EFFECT OF NUTS (PISTACHIO OR ALMONDS) CONSUMPTION ON LIPID PROFILE OF HYPERCHOLESTEROLEMIC RATS

GHADA Z A SOLIMAN
Assistant Professor of Biochemistry, National Nutrition Institute, Cairo, Egypt, Email: r.soliman2005@yahoo.com

Received: 23 July 2012, Revised and Accepted: 31 August 2012

ABSTRACT

Introduction: Nuts contain numerous beneficial nutritive and bioactive compounds like fatty acids, dietary fibbers, micronutrients, and phytochemicals that have shown favourable lipid-altering activity.

Aim: To study the effects of nut (Pistachio or Almonds) consumption on lipid profiles of hypercholesterolemic rats.

Materials and Methods: One hundred and eighty (180) adult male Sprague-Dawley rats, divided into 9 groups (20/group): G1: control; G2: hypercholesterolemic; G3-G6: hypercholesterolemic treated with hypocholesterolemic drug, pistachio, almonds, mixture of pistachio and almonds, G7-G9 normal rats treated with pistachio (G7), almonds (G8), mixture of pistachio and almonds (G9). The experiment lasted 6 weeks after treatment. Rats were sacrificed; blood was collected for biochemical analysis.

Results and Discussion: The results of this study showed that diet supplemented with pistachio and/or almond induced significant decrease in TC and LDL-C, VLDL TG and Phospholipids level, while HDL-C levels were unchanged compared to hypercholesterolemic group. The decrease being more in rats treated with mixture of both nuts. The overall result, therefore, is toward a less atherogenic lipid profile. The same was observed for MDA. This may be due to interactive or additive effects of the numerous bioactive constituents found in pistachio or almonds. Nuts are rich in several beneficial compounds, as ω-6 or ω-9 or ω-3 fatty acids which have demonstrated beneficial effects on blood cholesterol and lipoprotein profiles.

Conclusion: Results suggest that pistachio or almonds supplementation may improve blood lipids, ameliorate oxidative stress and this may be due to interactive or additive effects of the numerous bioactive constituents found in pistachio or almonds. Nuts may have beneficial applications in the prevention of cardiovascular diseases.

Keywords: Nuts, Fatty acids, CHD, CVD, Lipid profile, phytochemicals, dietary fibre, cholesterol.

INTRODUCTION

The World Health Organization (WHO) estimates that every year 12 million people worldwide die from cardiovascular diseases, with most of them being from the developing world (Kmietowicz, 2002). Nuts are recently recognized as “heart-healthy” foods by the U.S. Food and Drug Administration (US-FDA 2003, Kocyigit et al., 2006).

Because nuts have favourable fatty acid and nutrient profiles, there is growing interest in evaluating their role in cholesterol-lowering diets. Nuts are complex plant foods that are not only rich sources of unsaturated fat (nuts are low in saturated fatty acids and high in monounsaturated and polyunsaturated fatty acids) but also contain several non-fat constituents such as plant protein, fibre, micronutrients (e.g.: copper and magnesium), vitamins as A, C, E, plant sterols, and phytochemicals that may provide additional protective effects (Kris-Etherton et al., 1999 & 2001). These nutrients have shown favourable lipid-altering activity (Vorster et al., 2003). Nuts such as almonds and pistachios are rich in several beneficial compounds, such as ω-9 fatty acids, which have demonstrated beneficial effects on blood cholesterol and lipoprotein profiles (Brown 2003). It has been proposed that the bioactive compounds in nuts may help lower the risk factors of CVD by improving endothelial function (Ros 2009), blood pressure (Estruch et al., 2006) and the serum lipid profile (Giel & Kris-Etherton 2006) in addition to lowering oxidative stress (Jenkins et al., 2006 & 2002) and inflammation (Jiang 2006).

There are controversial results about the effects of pistachio nut and/or almonds consumption on the lipid profile of patients with hypercholesterolemia. There is limited information about the antioxidant effects of nuts and pistachios (Matthäus and Ozcan, 2006).

MATERIAL AND METHODS

Approval of the experimental protocol had been taken from the research ethics committee of General Organization of Teaching Hospitals & Institutes (GOTHI), Cairo, Egypt.

Animals and diet

Pistachio and almonds were purchased from local markets of Cairo, Egypt. They were analysed for their nutritive value according to AOAC (2010). They were analysed for their fatty acid profile using GC mass.

Fatty acid Profile

a) Lipid extraction was conducted using method of AOAC 2000. Separation of fatty acids (saponification, preparation of diazomethane, then methylation) was carried out using method of Vogel 1975.

b) Identification and determination of fatty acids was conducted using gas liquid chromatography (GLC, GC trace GC ULTRA) according to Farag et al., 1986. The gas chromatographic analysis was performed on a GC trace GC ULTRA equipped with an FID fitted with a column (30 m) packed with 70% cyanopropyl polysilphylene siloxane. The carrier gas was N2, with a flow rate of 1.5 ml/min. The column was run isothermally at 195 °C and the injector and detector were at 220 °C. The fatty acids were identified by the retention time by comparing with standards. Peak area was measured by using a computing integrator (PU-4810, Philips).

Experimental Design

One hundred and eighty (180) adult male Sprague-Dawley rats weighing 210±20 g, 3 months old were housed individually in stainless steel mesh cages. They were fed on standard diet for 10 days before experiments began (Adaptation period). The control diet was prepared according to Reeves et al., (1993), and National Research Council (NRC) Committee on Animal Nutrition, (1978). The diets were prepared every week in the laboratory. The water and diets were given ad libitum. Induction of hypercholesterolemia was carried out on 6 groups (G2-6; 100) by addition of cholesterol (2 %) + 0.25 % bile salts to the basal diet for 4 weeks. The animals (180) were divided into 9 groups (20 rat/group) as follows:

Group 1 (Control group, -ve control); rats fed on basal diets; Group 2: Hypercholesterolemic rats, (-ve control); Group 3: Hypercholesterolemic rats fed on basal diet supplemented with hypocholesterolemic drugs (Statin); Group 4: Hypercholesterolemic rats fed on basal diet supplemented with pistachio 2%; Group 5: Hypercholesterolemic rats fed on basal diet supplemented with almonds 2%; Group 6: Hypercholesterolemic rats fed on basal diet supplemented with pistachio and almonds, Group 7: Hypercholesterolemic treated with hypocholesterolemic drug, pistachio, almonds, mixture of pistachio and almonds, Group 8: Hypercholesterolemic treated with pistachio, almonds, mixture of pistachio and almonds, Group 9: Hypercholesterolemic treated with pistachio, almonds, mixture of pistachio and almonds, Group 2: Hypercholesterolemic rats treated with hypocholesterolemic drug, pistachio, almonds, mixture of pistachio and almonds, Group 3: Hypercholesterolemic rats treated with pistachio, almonds, mixture of pistachio and almonds.
supplemented with equal mixture of pistachio and almonds; Group 7: normal rats fed on basal diet supplemented with pistachio 2%; Group 8: normal rats fed on basal diet supplemented with almonds 2%; Group 9: normal rats fed on basal diet supplemented with equal mixture of pistachio and almonds. Diets and rats weight were done once/week.

At the end of the experimental period (6 weeks after treatment), rats were fasted over night before sacrificing, blood was collected, centrifuged; serum was stored at - 80 °C until analysis. Part of the blood is collected on tubes coated with EDTA. Some minerals as Mg, Cu, Fe etc... and some vitamins as A, C and E were determined in nuts.

Analytical Methods

The serum total cholesterol (TC) and serum high density lipoprotein cholesterol (HDL-C) level was determined using colorimetric enzymatic kits (SGM Italia, Rome, Italy), according to the method described by Allian et al., (1974) and Lopes-Virella et al., (1977) respectively. The serum low density lipoprotein cholesterol (LDL-C) level was determined using colorimetric enzymatic kits (SGM Italia, Rome, Italy), according to the method described by Fruchart et al., (1982) and Levy et al., (1981). The serum triglycerides (TG) level was determined using colorimetric enzymatic kits (SGM Italia, Rome, Italy), according to the method described by Bucolo et al., (1973). Very low density lipoprotein cholesterol (VLDL-C) level was calculated using the following equation: VLDL-C= TC-(HDL-C+LDL-

The plasma maloniedialdehyde (MDA) level was determined according to the method described by Draper and Hadley (1990). Amino acids (arginine and lysine) were determined using amino acid analyzer, AAA, Sykum using its instruction cited in the catalogue. Minerals were determined using atomic absorption according to AOAC 2010. Vitamins were determined using HPLC according to AOAC 2010.

Statistical Analysis

All results were expressed as the mean ± SE. Statistical analysis was performed with Statistical Package for the Social Science for Windows (SPSS, version 11.0, Chicago, IL, USA). The data were analyzed by one-way analysis of variance (ANOVA). To compare the difference among the groups, post hoc testing was performed by the Tukey test. Pearson’s correlation analysis was used to determine the correlation among the parameters assessed. The p-value < 0.05 was considered statistically significant (Dawson and Trapp, 2001).

RESULTS

Table (I-a) reveal nutritive value, mineral and vitamin content of pistachio and almonds nuts. The kernels are a rich source of oil (>50.5%). It contains protein (>18.6), carbohydrate (> 16.87%) and dietary fibre (>10.0%). Pistachio nut also contains high amounts of K and P, and various amounts of Ca, Mg and Fe. The caloric value of the pistachio nuts and almonds was 633.33, 613.3 Kcal/100 gm respectively of the edible parts which is in agreement with Breuer 1993, Food Composition Tables for Egypt 2006.

Table 1-a: Chemical analysis (nutritive value) of pistachio and almonds/100gm.

<table>
<thead>
<tr>
<th>Protein</th>
<th>As</th>
<th>Crude fibre gm/100 gm</th>
<th>Moisture %</th>
<th>Carbohydrate</th>
<th>Dietary fibre</th>
<th>Calories Kcal/100 gm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pistachio</td>
<td>20.71</td>
<td>2.1</td>
<td>4</td>
<td>1.43</td>
<td>5.18</td>
<td>16.87</td>
</tr>
<tr>
<td>Almonds</td>
<td>18.6</td>
<td>2.7</td>
<td>2.5</td>
<td>4</td>
<td>4.6</td>
<td>21.1</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Minerals (mg/100gm)</th>
<th>Vitamins</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca</td>
<td>Mg</td>
</tr>
<tr>
<td>107.1</td>
<td>3.9</td>
</tr>
<tr>
<td>4</td>
<td>3</td>
</tr>
<tr>
<td>215</td>
<td>18</td>
</tr>
</tbody>
</table>

ND: Not detected

Table (I-b) reveal fat content and amount of mono-, polyunsaturated and saturated fatty acids of pistachio nuts and almonds. Also it shows their fatty acid composition. Pistachio nuts and almonds are highly nutritious foods, its fat content as high as 53.67; 50.5 %, Table (I-b) reveals that the mean fatty acid composition of the pistachio and almonds is 60.51, 65.41% oleic acid; 27.69, 17.42% linoleic acid, 10% palmitic acid which agrees to somewhat with (Gamyi and Hayoglu 2007). It also reveals that pistachio and almonds are rich in unsaturated fatty acids which represent 88.71, 83.7% of fat respectively.

Table 1-b: Fat content and % MUSFA, PUSFA, SFA, fatty acid composition and arginine & lysine content of pistachio nuts & almonds

<table>
<thead>
<tr>
<th>Fat (gm/100 gm)</th>
<th>Relative % of fat</th>
<th>Amino acid</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total</td>
<td>MUSFA</td>
<td>PUSFA</td>
</tr>
<tr>
<td>Pistachio</td>
<td>53.67</td>
<td>32.75</td>
</tr>
<tr>
<td>Almonds</td>
<td>50.5</td>
<td>33.47</td>
</tr>
</tbody>
</table>

MUSFA: Monounsaturated Fatty Acids; PUSFA: Polyunsaturated Fatty Acids; SFA: Saturated Fatty Acids. ND: Not detected

Table (II) reveals that hypercholesterolemic rats showed significantly decreased body weight when compared with normal control group. At the end of the experiment, the mean levels of body weight of G3-G6 rats were significantly lower than control group but significantly higher than hypercholesterolemic rats but in control+pistachio, control + almonds, control+mixture groups (G7-G9), it shows no significant change in body weight compared to normal control group, but it show significantly higher body weight when compared with hypercholesterolemic rats or treated groups (G3-G6).

Table (III) reveals that cholesterol, LDL-C, VLDL-C, triacylglycerol, phospholipids and maloniedialdehyde levels were significantly higher (P< 0.001) in hypercholesterolemic-induced rat group (G2). Pistachio and/or almonds consumption significantly decreased cholesterol, LDL-C, VLDL-C, phospholipids TG and MDA levels (P < 0.001, respectively). The results agree with Grieal and Kris-Etherton (2006), Phung et al., (2009), Sheridan et al., (2007), Kocyigit et al., (2006) and Aksoy et al., (2007).
Mechanisms for the Action of Statins

Statins act by competitively inhibiting HMG-CoA reductase, the first committed enzyme of the HMG-CoA reductase pathway. Because statins are similar to HMG-CoA on a molecular level they take the place of HMG-CoA in the enzyme and reduce the activity by which it is able to produce mevalonate, the next molecule in the cascade that eventually produces cholesterol (synthesis of cholesterol in the liver), as well as a number of other compounds. They alter the conformation of the enzyme when they bind to its active site. This prevents HMG-CoA reductase from attaining a functional structure. The change in conformation at the active site mimics the change in conformation at the active site of an internal bile salt circulation into which migrates to the nucleus as well as from G1 to G2; c: significant from G3; d: significant from G4; e: significant from G5. Significant at P < 0.001.

Table 3: Effect of nuts (pistachio and/or almonds) consumption on lipid profile of hypercholesterolemic rats.

<table>
<thead>
<tr>
<th>S. Cholesterol</th>
<th>S. HDL-C</th>
<th>S. LDL-C</th>
<th>S. VLDL-C</th>
<th>TG</th>
<th>Phos.Lip</th>
<th>Chol HDL-C</th>
<th>HDL-C</th>
<th>LDL-C</th>
<th>MDA (μmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>mg/dl</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>G1 77.9±1.15</td>
<td>40.39±0.35</td>
<td>24.06±0.58</td>
<td>13.50±1.05</td>
<td>60.42±1.87</td>
<td>487.89±2.20</td>
<td>1.93±0.03</td>
<td>1.69±0.04</td>
<td>70.92±2.38</td>
<td></td>
</tr>
<tr>
<td>G2 202.46±2.42</td>
<td>41.62±0.64</td>
<td>132.90±1.75</td>
<td>27.94±2.37</td>
<td>100.81±3.51</td>
<td>847.29±1.107</td>
<td>3.71±0.09</td>
<td>2.18±0.01</td>
<td>124.11±2.72</td>
<td></td>
</tr>
<tr>
<td>G3 99.8±2.99</td>
<td>41.80±0.92</td>
<td>39.58±2.68 b</td>
<td>18.48±0.54</td>
<td>80.45±1.59</td>
<td>60.37±1.78</td>
<td>2.40±0.08</td>
<td>1.07±0.05</td>
<td>97.92±1.56 b</td>
<td></td>
</tr>
<tr>
<td>ab</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>G4 106.89±2.99</td>
<td>41.57±0.92</td>
<td>46.91±2.68 b</td>
<td>18.40±0.54</td>
<td>78.26±1.59 b</td>
<td>698.57±2.82</td>
<td>2.58±0.08</td>
<td>0.91±0.05</td>
<td>90.86±1.56</td>
<td></td>
</tr>
<tr>
<td>G5 107.06±1.67</td>
<td>42.77±0.48</td>
<td>45.21±1.16</td>
<td>19.08±0.97</td>
<td>60.37±1.56</td>
<td>697.06±7.55</td>
<td>2.51±0.05</td>
<td>0.95±0.03</td>
<td>90.73±1.77</td>
<td></td>
</tr>
<tr>
<td>G6 100.52±1.98</td>
<td>42.37±0.50</td>
<td>42.40±0.89 b</td>
<td>15.76±1.52</td>
<td>74.55±1.55</td>
<td>603.10±7.40</td>
<td>2.37±0.05</td>
<td>1.00±0.02</td>
<td>85.56±1.54</td>
<td></td>
</tr>
<tr>
<td>G7 72.49±0.81</td>
<td>39.37±0.59</td>
<td>23.84±0.48</td>
<td>9.27±0.45</td>
<td>66.29±1.37</td>
<td>478.34±9.12 b</td>
<td>2.43±0.04</td>
<td>0.95±0.03</td>
<td>68.97±1.51 b</td>
<td></td>
</tr>
<tr>
<td>G8 73.39±1.25</td>
<td>39.90±0.83</td>
<td>24.08±0.48</td>
<td>9.41±0.14</td>
<td>70.45±2.43</td>
<td>479.37±8.11 b</td>
<td>2.37±0.02</td>
<td>0.99±0.01</td>
<td>69.92±1.58 b</td>
<td></td>
</tr>
<tr>
<td>G9 66.94±1.50</td>
<td>39.89±0.87</td>
<td>18.21±0.81</td>
<td>8.94±0.16</td>
<td>55.56±1.56 b</td>
<td>484.57±7.82 b</td>
<td>2.33±0.06</td>
<td>1.09±0.05</td>
<td>65.86±1.56</td>
<td></td>
</tr>
</tbody>
</table>

DISCUSSION

Although nuts are nutrient that has high fat content, there is a fear of increased energy intake but surprisingly nut consumption has been associated with lower or no change in BMI with no fear of weight gain (Bos-Rastrollo et al., 2009; Mattes et al., 2008). Few studies have specifically examined the effects of nuts on body weight (Fraser et al., 2002). In our study there was no significant change in body weight of normal rats consuming nuts which agrees with Edwards et al., 1999 (in humans study using pistachio or almonds) and agree...
also with Spiller et al, 1992, Jenkins et al, 2002 for almonds, and disagree with Lovejoy et al, 2002 for almonds where a significant increase in body weight was found in their study. Many of the supplementation studies that have examined the effects of nuts on lipid profiles have not found negative effects on body weight (Jenkins et al, 2002; and Edwards et al., 1999). The non-significant change in body weight may be due to some degree of mal absorption of energy in nuts (Fraser et al., 2002). In this study a significant decrease in body weight of hypercholesterolemic rats were observed, but after supplementation of the diet with nuts the weight began to increase, still significantly lower than normal control but body weight gain of rats after the supplementation closely near body weight gain of normal group in this period (unpresented data).

In our study no direct correlation between weight gain and nut consumption were found which agree with Sabate’ 2003.

In this study, TC and TG levels were significantly increased in the hyperlipidaemic group compared with the control group. Also the results of this study showed that diet supplemented with pistachio and/or almond induced significant decrease in TC and LDL-C, VLDL level, while HDL-C levels were unchanged (the reduction in TC being attributed to changes in LDL-C) of hypercholesterolaemic rats compared to control group consuming normal control diets. Clinical and epidemiological studies have reported the beneficial effects of tree nuts and peanuts on serum lipid levels (Aksoy et al., 2007; Ème kil-Alturfan et al., 2007). On the other hand, there are controversial results about the effects of pistachio nut and/or almond consumption on the lipid profile of patients with hypercholesterolaemia. Our results disagree with Sheridan et al., 2007 who studied the effects of 15 % of the daily caloric intake in the form of pistachio nuts on the lipid profiles of free-living human subjects with primary and moderate hypercholesterolaemia and found no significant differences in TC and TG levels and agree with Edwards et al, 1999 who reported decreased TG and TC levels in patients with hypercholesterolaemia; and agree with Kocyigit et al., 2006 who observed non-significant decreases in TG levels in healthy volunteers.

The overall result, therefore, is toward a less atherogenic lipid profile. A 1% drop in serum cholesterol reduces the risk for CHD by 2% (Jain et al, 2007). Kinison et al., (1995) and Natarajan et al. 2003 have reported that changes in ratios of TC/HDL-C and LDL-C/HDL-C are better predictors of CHD risk reduction than changes in levels (Panagiotakos et al., 2003). The dietary intervention did alter these ratios in a cardio-protective direction. In our study the beneficial effects of pistachio and/or almonds consumption over the six-week intervention period were modest (the decrease reach > 66% in comparison with G2, it is even better >75% on combination of both nuts), so it is possible that the cumulative effect of long term consumption could prove cardio-protective and help lower coronary artery disease.

The primary sources of fat in the treated groups were from pistachio or almonds. This effect on lipids may be due to pistachio or almonds type of fats or to other factors as the influence of other substances contained in the nuts or in the diet.

Nuts are low in saturated fatty acids (SFA; 5.77 gm/100 gm for pistachio; and 8.22 gm/100 gm for almonds) and high in unsaturated fatty acids (USFA; 47.61 gm/100 gm for pistachio; and 42.27 gm/100 gm for almonds). The predominant type of unsaturated fatty acid in most nuts is MUFAs (32.75 gm/100 gm for pistachio; and 33.47 gm/100 gm for almond). USFA (MUFA and PUFA) contribute 88.71 (pistachio): 83.7 (almonds) % of the calories supplied by these nuts, and 33.47 gm/100 gm for almonds) and high in unsaturated fatty acid in most nuts is MUFAs (32.75 gm/100 gm for pistachio; and 33.47 gm/100 gm for almond). USFA (MUFA and PUFA) contribute 88.71 (pistachio): 83.7 (almonds) % of the energy from fat. 

There are persuasive evidences that dietary substitution of monounsaturated fatty acids (MUFA) or ω-6 polyunsaturated fatty acids (PUFA) for SFA lowers blood cholesterol and may have beneficial effects on inflammation, thrombosis, and vascular reactivity. MUFA may have an advantage over PUFA because enrichment of lipoprotein lipids with MUFA increases their resistance to oxidation. Intake of unsaturated fatty acids with nuts is intrinsically cardio protective (Kris-Etherton, 1999; and Kris-Etherton et al., 2001).

Cholesterol levels in the body result from two sources: absorption from the gastrointestinal tract and endogenous de novo synthesis. The reduction in the values of lipid profile levels may be due to inhibition of hepatic cholesterol synthesis, or the redistribution of cholesterol from plasma to the liver by the cholesterol metabolizing enzyme systems in the liver or the control of lipids utilization.

Nuts such as almonds and pistachios are rich in several beneficial compounds, as ω-6 or ω-9 or ω-3 fatty acids which have demonstrated beneficial effects on blood cholesterol and lipoprotein profiles (Sabate’ 2003). Also, it has been shown to elicit cardio protective effects. The highly unsaturated ω-3 fatty acids eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are primarily responsible for this effect (Stone et al., 1996). Metabolic studies have shown that consumption of α-PUFA lowers circulating cholesterol level (Ramachandran et al., 2003).

Nuts are complex plant foods that are not only rich sources of unsaturated fat but also contain several non-fat constituents such as plant protein, fiber, micronutrients (e.g.: copper and magnesium), vitamins as A, C, E, plant sterols, and phytochemicals that may provide additional protective effects (Kris-Etherton 1999; and Kris-Etherton et al., 2001). These nutrients have shown favourable lipid-altering activity (Vorster et al., 2003).

The first possibility may be the source of protein that was added to the diet. We used casein in the control diet which supplied about 16 g of milk protein/100 gm diet to replace the majority of the protein provided by pistachio or almonds. There is significant literature indicating that amino acid profiles, including the arginine: lysine ratio (pistachio: 2.21:1/19; almonds: 2.56:0.65) in nuts, have beneficial effects on blood lipids when compared with animal proteins (Jenkins et al., 1989). Further, protein in nuts has an arginine-rich amino acid profile that is thought to be protective (Kritcheksky et al., 1982). Among amino acids, the most abundant amino acid found in nuts, may account for the hypcholesterolemic effect observed in animal studies (Kuwoswa and Carroll 1994) or in human intervention trials (Maxwell et al., 2000). In addition, pistachios are relatively high in the semi essential amino acid arginine (2.21g/21.4g protein), which appears to maintain flexible arteries and to enhance blood flow by boosting nitric oxide, a compound that relaxes blood vessels. There is significant literature indicating that amino acid profiles, including the favorable arginine:lysine ratio in nuts, have beneficial effects on blood lipids when compared with animal proteins (Kuwoswa and Carroll 1994 and Jenkins et al., 1989).

A second possible explanation may be the contribution of the dietary fibers supplied by the pistachio or almonds (pistachio: 12.5 g/100g, of which 25 % is soluble fibre; almonds: 10.0 g/100g, of which 10% is soluble fibre respectively, providing 0.76 and 0.89 g of DR). Soluble fibre has been shown to reduce total and LDL-cholesterol concentrations and improve glycemic control (Anderson et al., 1994 and Brown et al., 1999). They concluded that 2–10 g/d of soluble fibre was associated with small but significant decreases in TC.

A third possible explanation is the presence of lipid-altering phytochemicals such as plant sterols and saponins that are found in almonds (Farquhar 1996; and Okenfull 1996). Also pistachios contain significant amounts of phytosterols and other phytochemical compounds such as polyphenols and elagic acid. The major phytoesterol component is β-sitosterol, which is one of several plant sterols implicated in cholesterol lowering (Jones et al., 1997). Studies showed that 2 g of plant sterols/d significantly reduces cholesterol absorption, which in turn decreases plasma TC and LDL-C concentrations (Farquhar 1996; Okenfull 1996 and Vorster et al., 2003).

Almonds contain a variety of phenolic compounds, localized principally in their skin, including flavonols, flavanols, flavonones, anthocyanins procyanidins (B2 and B3), and phenolic acids (caffeic acid, ferulic acid, p-coumaric acid, protocatechuc acid, vanillic acid) (Amarowicz et al., 2005 and Wijeratne et al., 2006). Almond flavonols and flavanols have been shown to be bioavailable and contribute to the antioxidant protection against LDL-C oxidation in vitro and in vivo (Jenkins et al, 2002 and Chen et al., 2005).
A Fourth possible explanation is that almost all nuts are good sources of minerals as magnesium, copper. Magnesium (Mg) level in nuts (pistachio and almonds), provide 8–10% of the DRI for this essential mineral in a 25 g. RDI of Mg is 360–400 mg/day for adult human (RDIs 1997) Magnesium is important since low magnesium status can contribute to myocardial infarction, and possibly hypertension. Magnesium is also critical to enzyme function. Copper (Cu) in nuts (pistachio, almonds), >50.5% of the DRI for copper (700 μg/day) (RDIs 1997) respectively and therefore nuts can be a significant source of this essential mineral (Allen et al., 1977). Copper plays a key role in hematopoiesis and diets low in copper has been associated with adverse changes in lipids, glucose tolerance, blood pressure, and electrocardiograms (Klevay 1993).

A Fifth possible explanation is nuts content of Vitamin E. Vitamin E in high doses (>100 IU/d) has been shown to reduce the risk of coronary heart disease (Rimm and Stampfer1997). This cardio-protective effect appears to be due to vitamin E-induced inhibition of LDL oxidation (vitamin E is transported in the LDL particle (Steinberg and Lewis 1997)), a key step in the atherogenic process. Nuts are a rich source of vitamin E, although the quantities obtained from typical nut consumption are far less than the amounts shown to have beneficial effects on coronary heart disease. Nonetheless, nut consumption is still an effective means of increasing vitamin E intake. RDI of vitamin E is 15 mg/day for adult humans. Almonds in particular are especially rich in many tocopherols, including α-tocopherol, the most active form of vitamin E, which has also shown potent anti-atherogenic effects (Food and Nutrition Board, 2004; Jenkins et al., 2002 and Chen et al., 2005).

It is apparent that fibre, vitamin E, arginine, phytosterols, and phenolic components from realistic amounts of nuts are not sufficient to exert individual hypocholesterolemic effects. In essence, it is possible that there are multiple small effects that contribute, and these are mediated by more than the lipid-lowering fatty acid composition.

A sixth possible explanation that might explain the cholesterol-lowering effects of pistachios is through Stearoyl-CoA desaturase (SCD) and cholesteryl ester transfer protein (CETP). Stearoyl-CoA desaturase (SCD) is the rate-limiting enzyme that catalyzes the synthesis of MUFA; 18:1 and 16:1 from SFAs; 18:0 and 16:0 and plays an important role in cholesterol, triacylglycerol, and lipoprotein metabolism. Stearoyl-CoA desaturase (SCD) plays an important role in lipid metabolism by catalyzing the synthesis of MUFA, mainly 18:1 and 16:1, from SFAs. The ratio of SFAs to MUFA in plasma reflects the membrane phospholipids composition, and increases in this ratio have been implicated in diseases such as CVD, obesity, and diabetes (Ntambi and Miyazaki 2004) so consumption of nuts that contains high levels of unsaturated fats resulted in a significantly lower ratio of 16:1/16:0. The direct correlations between change in SCD activity and lipids show lipoproteins suggest that SCD activity may contribute to the lipid-lowering effects of pistachios. Chole steryl ester transfer protein (CETP) is a plasma protein that plays a key role in reverse cholesterol transport by transferring cholesteryl esters (CEs) from HDL particles to LDL and VLDL particles in exchange for triacylglycerols. CETP may be antiatherogenic in that it increases the rate of reverse cholesterol transport, but it may be proatherogenic in that it transports CE from HDL, which is protective, to VLDL and LDL, which are atherogenic (Cuchel and Rader 2007). Studies in humans have shown that the intakes of SFAs (Schwab et al., 1996) and trans fatty acids (Van et al., 1995) increase CETP, whereas the intake of MUFA decreases (Jansen et al., 2000) and the intake of PUFA decreases (Bagdade et al., 1992) or has no effect on CETP (Thomas et al., 2004).

Oxidative stress (disruption of the balance between oxidative and antioxidative processes) plays an important role in the pathogenesis of atherosclerosis (Steinberg et al., 1989). A cholesterol rich diet results in increased lipid peroxidation by the induction of free radical production, followed by hypercholesterolemia, a major risk factor for atherosclerosis. It has been reported that hypolipoproteinemic atherosclerosis is associated with an increase in tissue concentration of lipid peroxidation products, malondialdehyde and conjugated dienes (Lorgeril et al., 1994).

Scientists have concluded that overproduction of reactive oxygen species (ROS) (oxidative stress) plays a pivotal role in the oxidation of LDL molecules, which get accumulated in the layers of blood vessels. Lipid oxidation due to generation of ROS is considered as an important factor in the initiation and progression of several diseases (Fasoyiroy and Adegoke 2006). The amount of lipid peroxide was measured by MDA assay, which is considered as indirect measure of the formation of lipid peroxides free radicals.

In the present study TBARS levels significantly increased in the hyperlipidaemic group. Decreased antioxidant levels are possibly due to their increased utilization combating excessive plasma oxidative stress in hypercholesterolaemic rats. Consequently, decreased TAA in the hyperlipidaemic group might be responsible for the increased peroxidation of the membrane lipids in this group since increased peroxidation of membrane lipids causes reduction in the activity of antioxidative enzymes. Disturbed balance between oxidants and antioxidants due to hyperlipidaemia has been shown before (Emekli-Alturfan et al., 2008). On the other hand, there is an increasing but inconclusive body of evidence suggesting that nuts improve antioxidant levels (Kocyigit et al., 2006; Gentile et al., 2007; Emekli-Alturfan et al., 2008). Consequently, in the present study pistachio and/or almonds supplementation in the hyperlipidaemic group significantly decreased TBARS levels when compared with the untreated hyperlipidaemic group.

The antioxidant effects of pistachio against oxidative damage might originate from phytochemicals in its content that have strong free radical scavenging ability (Tapiero et al., 2002; Tokusoglu et al., 2005; Kocyigit et al., 2006). Polyphenols, including flavonoids, can exert their antioxidant activity by inhibiting the activities of enzymes, including lipoperoxide and cyclooxygenase, by chelating metal ions, and, most importantly, by scavenging free radicals. Generally, polyphenols are potent free radical scavengers because phenolic groups are excellent nucleophiles (Tapiero et al., 2002). Moreover, it may be assumed that polyphenols in pistachio or almonds reinforce the antioxidant system. These results suggest that pistachio and/or almonds could be a useful compound to control hypercholesterolaemia by both improving the lipid profile and modulating oxidative stress. This modified balance between the antioxidative enzymes might be able to remove superoxides more efficiently (Tapiero et al., 2002; Tokusoglu et al., 2005; Kocyigit et al., 2006).

Jenkins et al., 2008 predicted that the higher intake of vitamin E, MUFA, and phenolic constituents with almond consumption, and the interactions between these nutrients, would increase the status of vitamin E and decrease the level of lipid peroxidation, specifically reducing the biomarkers of oxidative damage, serum MDA.

CONCLUSION
This study supports the benefits of a diet supplying a reasonable amount of fat as monounsaturated fat, while low in saturated fat, for control of plasma cholesterol. Results suggest that pistachio or almonds supplementation may improve blood lipids, ameliorate oxidative stress and this may be due to interactive or additive effects of the numerous bioactive constituents found in pistachio or almonds. Nuts may have beneficial applications in the prevention of cardiovascular diseases.

REFERENCES


10. Breuer C. Nutrient and mineral content of the most common nuts. The clipper. The journal for the international Trade in Processed Food, Dried Fruit and Nuts; 1993; 8: 12-20.


45. Krichelsky D, Tepper SA, Czarnecki SK, and Kurfield DM. Atherogenicity of animal and vegetable protein-influence of

52
the lysine to arginine ratio. Atherosclerosis, 1982; 41: 429–431.


