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ORIGINAL ARTICLE

Almond protects the liver in coronary artery disease: A randomized controlled clinical trial

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Abstract

Objective: To compare the effect of Pakistani and American almonds on serum concentration of liver enzymes in coronary artery disease patients.

Methods: The randomised controlled trial was conducted at the Cardiology Clinics of Aga Khan University Hospital, Karachi, from February to July, 2012, and comprised patients who were randomised into intervention PA and AA groups and the control NI groups. Subjects in the intervention groups were provided Pakistani and American varieties of almonds 10g/day respectively with instructions to soak them overnight, remove the skin and eat them before breakfast for 12 weeks. The control group underwent no intervention. Serum concentrations of aspartate transaminase, Alanine transaminase and gamma-glutamyl transferase were analysed and compared.

Results: Of the 150 subjects, 110(73.3%) completed the study. Of them, there were 38(34.5%) in PA group, 41(37.3%) in AA, and 31(28.2%) in the NI group. Dietary almonds significantly reduced serum concentrations of aspartate transaminase, alanine transaminase and gamma-glutamyl transferase in the two intervention groups compared to the controls group ($p<0.05$) at 12-week follow-up.

Conclusion: A low dose of almonds was found to be an effective strategy to protect the liver.

Keywords: Low dose, Soaked almonds, Transaminases, Transpeptidase, SGOT/SGPT. (JPMA 71: 791 2021)

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Introduction

Hepatic dysfunction is a common co-morbid condition of coronary artery disease (CAD). Although the precise cause and effect is still dubious, the crucial role of liver in regulating lipids and carbohydrate metabolism is not debatable. CAD patients experience insulin resistance (IR) and dyslipidaemia, with subsequent oxidative stress and chronic systemic inflammation. All these factors have independent deleterious effects on hepatocytes. The medications used for treating CAD or other co-morbidities further exacerbate the hepatic damage. Statins, for example, have well-recognised adverse effects on liver function.[1]

When the liver is under stress as in chronic CAD, the hepatocytes disintegrate, releasing aspartate aminotransferase (AST), alanine aminotransferase (ALT) and gamma-glutamyl transferase (GGT) into the blood. Interestingly, the implication of elevated serum level of these three biomarkers is not just limited to hepatic dysfunction, as they also imply cardio-metabolic abnormalities with sufficient evidence indicating that these hepatic biomarkers are also associated with cardiovascular disease (CVD) risk. Analysis of the British Women's Heart and Health Study and Meta-analysis involving around 3,000 women concluded that even minor increase in serum GGT by just 1U/L can increase the risk of CVD by 20%, and risk of CAD and stroke by 34%.[2] Another study involving more than 1,800 healthy men from the general population indicated GGT as a robust predictor of an acute coronary event independent of other CVD risk factors.[3] Moderately high GGT, still within normal range, is reported to increase the risk of an incident of CAD, and is an independent prognostic marker of re-infarction and cardiac death in CAD patients.[4]

Similar is the case for elevated aminotransferases, which are positively associated with severity of atherosclerosis and IR.[5] Results from the National Health and Nutrition Examination Survey showed that slight elevation of serum ALT concentrations, still within optimum range, is linked to higher CAD risk, and associated histological changes of fatty liver.[6] In contrast, AST is associated more with diabetic complications involving IR[7] and impaired glucose tolerance (IGT).[8]

Higher concentration of AST, ALT and GGT in the serum of CAD patients could indicate a concurrent or prospective non-alcoholic fatty liver disease, with dire health

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consequences. We need safer and inexpensive alternates, like nuts, which are good for cardio-metabolic health.[9] Almonds have shown to improve lipid profile,[10] glycaemic control,[11] and oxidative stress.[12] None of the animal studies looking at the hepatic effects of almonds have used whole or soaked almonds. Instead, they have used almond oils[13,14] and skin extracts.[15] To the best of our knowledge, there is no clinical study on hepato-protective effect of almonds except one[16] which has looked at AST and ALT in the context of weight reduction, but it used a very high dose of almonds 50 gm/daily and a hypo-caloric background diet, while limit itself to female subjects with high body-weights. Knowing that the cost of treatment is an important consideration when situation demands life-long use in chronic disorders, it was recently shown that a dose of almonds as low as 10g/day is effective in improving lipid profile and uric acid in CAD patients when used according to South-Asian tradition, taking them on empty stomach and after overnight soaking.[17,18] According to the Food and Agriculture Organisation of the United Nations, Pakistan ranks 17th on the list of almond-producing countries in the world.[19] Portugal ranks 19th and it has evaluated the biological properties of locally-grown almonds. India is not in the list of top 20 almond-growers,[19] and yet it has studied the nutritional properties of its almonds. To the best of our knowledge, there is no study exploring the medicinal properties of Pakistani almonds. The current study was planned to compare the effect of Pakistani and American almonds on serum concentration of liver enzymes in CAD patients.

Subjects and Methods

The randomised controlled trial (RCT) was conducted at the Cardiology Clinics of Aga Khan University Hospital, Karachi, from February to July, 2012. Approval was obtained from the institutional Clinical Trial Unit (CTU) and Ethical Review Committee (ERC), and the RCT was registered at the Australian New Zealand Clinical Trial Registry[20] (Trial number: ACTRN12614000036617; URL: http://www.anzctr.org.au). The study is an offshoot of an original research designed to inspect changes in serum high-density lipoprotein (HDL) levels as the primary outcome. The study design with eligibility criteria, participants' flow and sample size calculation, as well as the findings have already been published.[17,18]

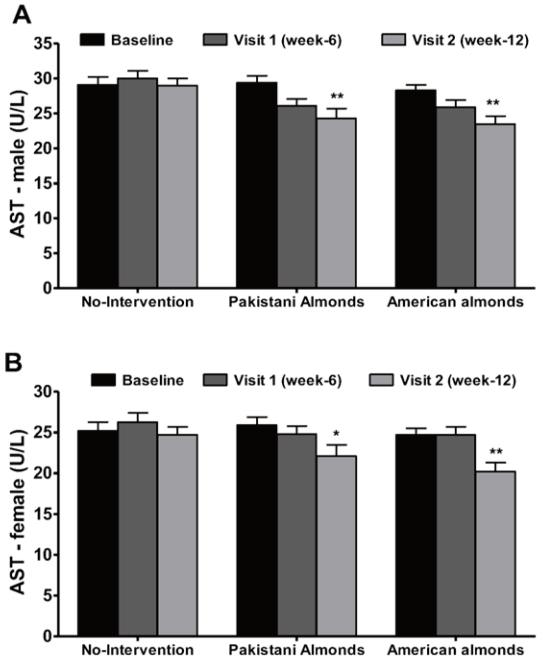
Those included were patients of CAD, as diagnosed by their respective cardiologists. After obtaining consent, baseline blood samples were collected, questionnaires were administered, and the participants were randomised into three groups using computer-generated block randomisation which was done using sealed envelopes provided by CTU. Group 1 was given no intervention (NI), Group 2 was given Pakistani almonds (PA) and Group-3 was given American almonds (AA).

Blood samples were taken at baseline and follow-up visits at week 6 and 12. Each time, vitals were recorded, and physical activity and food frequency questionnaires were administered. Pre-packed almonds 10g/d were provided to PA and AA groups. As per traditional recommendation, the participants were requested to soak the almonds overnight, remove the skin in the morning, and eat before breakfast. Compliance was monitored by patient diaries and reminder phone calls.

Serum concentrations of AST, ALT and GGT were estimated on an automated analyser (Roche Cobas c-111, PK) using commercially available kits. For comparing means, two-way analysis of variance (ANOVA) with repeated measure was used, followed by Bonferroni post-test. Results were expressed as means±standard error of mean (SEM), and $p<0.05$ was considered significant.

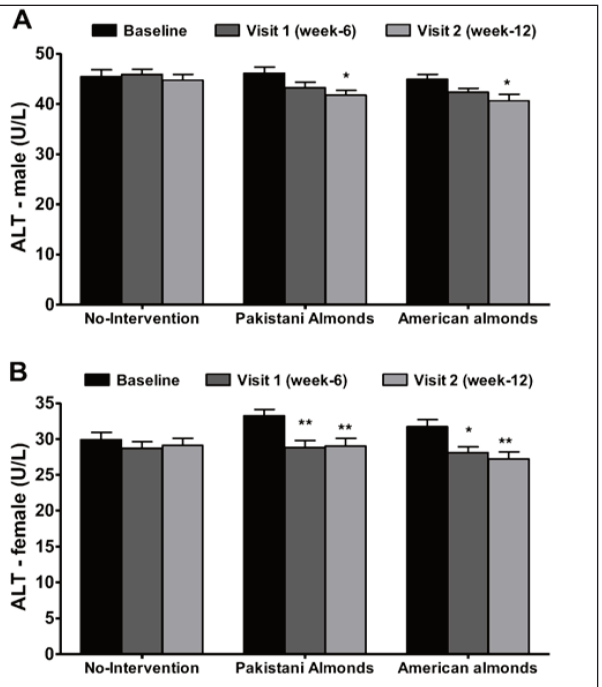
Results

Of the 150 subjects, 113 were males and 37 were females. The overall age range was 32-86 years, and mean body weight was 76±12kg. Of the total, 110 (73.3%) completed the study, and of them, 38 (34.5%) were in PA group, 41 (37.3%) in AA, and 31 (28.2%) in the NI group. Food



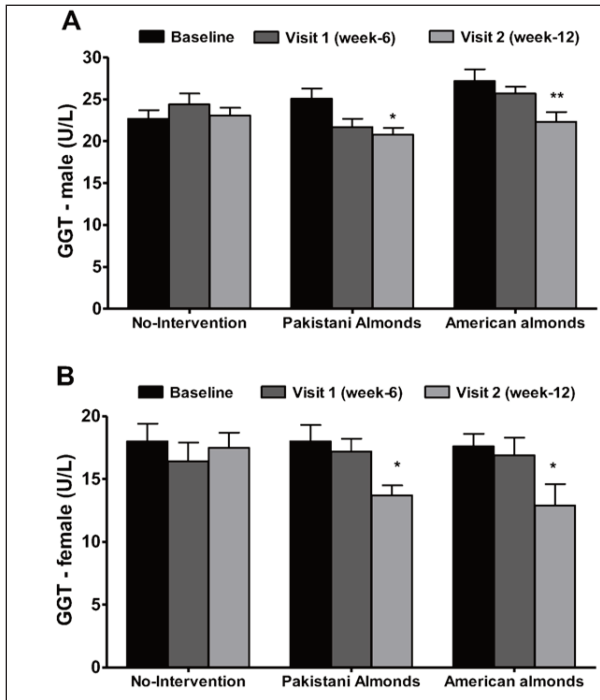
All values are expressed as mean±standard error of mean (SEM); *p-value < 0.05, ** p-value < 0.01

Figure-1: Impact of three-month almond supplementation on aspartate aminotransferase (AST) levels in male (A) and female (B) coronary artery disease (CAD) patients.



All values are expressed as mean±standard error of mean (SEM); *p-value < 0.05, ** p-value < 0.01

Figure-2: Impact of three-month almond supplementation on alanine aminotransferase (ALT) levels in male (A) and female (B) coronary artery disease (CAD) patients.



All values are expressed as mean±standard error of mean (SEM); *p-value < 0.05, ** p-value < 0.01

Figure-3: Impact of three-month almond supplementation on gamma-glutamyl transferase (GGT) levels in male (A) and female (B) coronary artery disease (CAD) patients.

consumption, physical activity patterns, and drug regimens remained unchanged across the groups during the study (p>0.05). PA and AA subjects showed significant improvement in serum concentrations of AST, ALT and GGT at 12 weeks compared to NI controls (p<0.05), as well as compared to their own respective baseline values (p<0.05). At 6 weeks, the values were not significant (p>0.05) Dietary supplementation of both Pakistani and American almonds produced similar effects (p>0.05).

Mean serum AST concentration in PA and AA groups decreased from 27.65±1 and 26.75 ±1.1 U/L at baseline to 25.45±1.1 and 25.3±1 U/L (p>0.05) at 6 weeks, and 23.2±1.1 and 21.85±1U/L (p<0.05) at 12 weeks. In the NI group, the concentrations were 27.2±0.9U/L at baseline, 28.2±1.1 at week 6 and 26.9+1U/L at week 12 (p>0.05) (Figure 1). ALT concentration in NI was 37.7±1.2U/L at baseline, 37.3±1.1 at week 6 and 36.9±1U/L at week 12 (p>0.05. ALT concentration dropped from 39.65±1.33 to 36+0.8 and then to 35.35±0.95 U/L (p< 0.05) in PA group, and from 38.3±1.1 to 35.2±0.7 and then to 33.9±0.8 U/L (p<0.05) in AA group (Figure 2).

Serum GGT concentrations at baseline were 20.35±1.2, 21.55±0.8 and 22.4±1.0U/L in NI, PA and AA groups, respectively. At week 6, the concentrations changed to 19.45±1.1 and 21.3±1 U/L in PA and AA respectively (p>0.05). The change in NI group was not significant (p>0.05). At week 12, GGT concentrations in the NI group remained unchanged at 20.3±1.6U/L (p>0.05), while in PA and AA groups it decreased to 17.25±1 and 17.6±0.9U/L (p<0.05) (Figure 3).

There was no significant change in systolic or diastolic blood pressure, heart rate or body-weights of the participants in any of the three groups (p>0.05).

Discussion

The findings showed a reduction in the concentration of AST, ALT and GGT in the serum of CAD patients, indicating the potential of almonds to protect the liver from damage and dysfunction. To the best of our knowledge, this is the first clinical trial on almonds, showing protection of CAD-associated hepatic complications at a very low dose when taken on empty stomach after overnight soaking. This is also the first attempt to scientifically evaluate Pakistani almond variety and show that the bio-efficacy of this cultivar is similar to American almonds. We need full-scale clinical trials and molecular research to find out exactly how whole almonds may prevent, and possibly treat, human hepatic dysfunction, but these preliminary results are significant on their own.

Hepatic effects of other components of almonds, like

oils[13,14] or skin extracts,[15] have also been tested in animal models. An earlier study[15] found that certain extracts of almond skin can protect hepatocytes from oxidative stress. They elaborated the anti-oxidative potential using cell and tissue preparations and particularised microsomal lipid peroxidation and cell death.[15] One study[13] assessed the hepatic effects of almond oil in a rat model of acute liver damage, and found reduction in AST and ALT, with a concurrent lowering of cholesterol, triglycerides, and lipoproteins in the serum. They also reported an anti-oxidative effect in superoxide dismutase (SOD), glutathione peroxidase (GSH) and malondialdehyde (MDA) assays.[13] A similar reduction in hepatic biomarkers, oxidative stress and lipid profile by almond oil was reported by a study[14] which used carbon tetrachloride (CCL4)-induced hepatic toxicity in rats. It found that a formulation containing almonds, flaxseeds and olive oil significantly reduced serum AST, ALT, cholesterol, triglycerides, MDA and SOD. In contrast, a study[21] used extracts of bitter almonds in the rat model of streptozotocin-induced diabetes. The extracts positively affected some of the liver fatty acids, inhibited post-prandial glycaemia and showed an antioxidant effect in the MDA and GSH assays.[22]

It has previously been shown in a rat model that dietary almond supplementation prevented high-fat diet-induced hepatic damage and potentially inhibited cholesterol synthesis via HMG-CoA reductase inactivation.[23] It was further observed that high-fat diet led to prominent fatty lesions on rat livers,[24] which were absent in the almond-treated rats (unpublished data). This encouraged us to explore whether dietary almonds can prevent fatty liver or associated complications in chronic diseases.

The only human study using almond intervention and liver enzymes[16] used a large dose of almond at 50 grams/day added to low-calorie diets of obese and overweight women. After three months, body-weights reduced with significant improvement in liver enzymes.[16] Conflictingly, we did not find a change in body-weight, which may be because of the hypocaloric versus habitual diets. Interestingly, using an almond dose that was five times less than that used earlier,[16] the current study observed a comparable reduction in liver enzymes. This means that soaking almonds overnight and eating before breakfast may have added benefits with better bioavailability. Previously, in rodent models, it was found that a low dose of almonds eaten on empty stomach is equally effective as high dose taken with food.[22,25,26] Digestion, absorption and content bio-accessibility is maximised when there are no hindering food particles.[26]

Raised hepatic biomarkers AST, ALT and GGT in the serum are not just interpreted as indicating hepatic destruction/dysfunction, but their implication in CVD prognosis is now being recognised by the scientific community, and research is directed towards understanding their role in the pathogenesis of atherosclerotic vascular diseases. While the precise mechanistic contributions of these biomarkers in CAD are still uncertain, it is known that hepatic damage compromises the physiological state and limits treatment options for CAD patients.

GGT is not confined only to hepatocytes. It is found on membranes of other cell types too where it is anticipated to be involved in atherosclerotic progression.[27] Active GGT has been identified in plaques that block the coronary arteries in CAD patients where it is shown to be producing reactive oxygen species which damages deoxyribonucleic acid (DNA), proteins, and circulating lipids.[28] Oxidised lipids in turn promote the formation of foam cell in the atheromatous plaques.

AST is also not specific to the liver cells, as it it is present in the heart, kidneys and blood cells among others. It may get released if any of these organs experience cellular damage due to oxidative stress or inflammation. ALT, however, is highly restricted to the cytoplasm of liver cells and elevated levels mostly indicate hepatocellular destruction.[24]

Conclusion

Almonds were found to potentially protect the liver of CAD patients at distinctly low dose of 10 g/day when taken on an empty stomach after overnight soaking. Knowing that natural edible products are safer than medications and act through multiple target sites, they can be promoted as preventive remedy to avert hepatic dysfunction associated with chronic diseases, like CVDs, and cost-effectiveness can be assured when taken in a South Asian traditional way.

Disclaimer: The original trial with high-density lipoprotein (HDL) as the primary outcome was part of a doctorate thesis. The results presented here were not included in the dissertation, as these analyses were conducted later.

Conflict of Interest: None.

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References

1. Golomb BA, Evans MA. Statin adverse effects: a review of the literature and evidence for a mitochondrial mechanism. Am J Cardiovasc Drugs 2008;8:373-418. doi: 10.2165/0129784-200808060-00004.
2. Fraser A, Harris R, Sattar N, Ebrahim S, Smith GD, Lawlor DA. Gamma-glutamyltransferase is associated with incident vascular events independently of alcohol intake: analysis of the British Women's Heart and Health Study and Meta-Analysis. Arterioscler Thromb Vasc Biol 2007;27:2729-35. doi: 10.1161/ATVBAHA.107.152298.

3. Meisinger C, Döring A, Schneider A, Löwel H. Serum gamma-glutamyltransferase is a predictor of incident coronary events in apparently healthy men from the general population. Atherosclerosis 2006;189:297-302. doi: 10.1016/j.atherosclerosis.2006.01.010.
4. Mao Y, Qi X, Xu W, Song H, Xu M, Ma W, et al. Serum gamma-glutamyl transferase: a novel biomarker for coronary artery disease. Med Sci Monit 2014;20:706-10. doi: 10.12659/MSM.890245.
5. Dessein PH, Woodiwiss AJ, Joffe BI, Norton GR. Aminotransferases are associated with insulin resistance and atherosclerosis in rheumatoid arthritis. BMC Cardiovasc Disord 2007;7:31. doi: 10.1186/1471-2261-7-31.
6. Ioannou GN, Weiss NS, Boyko EJ, Mozaffarian D, Lee SP. Elevated serum alanine aminotransferase activity and calculated risk of coronary heart disease in the United States. Hepatology 2006;43:1145-51. doi: 10.1002/hep.21171.
7. Schindhelm RK, Diamant M, Bakker SJ, van Dijk RA, Scheffer PG, Teerlink T, et al. Liver alanine aminotransferase, insulin resistance and endothelial dysfunction in normotriglyceridaemic subjects with type 2 diabetes mellitus. Eur J Clin Invest 2005;35:369-74. doi: 10.1111/j.1365-2362.2005.01502.x.
8. Oka R, Aizawa T, Yagi K, Hayashi K, Kawashiri M, Yamagishi M. Elevated liver enzymes are related to progression to impaired glucose tolerance in Japanese men. Diabet Med 2014;31:552-8. doi: 10.1111/dme.12345.
9. Ternus M, McMahon K, Lapsley K, Johnson G. Qualified health claim for nuts and heart disease prevention: development of consumer-friendly language. Nutr Today 2006;41:62-6.
10. Musa-Veloso K, Paulionis L, Poon T, Lee HY. The effects of almond consumption on fasting blood lipid levels: a systematic review and meta-analysis of randomised controlled trials. J Nutr Sci 2016;5:e34. doi: 10.1017/jns.2016.19.
11. Chen CM, Liu JF, Li SC, Huang CL, Hsirh AT, Weng SF, et al. Almonds ameliorate glycemic control in Chinese patients with better controlled type 2 diabetes: a randomized, crossover, controlled feeding trial. Nutr Metab (Lond) 2017;14:51. doi: 10.1186/s12986-017-0205-3.
12. Choudhury K, Clark J, Griffiths HR. An almond-enriched diet increases plasma α-tocopherol and improves vascular function but does not affect oxidative stress markers or lipid levels. Free Radic Res 2014;48:599-606. doi: 10.3109/10715762.2014.896458.
13. Jia XY, Zhang QA, Zhang ZQ, Wang Y, Yuan JF, Wang HY, et al. Hepatoprotective effects of almond oil against carbon tetrachloride induced liver injury in rats. Food Chem 2011;125:673-8. doi.org/10.1016/j.foodchem.2010.09.062
14. Fadlallah A, Saleh Abd Elal Z, Fadlalla E. Protective effect of olive, almond and flaxseed oil against carbon tetrachloride-induced hepatotoxicity in rat models. African J Biol Sci 2013;6:243-59.
15. Dong Q, Banaich MS, O'Brien PJ. Cytoprotection by almond skin extracts or catechins of hepatocyte cytotoxicity induced by hydroperoxide (oxidative stress model) versus glyoxal or methylglyoxal (carbonylation model). Chem Biol Interact 2010;185:101-9. doi: 10.1016/j.cbi.2010.03.003.
16. Abazarfard Z, Eslamian G, Salehi M, Keshavarzi S. A Randomized Controlled Trial of the Effects of an Almond-enriched, Hypocaloric Diet on Liver Function Tests in Overweight/Obese Women. Iran Red Crescent Med J 2016;18:e23628. doi: 10.5812/ircmj.23628.
17. Jamshed H, Sultan FA, Iqbal R, Gilani AH. Dietary Almonds Increase Serum HDL Cholesterol in Coronary Artery Disease Patients in a Randomized Controlled Trial. J Nutr 2015;145:2287-92. doi: 10.3945/jn.114.207944.
18. Jamshed H, Gilani AU, Sultan FA, Amin F, Arslan J, Ghani S, Masroor M. Almond supplementation reduces serum uric acid in coronary artery disease patients: a randomized controlled trial. Nutr J 2016;15:77. doi: 10.1186/s12937-016-0195-4.
19. Food and Agriculture Organization of the United Nations. Countries by commodity – Almonds, with shell. [Online] 2018 [Cited 2020 July 01]. Available from URL: http://www.fao.org/faostat/en/#rankings/countries_by_commodity
20. Australian New Zealand Clinical Trial Registry. Trial # ACTRN12614000036617. [Online] [Cited 2020 July 01]. Available from URL: http://www.anzctr.org.au
21. Demir E, Yilmaz O. The Effect of Bitter Almond Oil on The Some Alterations in Liver Tissue of Experimental Diabetic Rats. Van Vet J 2015;26:17-23.
22. Arslan J, Ahmed T, Gilani AH. Soaked Almonds Exhibit Vitamin E-dependent Memory Protective Effect in Rodent Models. Int J Pharmacol 2017;13:448-56.
23. Jamshed H, Gilani AH. Almonds inhibit dyslipidemia and vascular dysfunction in rats through multiple pathways. J Nutr 2014;144:1768-74. doi: 10.3945/jn.114.198721.
24. Jamshed H, Arslan J, Gilani AU. Cholesterol-cholate-butterfat diet offers multi-organ dysfunction in rats. Lipids Health Dis 2014;13:194. doi: 10.1186/1476-511X-13-194.
25. Jamshed H, Gilani AH. Lower Dose of Almonds Exhibits Vasculo-protective Effect when Given in Empty Stomach. Int J Pharmacol 2015;11:122-9.
26. Mandalari G, Faulks RM, Rich GT, Lo Turco V, Picout DR, Lo Curto RB, et al. Release of protein, lipid, and vitamin E from almond seeds during digestion. J Agric Food Chem 2008;56:3409-16. doi: 10.1021/jf073393v.
27. Bradley R. Gamma glutamyltransferase (GGT) as a biomarkers of atherosclerosis. In: Patel VB, Preedy VR, eds. Biomarkers in cardiovascular disease. Dordrecht, Netherlands: Springer, 2016; pp 673-702.
28. Emdin M, Pompella A, Paolicchi A. Gamma-glutamyltransferase, atherosclerosis, and cardiovascular disease: triggering oxidative stress within the plaque. Circulation 2005;112:2078-80. doi: 10.1161/CIRCULATIONAHA.105.571919.