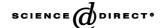


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# Hypolipidemic and hypocholesterolemic effect of argan oil (*Argania spinosa* L.) in *Meriones shawi* rats

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#### **Abstract**

The potential health benefits of various dietary oils in relation to cardiovascular disease and cancer are recently receiving considerable attention. The main proposal of this study is to investigate the effect of dietary argan oil, obtained from seeds of  $Argania\ spinosa\ L$ . (Sapotaceae) endemic from Morocco, on serum lipids composition. Hyperlipidemia was induced by high calorie and cholesterol (HCC) diet administration in 16 rats ( $Meriones\ shawi$ , a rodent of the Gerbillideae family). Eight rats were treated with argan oil ( $1\ ml/100\ g$  weight) daily by oral route during 7 weeks (treated group). Control animals were also fed with HCC diet for 7 weeks. After 7-week treatment with argan oil, blood lipoproteins were significantly reduced. Total cholesterol decreased with 36.67% (P < 0.01), low density lipoprotein (LDL)-cholesterol in 67.70% (P < 0.001), triglycerides in 30.67% (P < 0.05) and body weight in 12.7% (P < 0.05). High density lipoprotein (HDL)-cholesterol concentration remained unaltered. These results indicate the beneficial effect of argan oil in the treatment of the hyperlipidemia and hypercholesterolemia. This effect will be related with the polyunsaturated fatty acids and other constituents of studied oil.

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Keywords: Argan oil; Hypolipidemia; Cholesterol; HDL; LDL; Triglycerides; Rat

# 1. Introduction

Argan oil obtained from *Argania spinosa* L. seeds is eaten raw in southwest of Morocco where it represents 25% of diet fat and 9% of annual production (Rahmani, 1989). This oil is also used in traditional medicine for its cosmetic, bactericide and fungicide properties (Boukhobza and Pichon, 1988).

Chemical analysis of this oil highlighted a glyceride fraction (99%) that is mainly rich in polyunsaturated fatty acids like oleic C18:1 (47.7%) and linoleic C18:2 acid (29.3%) (Chimi et al., 1994). Studies with the unsaponifiable fraction revealed that argan oil is rich in tocopherol (62.0 mg/100 g), mainly  $\alpha$ -tocopherol. This compound makes argan oil a very important source of Vitamin E and also contributes to better preservation of this oil since it protects against oxidation (Chimi et al., 1988).

The prevalence of coronary artery disease morbidity and mortality is particularly low in Mediterranean regions, where the traditional diet, which is rich in olive oil, provide high amounts of monounsaturated fatty acids (Kafatos and Mamalakis, 1993). It has been know for many years that the type of fat in the diet influences blood lipids levels, and, consequently, the risk of the development of atherosclerosis and related cardiovascular diseases (Hegsted et al., 1965). Early studies showed that dietary intake of n-6 polyunsaturated fatty acids, which are abundant in vegetable oils, are inversely related to the incidence of cardiovascular disease. Dietary linoleic acid (C18: 2n-6) serves as a precursor for biosynthesis of arachidonic acid (C20: 4n-6), the substrate for eicosanoid synthesis. It has long been accepted as having hypocholesterolemic effects (Keys et al., 1957; Hegsted et al., 1965). More recently, acid derivatives, particularly gamma-linolenic acid, were found to be even more potent in reducing blood cholesterol in humans and rats (Takada et al., 1994). Those studies pointed out that vegetable oil rich in polyunsaturated fatty acids are able to decrease the plasmatic levels of VLDL and low density lipoprotein

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(LDL)-cholesterol and to increase the plasmatic level of high density lipoprotein (HDL)-cholesterol (Jacotot and Lasserre, 1988; Campbell et al., 1994; Michihiro et al., 1996).

## 2. Material and methods

## 2.1. Argan oil extraction

Argan oil used was extracted by artisan methods from fresh seeds collected the same year in order to prevent auto-oxidative reactions. The extraction was carried out in Essaouira (southwest Morocco). The principle of traditional extraction was reported by Charrouf and Guillaume (1999).

#### 2.2. Animals and diet preparation

Adult male *Meriones shawi* rats (12–14 weeks old) were fed with a high calorie and cholesterol (HCC) diet by enriching the chow with 1% cholesterol and 3% lard (composition of diet: protein (14%), carbohydrate (61%), sugar (9%), fats (20%), salts (4%), vitamin mix (1%)). Each animal received 16 g of chow daily during 7 weeks and had free access to water. Animals were maintained individually in the metabolic cage in a temperature-controlled room (22  $\pm$  2 °C) under a light/dark cycle of 12 h.

Once hyperlipidemia was reached, the rats were randomly divided in two groups of eight animals according to the treatment received. Treated animals received argan oil (1 ml/100 g weight) by gastric intubations everyday during 7 weeks together with HCC. Control animals received HCC chow and 1 ml/100 g weight of distilled water. Rats from both groups were weighted every week.

Blood was collected by retro-orbital sinus punction before starting HCC feeding in day (0), day (45) (before treatment with argan oil or water) and day (90) (after the treatment was finished). Blood was centrifuged for 15 min at 3500 rpm. Plasma was used for determination of total cholesterol, HDL, LDL and triglycerides.

#### 2.3. Biochemical determination

Total cholesterol was measured by an enzymatic method based on the liberation of ester of cholesterol lipoproteins by effect of detergents.

C-HDL concentration in plasma was determined by the enzymatic method based on specific precipitation of VLDL and LDL in the presence of magnesium ions. Plasma levels of triglycerides and C-LDL were assessed by precipitation in the presence of amphiphilic polymers (Esderst and Michirina, 1979; Fruchart, 1988).

# 2.4. Statistical analysis

Results are expressed as mean  $\pm$  S.E.M. of seven to eight values obtained from different animals. Student's t-test was

used for statistical analysis. P < 0.05 values were considered as significant different.

## 3. Results and discussion

The results obtained showed an improvement of lipid profile in rats with 7-week treatment of argan oil (1 ml/100 g) weight). Significant decreases were observed in the plasma concentration of triglycerides (30.76%, P < 0.05, Fig. 1), total cholesterol (36.67%, P < 0.001, Fig. 2), and LDL (67.70%, P < 0.001, Fig. 3). However, the increase the HDL plasma concentration was not significant (3.05%, Fig. 4). It is also interesting to point out the decrease in body weight (12.70%, P < 0.05, Fig. 5).

Although this improvement in plasma lipid concentration, they did not reach initial values. This fact may be due to the HCC diet that was administered together with argan oil. This effect of argan oil was engendered by different mechanisms.

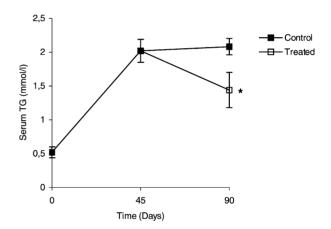


Fig. 1. Serum triglycerides (TG) level before and after argan oil treatment in *Meriones shawi* rat. Values represent mean  $\pm$  S.E.M. of eight rats. \*P < 0.05 vs. control group.

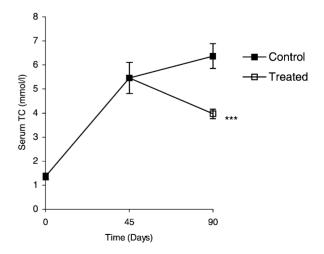


Fig. 2. Serum total cholesterol (TC) level before and after argan oil treatment in *Meriones shawi* rat. Values represent mean  $\pm$  S.E.M. of eight rats. \*\*\*\*P < 0.001 vs. control group.

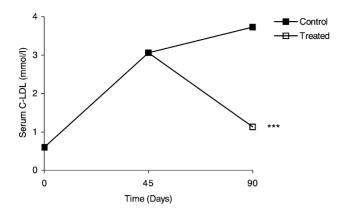


Fig. 3. Serum LDL-cholesterol level before and after argan oil treatment in *Meriones shawi* rat. Values represent mean  $\pm$  S.E.M. of eight rats. \*\*\*P < 0.001 vs. control group.

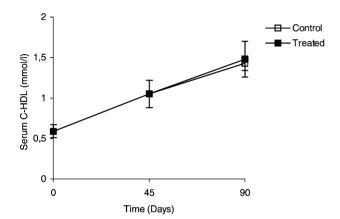


Fig. 4. Serum HDL-cholesterol level before and after argan oil treatment in *Meriones shawi* rat. Values represent mean  $\pm$  S.E.M. of eight rats.

In order to explain the hypolipidaemic effect of argan oil, it is interesting to consider its richness in unsaturated fat with a ratio (polyunsaturated/saturated) of 3.34 and a ratio (monounsaturated/polyunsaturated) of 1.63. Those

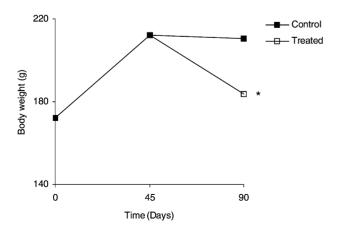


Fig. 5. Body weight level before and after argan oil treatment in *Meriones shawi* rat. Values represent mean  $\pm$  S.E.M. of eight rats. \*P < 0.01 vs. control group.

unsaturated fatty acid has shown beneficial effects like decrease of cholesterolemia and proatherogenic lipoproteins levels. Polyunsaturated fatty acids have protective effects against oxidation, which is explained by their double bonds.

Argan oil is very rich in oleic and linoleic acid. Oleic acid is a 47.7% unsaturated acid that is difficult to oxidize and that is involved in the fluidity of lipoproteins and as a consequence in generation of HDL (Sola et al., 1990). Dietary linoleic acid serves as a precursor for biosynthesis of arachidonic acid, the substance for eicosanoid synthesis through activity of the enzyme cyclooxygenase and 5-lipoxygenase. It has long been accepted as having hypocholesterolemic effects (Keys et al., 1957; Hegsted et al., 1965). More recently, linoleic acid derivatives, particularly gamma-linolenic acid were found to be even more potent in reducing blood cholesterol in humans and rats (Takada et al., 1994). Richard et al. (1990), also reported that a dietary intake of 3-8 mmol gamma-linolenic acid per day reduce serum total and LDL cholesterol levels. However, linoleic acid in argan oil artisan extracted is more stable because of the low concentration of conjugated dienes, which are the main precursors in oxidative reactions (Chimi et al., 1994).

The decrease in cholesterol concentration could be due to a low intestinal absorption of cholesterol because of the activity of saponins in argan oil. Other compounds present in argan oil that could be preventing lipoprotein structural alteration are beta-carotenes and  $\alpha\text{-tocopherol},$  since this compound has Vitamin E action and it is one of the main natural antioxidants. Cabalero (1995) has reported that a supplement of Vitamin E decreases oxygen free radical generation and induces an increase of glucose utilization and response to insulin in normal and diabetic patients.

Another study carried out with some vegetal oils, confirmed our observation (Jacotot, 1990). They show that substitution of the intake of saturated fatty acids by unsaturated fat induce a decrease in total cholesterol concentration. The effects of sunflower, olive and rape oils in diet are remarkable since they have a low concentration of saturated fat. On the other hand, cholesterol concentrations are increased after a diet with corn oil although saturated fat concentrations are similar to those in olive and rape oils. They have also observed an increase of HDL and decrease of LDL without changing the total cholesterol concentration after 6 months diet with olive oil, that is poor in polyunsaturated fatty acids (Jacotot, 1990). In the same study, it was found that a diet rich in polyunsaturated acid, not only increased the ratio HDL/LDL, but also decreased total cholesterol concentration.

Depontel (1989) showed that 4-month administration of olive oil to hypercholesterolemic patients induced a decrease of 26% of plasma lipids concentration. Although we have used a different model, it is interesting to point out that this percentage is lower than that obtained in our study with argan oil.

When comparing two different diets, one rich in monounsaturated fat and the other with a high concentration of polyunsaturated fatty acids, similar decreases in total cholesterol and C-LDL were observed. However, the increase in HDL and ApoA1 was higher after a diet rich in monounsaturated fat (Mata et al., 1992). This diet also induced an improvement in glucose, lipids and insulin concentration in non-insulin dependent diabetic patients (Campbell et al., 1994). In accordance with our results, Bennani-Kabchi et al. (1995) showed that olive oil decreased total and LDL-cholesterol in diabetic patients. However, the increase in HDL was only significant after a diet rich in olive oil. The effects of argan oil on body weight shown in the present study were not observed with olive oil.

In summary, argan oil could be used as an equilibrated diet since it has shown to elicit similar results to other oils like olive oil. It also contains some products together with therapeutic effects that are present in olive, sunflower or rape oils.

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