

[8] Singal AK, Jampana SC, Weinman SA. Antioxidants as therapeutic agents for liver disease. Liver Int 2011; 31: 1432-48.

[9] Le TH, Caldwell SH, Redick JA, Sheppard BL, Davis CA, Arseneau KO, Iezzoni JC, Hespenheide EE, Al-Osaimi A, Peterson TC. The zonal distribution of megamitochondria with crystalline inclusions in nonalcoholic steatohepatitis. Hepatology 2004; 39: 1423-9.

[10] Clarke MW, Burnett JR, Croft KD. Vitamin E in human health and disease. Crit Rev Clin Lab Sci 2008; 45: 417-50.

[11] Erhardt A, Stahl W, Sies H, Lirussi F, Donner A, Häussinger D. Plasma levels of vitamin E and carotenoids are decreased in patients with Nonalcoholic Steatohepatitis (NASH). Eur J Med Res 2011; 16: 76-8.

[12] Nan YM, Wu WJ, Fu N, Liang BL, Wang RQ, Li LX, Zhao SX, Zhao JM, Yu J. Antioxidants vitamin E and 1-aminobenzotriazole prevent experimental non-alcoholic steatohepatitis in mice. Scand J Gastroenterol 2009; 44: 1121-31.

[13] Phung N, Pera N, Farrell G, Leclercq I, Hou JY, George J. Pro-oxidant-mediated hepatic fibrosis and effects of antioxidant intervention in murine dietary steatohepatitis. Int J Mol Med 2009; 24: 171-80.

[14] Soden JS, Devereaux MW, Haas JE, Gumpricht E, Dahl R, Gralla J, Traber MG, Sokol RJ. Subcutaneous vitamin E ameliorates liver injury in an in vivo model of steatocholestasis. Hepatology 2007; 46: 485-95.

[15] Iida C, Fujii K, Koga E, Washino Y, Kitamura Y, Ichi I, Abe K, Matsura T, Kojo S. Effect of alpha-tocopherol on carbon tetrachloride intoxication in the rat liver. Arch Toxicol 2009; 83: 477-83.

[16] Peng L, Wang J, Li F. Weight reduction for non-alcoholic fatty liver disease. Cochrane Database Syst Rev 2011; CD003619.

[17] Keating SE, Hackett DA, George J, Johnson NA. Exercise and non-alcoholic fatty liver disease: a systematic review and meta-analysis. J Hepatol 2012; 57: 157-66.

[18] Radimer K, Bindewald B, Hughes J, Ervin B, Swanson C, Picciano MF. Dietary supplement use by US adults: data from the National Health and Nutrition Examination Survey, 1999-2000. Am J Epidemiol 2004; 160: 339-49.

[19] Sanyal AJ. Pioglitazone, vitamin E, or placebo for nonalcoholic steatohepatitis. N Engl J Med 2010; 362: 1675-85.

[20] Dufour JF, Oneta CM, Gonvers JJ, Bihl F, Cerny A, Cereda JM, Zala JF, Helbling B, Steuerwald M, Zimmermann A; Swiss Association for the Study of the Liver. Randomized placebo-controlled trial of ursodeoxycholic acid with vitamin e in nonalcoholic steatohepatitis. Clin Gastroenterol Hepatol 2006; 4: 1537-43.

[21] Harrison SA, Torgerson S, Hayashi P, Ward J, Schenker S. Vitamin E and vitamin C treatment improves fibrosis in patients with nonalcoholic steatohepatitis. Am J Gastroenterol 2003; 98: 2485-90.

[22] Lavine JE, Schwimmer JB, Van Natta ML, Molleston JP, Murray KF, Rosenthal P, Abrams SH, Scheimann AO, Sanyal AJ, Chalasani N, Tonascia J, Ünalp A, Clark JM, Brunt EM, Kleiner DE, Hoofnagle JH, Robuck PR; Nonalcoholic Steatohepatitis Clinical Research Network. Effect of vitamin E or metformin for treatment of nonalcoholic fatty liver disease in children and adolescents: the TONIC randomized controlled trial. JAMA 2011; 305: 1659-68.

[23] Nobili V, Manco M, Devito R, Di Ciommo V, Comparcola D, Sartorelli MR, Piemonte F, Marcellini M, Angulo P. Lifestyle intervention and antioxidant therapy in children with nonalcoholic fatty liver disease: a randomized, controlled trial. Hepatology 2008; 48: 119-28.

[24] Chang CY, Argo CK, Al-Osaimi AM, Caldwell SH. Therapy of NAFLD: antioxidants and cytoprotective agents. J Clin Gastroenterol 2006; 40 Suppl 1: S51-60.

[25] Kleiner DE, Brunt EM, Van Natta M, Behling C, Contos MJ, Cummings OW, Ferrell LD, Liu YC, Torbenson MS, Unalp-Arida A, Yeh M, McCullough AJ, Sanyal AJ; Nonalcoholic Steatohepatitis Clinical Research Network. Design and validation of a histological scoring system

for nonalcoholic fatty liver disease. Hepatology 2005; 41: 1313-21.
[26] Moher D, Pham B, Jones A, Cook DJ, Jadad AR, Moher M, Tugwell P, Klassen TP. Does quality of reports of randomised trials affect estimates of intervention efficacy reported in meta-analyses? Lancet 1998; 352: 609-13.
[27] Follmann D, Elliott P, Suh I, Cutler J. Variance imputation for overviews of clinical trials with continuous response. J Clin Epidemiol 1992; 45: 769-73.
[28] Higgins JP, Thompson SG. Quantifying heterogeneity in a meta-analysis. Stat Med 2002; 21: 1539-58.
[29] DerSimonian R, Laird N. Meta-analysis in clinical trials. Control Clin Trials 1986; 7: 177-88.
[30] Egger M. Bias in meta-analysis detected by a simple, graphical test. BMJ 1997; 315: 629-34.
[31] Lavine JE. Vitamin E treatment of nonalcoholic steatohepatitis in children: a pilot study. J Pediatr 2000; 136: 734-8.
[32] Yakaryilmaz F, Guliter S, Savas B, Erdem O, Ersoy R, Erden E, Akyol G, Bozkaya H, Ozenirler S. Effects of vitamin E treatment on peroxisome proliferator-activated receptor-alpha expression and insulin resistance in patients with non-alcoholic steatohepatitis: results of a pilot study. Intern Med J 2007; 37: 229-35.
[33] Adams LA, Sanderson S, Lindor KD, Angulo P. The histological course of nonalcoholic fatty liver disease: a longitudinal study of 103 patients with sequential liver biopsies. J Hepatol 2005; 42: 132-8.
[34] Miller ER 3rd, Pastor-Barriuso R, Dalal D, Riemersma RA, Appel LJ, Guallar E. Meta-analysis: high-dosage vitamin E supplementation may increase all-cause mortality. Ann Intern Med 2005; 142: 37-46.