Hepat Mon. 2020 July; 20(7):e102125. doi: 10.5812/hepatmon.102125.

Published online 2020 July 28. **Research Article**

Effect of *Beta vulgaris* Extract on Liver Enzymes in Patients with Non-Alcoholic Fatty Liver Disease: A Randomized Clinical Trial

Nikta Afzali [1], Seyed Shayan Ebadi [2], Hasan Afzali [3], Mohammad Reza Sharif [3], Mahdi Vazirian [1, 4], Seyed Alireza Ebadi [5, *], Vahid Shahkarami [6] and Habibollah Rahimi [7]

[1] Department of Pharmacognosy, Faculty of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran
[2] Student Research Committee, Shahid Beheshti University of Medical Sciences, Tehran, Iran
[3] Infectious Disease Research Center, Kashan University of Medical Sciences, Kashan, Iran
[4] Medicinal Plants Research Center, Faculty of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran
[5] Department of Internal Medicine, School of Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran
[6] Department of Radiology, Lorestan University of Medical Sciences, Khorramabad, Iran
[7] Department of Biostatistics and Epidemiology, School of Public Health, Kashan University of Medical Sciences, Kashan, Iran

[*] *Corresponding author*: : Department of Internal Medicine, School of Medicine, Shahid Beheshti University of Medical Sciences, SBUMS-Arabi Ave, Daneshjoo Blvd, Tehran, Iran. Tel: +98-9121231950, Email: sa.ebadi@sbmu.ac.ir

Received 2020 February 24; **Revised** 2020 May 16; **Accepted** 2020 July 09.

Abstract

Background: The prevalence of non-alcoholic fatty liver disease (NAFLD) has increased in recent decades. There are some concerns about the efficacy and side effects of drugs used for the treatment of NAFLD.
Objectives: Therefore, new treatment methods and modalities are needed. This study aimed to determine the efficacy of *Beta vulgaris* extract in the treatment of NAFLD.
Methods: This is a double-blind, parallel-group, randomized clinical trial. This clinical trial was conducted from November 2018 to April 2019 in Shahid Beheshti Hospital of Kashan, Iran. Among 143 NAFLD patients who met the inclusion criteria, 120 patients agreed to participate in the study. Subsequently, they were divided into two equal groups via simple randomization. *The Beta vulgaris* group received *Beta vulgaris* extract, alongside standard NAFLD treatment, including vitamin E and *Silybum marianum* extract (Livergol). The placebo group received standard NAFLD treatment, as well as a placebo instead of *Beta vulgaris* extract. The levels of aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP), fasting blood sugar (FBS), low-density lipoprotein (LDL), and high-density lipoprotein (HDL) were evaluated and compared between the groups. Variables were measured at the beginning of the study and after three and six months.
Results: Overall, 52% of the participants were male. The mean (SD) age of *Beta vulgaris* and placebo groups was 47.5 (10.5) and 46.4 (8.7) years, respectively. The results of between-group analysis revealed that AST significantly reduced in the *Beta vulgaris* group, compared to the placebo group ($P = 0.04$). Conversely, ALT reduction was not significant in the groups. The significant interaction between time and groups indicated that the effect of Beta vulgaris on ALT increased over time ($P < 0.001$). Moreover, the ALP, FBS, LDL, and HDL levels significantly improved in the *Beta vulgaris* group compared to the placebo group ($P < 0.05$).
Conclusions: Integration of *Beta vulgaris* extract in the standard treatment of NAFLD could significantly improve AST, ALP, FBS, LDL, and HDL. This study also revealed that the effect of *Beta vulgaris* on ALT increased over time.

Keywords: Diabetes Mellitus, Alanine Transaminase, Aspartate Transaminase, Non-Alcoholic, Fatty Liver Disease (NAFLD), *Beta vulgaris*, Beetroot

1. Background

Non-alcoholic fatty liver disease (NAFLD) is the most common cause of chronic liver disease around the world (1). The prevalence of NAFLD has increased in the past two decades, and the Middle East and South American countries account for the highest prevalence of NAFLD (2). NAFLD is also the second leading cause of liver transplant and the third leading cause of hepatocellular carcinoma (3, 4). The potential of NAFLD to progress to advanced fibrosis, cirrhosis, and hepatocellular carcinoma makes it a formidable disease with high morbidity and mortality. Moreover, NAFLD is highly associated with some metabolic comorbidities, including type II diabetes mellitus, hyperlipidemia, obesity, and metabolic syndrome (5, 6). Therefore, some effective treatment methods are needed to treat patients in the early stages of the disease and prevent its

progression into end-stage liver disease.

Currently, there is no standard pharmacological drug for the treatment of NAFLD. Management of some metabolic comorbidities, including obesity, type II diabetes mellitus, and hyperlipidemia, is the main goal of NAFLD treatment (7). Some agents, which are used for the treatment of NAFLD, include insulin-sensitizing agents (pioglitazone), hypolipidemic agents (gemfibrozil), and antioxidants (vitamin E) (8). However, these agents are not broadly recommended due to their adverse effects and unapproved effectiveness in NAFLD (9, 10). Furthermore, promising results have been reported in NAFLD treatment by using pentoxifylline and obeticholic acid. However, the safety and side effects of these agents are not well-established (11-13). Consequently, it seems necessary to find effective agents with minimum side effects.

Beta vulgaris is an herbal agent used as an anti-fever medication in ancient Roma (14). It has been shown that *Beta vulgaris* reduces blood sugar and induces glucose storage as glycogen in the liver (15). Experimental studies on rats revealed that *Beta vulgaris* reduces the level of liver enzymes, including aspartate transaminase (AST), alanine transaminase (ALT), and alkaline phosphatase (ALP) (16). Nevertheless, there have been some controversies about the effects of *Beta vulgaris* on liver enzymes in previous clinical trials. In order to obtain consistent results regarding the effectiveness of *Beta vulgaris* in NAFLD, further studies are needed.

2. Objectives

Therefore, the objective of this study was to determine the efficacy of *Beta vulgaris* extract in the treatment of NAFLD.

3. Methods

This study was approved by the Ethics Committee of Tehran University of Medical Sciences (TUMS) (code: IR.TUMS.PSRC.REC.1396.4044). It was also registered in the Iranian Registry of Clinical Trials (code: IRCT20121017011145N20). The identity and information of the participants remained confidential. Moreover, informed consent was obtained from eligible volunteers prior to participation in the study. All of the participants could withdraw from the study at any time.

3.1. Study Sample

This double-blind, parallel-group, randomized clinical trial was conducted from November 2018 to April 2019 at Shahid Beheshti Hospital of Kashan, Iran. This hospital is a referral teaching hospital, affiliated to Kashan University of Medical Sciences (KUMS).

3.2. Patients and Methods

This study was conducted among patients with NAFLD who were referred from specialized outpatient clinics of KUMS. The participants were selected by purposive sampling based on eligibility criteria. The inclusion criteria for patients were age between 18 and 70 years and a primary diagnosis of NAFLD. The diagnostic criteria for NAFLD included ultrasound evidence of fatty liver (grade II or above) and increased level of liver enzymes (two or three times higher than normal). Ultrasonography was performed by an expert radiologist, and the diagnosis was established by a gastroenterologist. The grade of fatty liver was determined according to the Rumack ultrasound criteria, and the liver biochemical profile was measured based on standard protocols (14).

On the other hand, patients with liver diseases, including Wilson's disease, hemochromatosis, alcoholic fatty liver disease, autoimmune liver disease, or cirrhosis, were excluded from the study. Also, pregnant and lactating women were excluded. Finally, eligible participants were divided equally into case and control groups via simple randomization. For the randomization, the groups were named as *Beta vulgaris* group and placebo group. Then, the sequence of groups was drawn up by coin tossing. The *Beta vulgaris* group received vitamin E pearl (300 IU/twice daily), Livergol tablet (140 mg/daily), and *Beta vulgaris* capsule (400 mg/daily) for six months. On the other hand, the placebo group received the same dosages of vitamin E pearl and Livergol tablet, besides placebo capsules instead of *Beta vulgaris* capsules for the same amount of time. Also, the participants were asked to do not use *Beta vulgaris* or its products during the study. The probable intervention complications were followed up closely via telephone contacts. In the first two weeks, three participants of the *Beta vulgaris* group had mild gastrointestinal symptoms, including diarrhea and nausea. Also, one participant of the placebo group had moderate diarrhea in the first week of intervention, who was excluded from the study.

The demographic and anthropometric characteristics of the participants, including gender, age, weight, height, family history of NAFLD, and history of diet and exercise, were determined at baseline. In addition, the levels of AST,

ALT, ALP, prothrombin time (PT), triglyceride (TG), cholesterol (CHL), high-density lipoprotein (HDL), low-density lipoprotein (LDL), fasting blood sugar (FBS), and albumin (ALB), as well as the grade of fatty liver, were measured at baseline and three and six months after the intervention.

3.3. Preparation of Beta vulgaris Extract

About 70 kg of *Beta vulgaris* root (common beet) was purchased and confirmed by a botanist. Next, the roots were cleaned and extracted according to the maceration technique using 70% ethanol by a specialist in the Medicinal Plants Laboratory of Tehran Faculty of Pharmacy, Tehran, Iran. The extracted liquid was concentrated by a rotary evaporator and dried by a spray dryer. Subsequently, 400 mg of the dried extract of *Beta vulgaris* root was added to a capsule with the same color and size as the placebo capsule. In addition, microbial experiments were carried out on *Beta vulgaris* extracts to find any possible contamination with pathogens, such as *Escherichia coli*, *Salmonella*, and *Bacillus cereus*.

3.4. Data Analysis

Chi-square test was used to evaluate qualitative variables. Independent samples *t*-test and one-way analysis of variance (ANOVA) were also used to determine differences between the mean values of the groups. The therapeutic effects of *Beta vulgaris* were evaluated using repeated measures analysis. Before analysis, the model preassumptions, including normality and sphericity, were examined using histograms, box plots, and Mauchly's tests. All the statistical analyses were conducted in SPSS version 23, and the level of significance was set at 0.05.

4. Results

Among the 143 patients who met the study criteria, 120 patients agreed to participate in the study (83.9% response rate). However, two patients from the placebo group left the study due to gastrointestinal side effects (e.g., vomiting and diarrhea). In addition, one patient from the placebo group was excluded from the analysis due to unusual values. The study flowchart is presented in Figure 1. Overall, 62 (52%) patients were male, and 55 (48%) patients were female. The mean (SD) age of the participants was 46.9 (9.7) years, ranging from 18 to 69 years. Other sociodemographic characteristics are shown in Table 1.

The mean levels of AST, as one of the main biomarkers of NAFLD, were 51.0 ± 20.9 and 55.5 ± 16.9 mg/dL in the *Beta vulgaris* and placebo groups, respectively. Also, the mean level of ALT was 61.7 ± 26.2 and 56.7 ± 12.9 mg/dL in the *Beta vulgaris* and placebo groups, respectively. There was no significant difference in the mean level of biomarkers between the groups at the beginning of the study. Other biochemical characteristics of the participants are presented in Table 2.

After 60 days of the intervention, the results of intra-group comparisons based on the repeated measures analysis indicated that the AST level decreased significantly over time ($F = 74.8$, $P < 0.001$). Also, intra-group comparisons based on repeated measures analysis showed that ALT significantly decreased during and at the end of the study ($F = 83.58$, $P < 0.001$). In other words, treatments were effective in both *Beta vulgaris* and placebo groups. In addition, the inter-group analysis revealed a significant reduction in the AST level in the *Beta vulgaris* group, compared to the placebo group ($F = 4.08$, $P = 0.04$). Contrary to AST, inter-group analysis of ALT showed no significant reduction over time ($F = 4.67$, $P = 0.94$). However, the inter-group analysis indicated a significant reduction in ALP ($F = 6.47$, $P = 0.01$), FBS ($F = 4.13$, $P = 0.04$), and LDL ($F = 6.43$, $P = 0.01$) and a significant increase in HDL ($F = 5.27$, $P = 0.02$) over six months. Other changes in biomarkers over time are shown in detail in Table 3.

Analysis of the interaction between time and groups showed a significant interaction regarding ALT ($F = 11.84$, $P < 0.001$). In other words, the effect of *Beta vulgaris* on ALT increased over time. However, there was no interaction between time and groups regarding AST. In other words, the effect of *Beta vulgaris* did not change over time. The trends of AST and ALT changes over time are presented in Figure 2.

5. Discussion

The present study showed that the use of *Beta vulgaris*, alongside the standard treatment of NAFLD, could have positive effects on the biochemical markers of patients with NAFLD. Integration of *Beta vulgaris* in the treatment regimen of NAFLD patients significantly decreased AST and ALP as the main biomarkers of hepatic disease, compared to the standard treatment. Since elevated AST is associated with higher grades of fibrosis among NAFLD patients (17), improvement of AST is a promising way to prevent the progression of liver fibrosis. Although *Beta vulgaris* extract could not have significant effects on ALT in this study, the interaction between time and groups revealed that the effect of *Beta vulgaris* on ALT increased over time. It is recommended that future studies evaluate the effect of *Beta vulgaris* on ALT for more than six months.

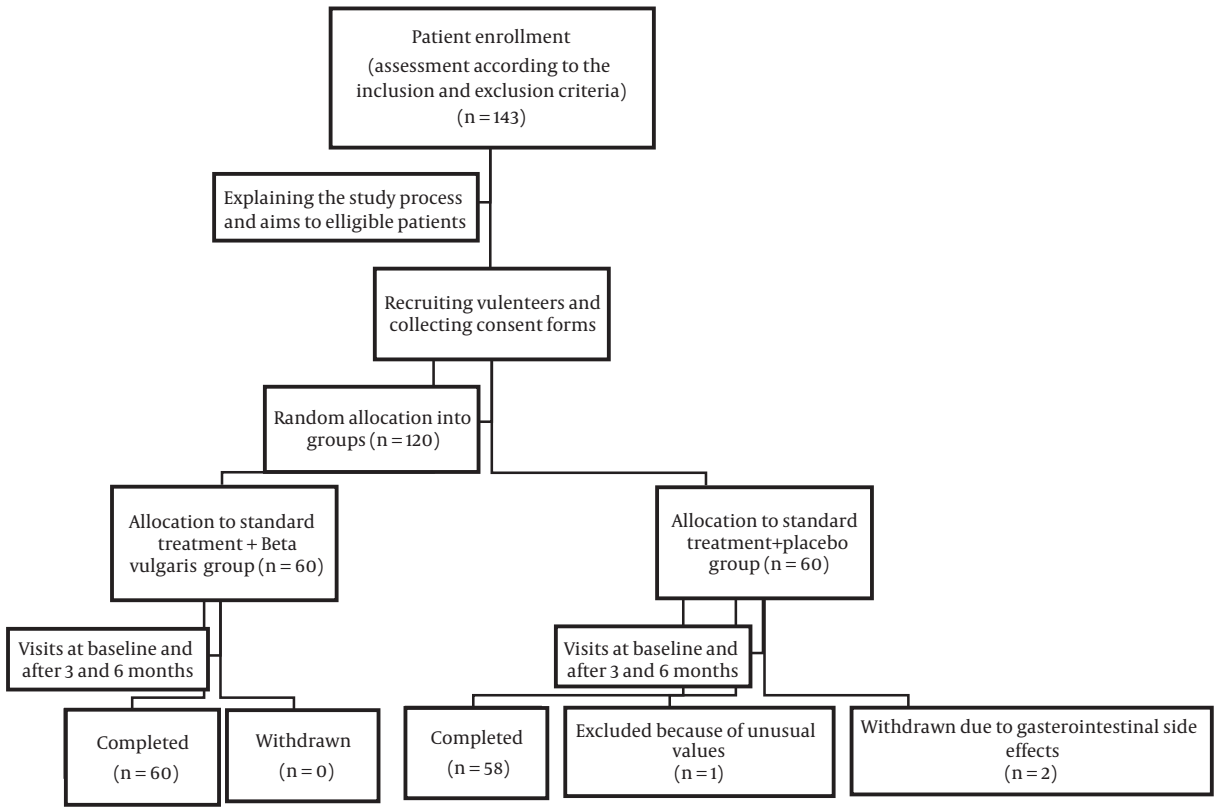

Figure 1. The study flowchart

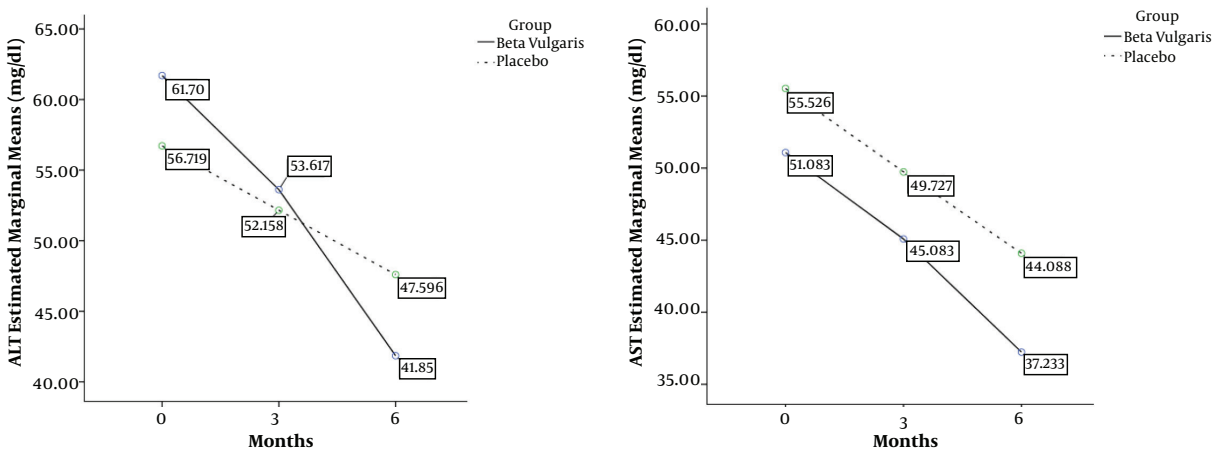

Figure 2. AST and ALT changes over time

Furthermore, *Beta vulgaris* extract had significant positive effects on other biomarkers, including FBS, LDL, and HDL. Since the treatment of comorbidities, such as diabetes mellitus, obesity, and hyperlipidemia, is one of the goals of NAFLD treatment, *Beta vulgaris* extract can improve the efficacy of other agents prescribed for NAFLD. To gain a clinical insight into the effect of *Beta vulgaris*, Cohen's d index was calculated using the Klauer's approach

Table 1. Descriptive Analysis of Demographic Characteristics and Other Variables in the Study Groups[a]

Variables	Study Groups		P Value
	Beta vulgaris Group (N = 60)	Placebo Group (N = 57)	
Age	47.5 ± 10.5	46.4 ± 8.7	0.55
BMI	30.4 ± 4.4	29.3 ± 4.4	0.18
Sex			0.65
Male	33 (53.2)	29 (46.7)	
Female	27 (49.0)	28 (50.9)	
Family history of NAFLD			0.63
No	29 (50.8)	28 (49.1)	
Yes	29 (50)	29 (50)	
Cirrhosis	2 (100)	0(0)	
Diet status			< 0.01[b]
Change from meat to vegetarian diet	0 (0)	0 (0)	
Change from vegetarian to meat diet	0 (0)	0 (0)	
On a diet in the last six months	10 (29.4)	24 (70.5)	
Losing weight in the last six months	3 (75.0)	1 (25.0)	
None	47 (59.4)	32 (40.5)	
Exercise status			0.71
Using bodybuilding devices	0 (0)	0 (0)	
Walking for 15 to 30 min every day in a week	9 (47.3)	10 (52.6)	
Walking for 15 to 30 min every other day in a week	29 (60.4)	19 (39.5)	
Walking for 15 to 30 min once a week	11 (42.3)	15 (57.6)	
Walking for 15 to 30 min once in two weeks	6 (46.1)	7 (53.8)	
Walking for 15 to 30 min once a month	1 (50.0)	1 (50.0)	
Without exercise	4 (44.4)	5 (55.5)	
BMI			0.19
Normal (18.5 - 24.9)	4 (33.3)	8 (66.7)	
Overweight (25 - 29.9)	26 (47.3)	29 (52.7)	
Obesity (≥ 30)	30 (60.0)	20 (40.0)	
Stage of NALD			0.03[b]
Grade 1	13 (48.1)	14 (51.9)	
Grade 1.5	2 (22.2)	7 (77.8)	
Grade 2	20 (43.5)	26 (56.5)	
Grade 2.5	11 (64.7)	6 (35.3)	
Grade 3	14 (77.8)	4 (22.2)	

[a] Values are expressed as mean ± SD or No. (%).
[b] P < 0.05.

(0.3 for AST and 0.6 for ALT). According to the Cohen's table, the effect size of *Beta vulgaris* was small for AST and medium for ALT. Therefore, despite the significant P value of AST, its effect size was small, while despite the non-significant P value of ALT, it was more significantly affected from a clinical point of view.

To the best of our knowledge, this is the second study evaluating the efficacy of *Beta vulgaris* extract in NAFLD patients. In the first study by Srivastava et al. (18), it was found that *Beta vulgaris* extract had no significant effects on the

Table 2. Biochemical Characteristics of the *Beta vulgaris* and Placebo Groups[a, b]

Variables	Study Groups		P Value
	Beta vulgaris Group (N = 60)	Placebo Group (N = 57)	
AST	51.0 ± 20.9	55.5 ± 16.9	0.21
ALT	61.7 ± 26.2	56.7 ± 12.9	0.19
ALP	199.9 ± 58.3	217.5 ± 43.9	0.06
FBS	92.4 ± 6.5	92.3 ± 8.3	0.06
PT	12.2 ± 0.69	12.0 ± 0.65	0.18
TG	191.0 ± 50.2	178.5 ± 23.9	0.09
CHL	192.7 ± 29.7	200.6 ± 22.2	0.11
LDL	115.1 ± 27.5	119.7 ± 21.7	0.31
HDL	40.9 ± 10.8	38.9 ± 4.9	0.20
ALB	4.4 ± 0.4	4.4 ± 0.4	0.88

[a] Values are expressed as mean ± SD.
[b] $P < 0.05$.

liver enzymes of NAFLD patients during 12 weeks. As shown in our study, the effect of *Beta vulgaris* on liver enzymes, especially ALT, increased over time. Therefore, the shorter duration of the study by Srivastava et al. compared to our study may be the cause of the discrepancy between the results. Also, considering the unclear dosage of *Beta vulgaris* supplement in the study by Srivastava et al. (18), insufficient dosage may be responsible for this discrepancy. Nevertheless, the lipid profile of patients in the *Beta vulgaris* group significantly decreased, compared to the placebo group. These results are consistent with our findings regarding the effect of *Beta vulgaris* extract on the lipid profile of patients with NAFLD.

In another study evaluating the hepatoprotective effects of *Beta vulgaris* on liver damage in male Sprague-Dawley rats, it was found that *Beta vulgaris* juice exerted hepatoprotective effects on liver damage in a dose-dependent manner (19). This finding supports our hypothesis about the discrepancy between our results and the study by Srivastava and colleagues. Moreover, Ozsoy-Sacan et al. (20) in their study, which assessed the effect of *Beta vulgaris* extract on the liver of diabetic rats, found that AST, ALT, ALP, total lipid profile, and blood glucose level decreased significantly in the intervention group, compared to the placebo group. Additionally, the anti-hyperglycemic and anti-lipidemic activities of *Beta vulgaris* have been confirmed in different studies (21-23). Therefore, the positive effects of *Beta vulgaris* extract on the glycemic status and lipid profile of patients with NAFLD can help physicians manage other comorbidities of NAFLD and improve the outcomes of treatment.

As a result, the integration of *Beta vulgaris* in the treatment regimen of NAFLD patients has positive effects on liver enzymes and other biochemical markers associated with NAFLD. Since *Beta vulgaris* is a highly available and low-cost medicinal plant, utilization of *Beta vulgaris* alongside other treatments of NAFLD can be considered as a new treatment with satisfactory clinical results. However, further studies are needed to evaluate the exact effects and possible side effects of *Beta vulgaris* on NAFLD.

Despite our findings, this study had two limitations. First, the FibroScan, as one the best noninvasive tests to quantify liver fibrosis, has not been used in this study due to economic considerations. Second, this study was performed for six months due to time limitations. According to our findings, it is recommended that future studies try to find the effect of *Beta vulgaris* consumption on NAFLD for more than six months.

5.1. Conclusions

The addition of *Beta vulgaris* extract to the standard treatment of NAFLD could significantly decrease the levels of AST and ALP. Although *Beta vulgaris* extract could not improve ALT, the interaction between time and groups showed that the effect of *Beta vulgaris* on ALT increased over time. Also, this study showed that the integration of *Beta vulgaris* extract, due to its positive effects on FBS, LDL, and HDL, could help physicians manage other metabolic comorbidities of NAFLS. It is recommended that future studies evaluate the possible side effects of *Beta vulgaris*. In addition, longer studies are needed in order to assess ALT changes over time.

Footnotes

Authors' Contribution: Study concept and design: NA, HA, MS, MV, SAE, and VS. Acquisition of data: NA, SSE, HA, MV, and VS. Analysis and interpretation of data: SSE, HR. Drafting of the manuscript: NA, SSE, HA, MS, SAE, and HR. Critical revision of the manuscript for important intellectual content: NA, SSE, HA, MS, MV, SAE, and VS. Statistical analysis: SSE, HR. Administrative, technical, and material support: NAHA, MS, MV, SSE, SAE, VS, and HR. Study supervision: SAE and HA.

Clinical Trial Registration Code: The clinical trial registration code is IRCT20121017011145N20 and link is https://en.irct.ir/trial/29809.

Conflict of Interests: There is no conflict of interest in this study.

Table 3. Descriptive Analysis of Biomarkers During the Study[a]

Variable	Study Groups						Between-Group Comparisons
	Beta vulgaris Group (N = 60)			Placebo Group (N = 57)			
	0th	3th	6th	0th	3th	6th	P Value
AST	51.0 ± 20.9	45.0 ± 16.0	37.2 ± 11.7	55.5 ± 16.9	49.7 ± 14.0	44.0 ± 11.4	0.04[b]
ALT	61.7 ± 26.2	53.6 ± 21.9	41.8 ± 15.0	56.7 ± 12.9	52.1 ± 13.5	47.5 ± 15.0	0.94
ALP	199.9 ± 58.3	198.1 ± 42.3	208.3 ± 38.4	217.5 ± 43.9	218.7 ± 36.9	225.3 ± 36.7	0.01[b]
FBS	92.4 ± 6.5	90.2 ± 4.6	87.0 ± 5.2	92.3 ± 8.3	92.0 ± 5.6	91.1 ± 5.7	0.04[b]
PT	12.2 ± 0.69	12.0 ± 0.4	12.0 ± 0.7	12.0 ± 0.65	12.0 ± 0.5	12.2 ± 0.6	0. 74
TG	191.0 ± 50.2	174.2 ± 38.2	159.5 ± 25.7	178.5 ± 23.9	173.4 ± 19.3	167.6 ± 16.0	0.71
CHL	192.7 ± 29.7	182.7 ± 27.2	174.4 ± 20.8	200.6 ± 22.2	190.5 ± 18.4	181.1 ± 17.9	0.05
LDL	115.1 ± 27.5	106.6 ± 21.7	95.9 ± 17.3	119.7 ± 21.7	114.8 ± 15.5	108.0 ± 14.3	0.01[b]
HDL	40.9 ± 10.8	45.8 ± 10.0	52.5 ± 11.3	38.9 ± 4.9	41.6 ± 5.6	48.4 ± 7.3	0.02[b]
ALB	4.4 ± 0.4	4.4 ± 0.4	4.4 ± 0.4	4.4 ± 0.4	4.3 ± 0.4	4.4 ± 0.4	0.55

[a]Values are expressed as mean ± SD.
[b]Significant difference at 0.05.

Ethical Approval: The ethical approval code is IR.TUMS.PSRC.REC.1396.4044 and link is https://en.irct.ir/trial/29809.

Funding/Support: This study was supported by a teaching and research scholarship from the Faculty of Pharmacy of Tehran University of Medical Sciences.

Informed Consent: Informed constant has been signed by the participants.

References

1. Chalasani N, Younossi Z, Lavine JE, Diehl AM, Brunt EM, Cusi K, et al. The diagnosis and management of non-alcoholic fatty liver disease: practice guideline by the American Gastroenterological Association, American Association for the Study of Liver Diseases, and American College of Gastroenterology. *Gastroenterology*. 2012;**142**(7):1592–609. doi: 10.1053/j.gastro.2012.04.001. [PubMed: 22656328].
2. Perumpail BJ, Khan MA, Yoo ER, Cholankeril G, Kim D, Ahmed A. Clinical epidemiology and disease burden of nonalcoholic fatty liver disease. *World J Gastroenterol*. 2017;**23**(47):8263–76. doi: 10.3748/wjg.v23.i47.8263. [PubMed: 29307986]. [PubMed Central: PMC5743497].
3. Wong RJ, Aguilar M, Cheung R, Perumpail RB, Harrison SA, Younossi ZM, et al. Nonalcoholic steatohepatitis is the second leading etiology of liver disease among adults awaiting liver transplantation in the United States. *Gastroenterology*. 2015;**148**(3):547–55. doi: 10.1053/j.gastro.2014.11.039. [PubMed: 25461851].
4. Younossi ZM, Otgonsuren M, Henry L, Venkatesan C, Mishra A, Erario M, et al. Association of nonalcoholic fatty liver disease (NAFLD) with hepatocellular carcinoma (HCC) in the United States from 2004 to 2009. *Hepatology*. 2015;**62**(6):1723–30. doi: 10.1002/hep.28123. [PubMed: 26274335].
5. Mansour-Ghanaei F, Joukar F, Najafi Mobaraki S, Mavaddati S, Hassanipour S, Sepehrimanesh M. Prevalence of non-alcoholic fatty liver disease in patients with diabetes mellitus, hyperlipidemia, obesity and polycystic ovary syndrome: A cross-sectional study in north of Iran. *Diabetes Metab Syndr Clin Res Rev*. 2019;**13**(2):1591–6.
6. Fattahi MR, Niknam R, Safarpour A, Sepehrimanesh M, Lotfi M. The Prevalence of Metabolic Syndrome In Non-alcoholic Fatty Liver Disease; A Population-Based Study. *Middle East J Dig Dis*. 2016;**8**(2):131–7. doi: 10.15171/mejdd.2016.18. [PubMed: 27252820]. [PubMed Central: PMC4885612].
7. Teli MR, James OF, Burt AD, Bennett MK, Day CP. The natural history of nonalcoholic fatty liver: A follow-up study. *Hepatology*. 1995;**22**(6):1714–9. [PubMed: 7489979].
8. Panahi Y, Ghamarchehreh ME, Beiraghdar F, Zare R, Jalalian HR, Sahebkar A. Investigation of the effects of Chlorella vulgaris supplementation in patients with non-alcoholic fatty liver disease: A randomized clinical trial. *Hepatogastroenterology*. 2012;**59**(119):2099–103. doi: 10.5754/hge10860. [PubMed: 23234816].
9. Lewis JD, Ferrara A, Peng T, Hedderson M, Bilker WB, Quesenberry CJ, et al. Risk of bladder cancer among diabetic patients treated with pioglitazone: Interim report of a longitudinal cohort study. *Diabetes Care*. 2011;**34**(4):916–22. doi: 10.2337/dc10-1068. [PubMed: 21447663]. [PubMed Central: PMC3064051].
10. Schurks M, Glynn RJ, Rist PM, Tzourio C, Kurth T. Effects of vitamin E on stroke subtypes: meta-analysis of randomised controlled trials. *BMJ*. 2010;**341**:c5702. doi: 10.1136/bmj.c5702. [PubMed: 21051774]. [PubMed Central: PMC2974412].
11. Zein CO, Yerian LM, Gogate P, Lopez R, Kirwan JP, Feldstein AE, et al. Pentoxifylline improves nonalcoholic steatohepatitis: A randomized placebo-controlled trial. *Hepatology*. 2011;**54**(5):1610–9. doi: 10.1002/hep.24544. [PubMed: 21748765]. [PubMed Central: PMC3205292].
12. Van Wagner LB, Koppe SW, Brunt EM, Gottstein J, Gardikiotes K, Green RM, et al. Pentoxifylline for the treatment of non-alcoholic steatohepatitis: A randomized controlled trial. *Ann Hepatol*. 2011;**10**(3):277–86. [PubMed: 21677329].
13. Rinella ME. Nonalcoholic fatty liver disease: A systematic review. *JAMA*. 2015;**313**(22):2263–73. doi: 10.1001/jama.2015.5370. [PubMed: 26057287].
14. Hamedi S, Honarvar M. Beta vulgaris - a mini review of traditional uses in Iran, phytochemistry and pharmacology. *Curr Drug Discov Technol*.

2019;**16**(1):74–81. doi: 10.2174/1570163815666180308142912. [PubMed: 29521241].
15. Morra V, De Bonis A, Grifa C, Langella A, Cavassa L, Piovesan R. Minero-petrographic study of cooking ware and pompeian red ware (Rosso Pompeiano) from Cuma (Southern Italy). *Archaeometry*. 2013;**55**(5):852–79. doi: 10.1111/j.1475-4754.2012.00710.x.
16. Rumack CM, Wilson SR, Charboneau JW, Levine D. *Diagnostic ultrasound*. New York: Elsevier/Mosby; 2011.
17. Alizadeh A, Mansour-Ghanaei F, Roozdar A, Joukar F, Sepehrimanesh M, Hojati SA, et al. Laboratory Tests, Liver Vessels Color Doppler Sonography, and FibroScan Findings in Patients with Nonalcoholic Fatty Liver Disease: An Observation Study. *J Clin Imaging Sci*. 2018;**8**:12. doi: 10.4103/jcis.JCIS_93_17. [PubMed: 29692949]. [PubMed Central: PMC5894278].
18. Srivastava S, Siddiqi Z, Singh T, Bala L. Beetroot supplementation on non - alcoholic fatty liver disease patients. *Curr Res Nutr Food Sci J*. 2019;**7**(1):96–101. doi: 10.12944/crnfsj.7.1.10.
19. Olumese FE, Oboh HA. Hepatoprotective effect of beetroot juice on liver injury in male Sprague–Dawley rats. *Ann Trop Pathol*. 2018;**9**(1):83. doi: 10.4103/atp.atp_34_17.
20. Ozsoy-Sacan O, Karabulut-Bulan O, Bolkent S, Yanardag R, Ozgey Y. Effects of chard (Beta vulgaris L. var cicla) on the liver of the diabetic rats: A morphological and biochemical study. *Biosci Biotechnol Biochem*. 2004;**68**(8):1640–8. doi: 10.1271/bbb.68.1640. [PubMed: 15322346].
21. Ul Kabir A, Samad MB, Ahmed A, Jahan MR, Akhter F, Tasnim J, et al. Aqueous fraction of Beta vulgaris ameliorates hyperglycemia in diabetic mice due to enhanced glucose stimulated insulin secretion, mediated by acetylcholine and GLP-1, and elevated glucose uptake via increased membrane bound GLUT4 transporters. *PLoS One*. 2015;**10**(2). e0116546. doi: 10.1371/journal.pone.0116546. [PubMed: 25647228]. [PubMed Central: PMC4315578].
22. Mirmiran P, Houshialsadat Z, Gaeini Z, Bahadoran Z, Azizi F. Functional properties of beetroot (Beta vulgaris) in management of cardio-metabolic diseases. *Nutr Metab (Lond)*. 2020;**17**:3. doi: 10.1186/s12986-019-0421-0. [PubMed: 31921325]. [PubMed Central: PMC6947971].
23. Babarykin D, Smirnova G, Markovs J, Vasiljeva S, Basova N, Simanis R, et al. Therapeutic effect of fractionated by ultrafiltration red beetroot (Beta vulgaris L.) juice in rats with food-induced fatty liver. *Euro J Biol Res*. 2019;**9**(1):1–9.