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Effect of pistachio consumption on plasma lipoprotein subclasses in pre-diabetic subjects

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Abstract *Background and aims:* Nuts have been demonstrated to improve several cardiovascular risk factors and the lipid profile in diabetic and pre-diabetic subjects. However, analysis of conventional serum lipid profiles does not completely explain the atherogenic risk associated with pre-diabetes. We therefore investigated whether chronic consumption of pistachio modifies the lipoprotein subclasses to a healthier profile in pre-diabetic subjects.
Methods and results: Randomized cross-over clinical trial in 54 subjects with pre-diabetes. Subjects consumed a pistachio-supplemented diet (PD, 50% carbohydrates, 33% fat, including 57 g/d of pistachios daily) and a control diet (CD, 55% carbohydrates, 30% fat) for 4 months each, separated by a 2-week wash-out. Diets were isocaloric and matched for protein, fiber and saturated fatty acids. Nuclear magnetic resonance (NMR) was performed to determine changes in plasma lipoprotein subclasses. Small low-density lipoprotein particles (sLDL-P) significantly decreased after pistachio consumption compared to the nut-free diet ($P = 0.023$). The non-high-density lipoprotein particles (non-HDL-P i.e. VLDL-P plus LDL-P) significantly decreased under the PD compared to CD ($P = 0.041$). The percentage of sHDL-P increased by 2.23% after the PD compared with a reduction of 0.08% after the CD ($P = 0.014$). Consequently, the overall size of HDL-P significantly decreased in the PD ($P = 0.007$).
Conclusion: Chronic pistachio consumption could modify the lipoprotein particle size and subclass concentrations independently of changes in total plasma lipid profile, which may help to explain the decreased risk of cardiovascular disease and mortality associated with those individuals who frequently consumed nuts.
Registration number: This study is registered at www.clinicaltrials.gov as NCT01441921.

Abbreviations: CV, cardiovascular; CD, control diet; DOSY, diffusion-ordered ^{1}H NMR spectroscopy; DSTE, double stimulated echo; EPIRDEM, Effect of Pistachio Intake on Insulin Resistance and Type 2 Diabetes Mellitus; FID, finite impulse decay; HDL, high-density lipoprotein; IRAS, Insulin Resistance Atherosclerosis Study; ITT, intention-to-treat; LDL, low-density lipoprotein; LED, longitudinal eddy current delay; NMR, nuclear magnetic resonance; -P, particle; PD, pistachio diet; PLS, partial least-squares; PP, per protocol; T2DM, type 2 diabetes mellitus; VLDL, very low-density lipoprotein.

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Introduction

Atherogenic dyslipidemia is a common feature of type 2 diabetes (T2DM) characterized by high levels of serum triglycerides, low HDL-cholesterol (HDL-C) concentrations and a relative increase in the number of small dense LDL particles (sLDL-P). Nonetheless, the lipid and lipoprotein profile often displays other abnormalities in the pre-diabetic stage, which may contribute to the subsequent increased risk of developing T2DM and cardiovascular (CV) disease. In this regard, large VLDL and small LDL particles have been related with a higher severity and incidence of coronary artery disease and type 2 diabetes [1]. However, results on HDL subfractions are more controversial. Thus, whereas some studies showed an association between small HDL-P and coronary risk [2], others found that small and medium-sized HDL particles were associated with a lower risk of total stroke [3].

While conventional cardiovascular prevention strategies focus on decreasing LDL-C concentrations, increasing data suggests that preventive and therapeutic strategies could be focusing on lipoprotein subfractions abnormalities [4]. However, there is a lack of information on the potential modulatory effects of diet on lipoprotein subfractions and its effects on health and disease.

As far as nutritional factors are concerned, both epidemiological and clinical studies have provided a body of scientific evidence on the cardioprotective effects of nuts and their lipid-lowering properties. A pooled analysis of 25 clinical trials including different types of nuts showed a significant dose-related reduction in total cholesterol and LDL-C, but no effect on HDL-C or triglycerides (except in participants with hypertriglyceridemia) after nut consumption [5]. However, only one study has assessed the effect of nut consumption on the composition and particle size of lipoprotein subfractions. This study showed beneficial changes in lipid distribution in lipoprotein subfractions after walnut consumption, and no changes in plasma lipid composition [6]. Compared to other nuts, pistachios have lower fat (mostly from poly- and mono-unsaturated fatty acids) and energy content, and higher levels of fiber (both soluble and insoluble), potassium, phytosterols, γ-tocopherol, vitamin K, xanthophyll and carotenoids thereby contributing to explain the beneficial relation between pistachio consumption and health-related outcomes [7].

The aim of the present study is to evaluate the effect of chronic intake of pistachios on lipoprotein size and subclass concentration in pre-diabetic subjects as a potential mechanism for decreasing the cardiovascular risk associated with pre-diabetes.

Methods

Study characteristics

The EPIRDEM (Effect of Pistachio Intake on Insulin Resistance and Type 2 Diabetes Mellitus) study is a randomized, controlled, cross-over trial with a four-month dietary intervention in each period conducted in pre-diabetic subjects. The institutional review board of the Universitary Hospital of Sant Joan de Reus approved the study protocol in September 2011. Executed informed consent was obtained from all study participants. The trial was registered in Clinical Trials (identifier NCT01441921).

Study population

Eligible participants were community-living men and women aged between 25 and 65 years, body mass index $\leq$ 35 kg/m^2 and pre-diabetes was considered when fasting plasma glucose levels were between 100 and 125 mg/dL. Subjects were excluded if they met one of the following criteria: a) diabetes mellitus or using oral anti-diabetic drugs; b) alcohol, tobacco or drug abuse; c) frequent consumption of nuts or known history of allergy to them; d) use of plant sterols, psyllium, fish oil supplements and multivitamins, vitamin E or other antioxidant supplements, e) bad dentition, involving difficulty to chew pistachios; f) following a vegetarian or a hypocaloric diet to lose weight; g) being pregnant or wishing to become pregnant 9 months before or during the study, lactating 6 weeks before or during the study; h) significant liver, kidney, thyroid or other endocrine diseases; i) medical, dietary or social conditions that hinder compliance with the intervention.

Study design

A 15-days run-in period proceeded the four-month treatment period. A 2-week wash-out period separated the 2 crossover sequences. At baseline, data on medical history, physical examination and fasting blood for biochemical analysis were collected. Subjects who met the inclusion criteria were randomly assigned to one of the two different intervention sequences, before the 15-days run-in period, using a computer-generated random-number table. They were instructed to follow a normocaloric diet that provided 50% of energy as carbohydrates, 15% as protein, and 35% as total fat during the 2 weeks proceeding each study period.

The isocaloric diet was individually calculated using WHO equations adjusted by the estimated energy expenditure in physical-activity leisure-time. After the 2-week run-in period, subjects were randomized to one of the two sequences: starting with a control diet (CD) followed by the pistachio supplemented diet (PD), or starting with the pistachio diet followed by the control diet. The main characteristics of both intervention diets have already been published [8]. Participants allocated to the pistachio diet (PD) were supplemented with 2 ounces of pistachio (57 g/day, half roasted and half roasted and salted). In the control diet (CD), the energy intake of other fatty foods, mostly olive oil, was adjusted to compensate for the energy from pistachios included in the PD. Adherence to the intervention period was assessed by counting the empty sachets of pistachio administered and by measuring