

Biological Activity of *Opuntia ficus indica* Cladodes II: Effect on Experimental Hypercholesterolemia in Rats

E.M. Galati¹, M.M. Tripodo², A. Trovato¹, A. d'Aquino¹ and M.T. Monforte¹

¹Pharmaco-Biological Department, Messina, Italy; ²Department of Organic and Biological Chemistry, Messina, Italy

Abstract

The influence of a daily administration of lyophilized cladodes of *Opuntia ficus indica* (L.) Mill. (Cactaceae) on lipid metabolism was investigated in hypercholesterolemic rats. The preparation was administered either for 15 or 30 days (by gavage at a dose of 1 g/kg in aqueous vehicle in a volume of 1.5 ml/100 g b.w.). Cholesterol, HDL, LDL and triglyceride plasma levels were assayed. Statistically significant reductions in cholesterol, LDL and triglyceride plasma levels were found. The treatment was more effective after 30 days. The effects are probably due to the high fiber content of the cladodes, but other active ingredients could be operating in conjunction with this.

Keywords: *Opuntia ficus indica* (L.) Mill., cholesterol, LDL, HDL, triglycerides, hypercholesterolemic rats, waste matter.

Introduction

Opuntia ficus indica (L.) Mill. (Cactaceae) is indigenous to the dry regions of the new world and is also fairly abundant in the Mediterranean area. The plant is widely cultivated in Sicily for its edible fruit and various parts of the plant are employed in Sicilian folk medicine. Literature data reveal that it is used for many medical purposes (Barbera & Inglese, 1993; Pimienta, 1990), but little has been reported to date on its biological activity.

As part of an ongoing project financed by the Regione Siciliana, we have reported elsewhere on the anti-inflammatory, diuretic, anti-ulcer and cicatrizant properties (Galati et al., 2000, 2002; Mondello et al., 2000; Trovato et al., 2000) of the waste matter from *O. ficus indica*.

The objective of the present work was to examine the hypolipidemic activity of the cladode phytocomplex of *O. ficus indica* and to investigate its potential in an industrial context as a source of commercial pectins.

Several studies have been carried out on *O. ficus indica*, especially with regard to its nutritional value in human and animal foodstuffs, but literature data on its biological effects or its active ingredients are scanty.

Trachtenberg and Mayer (1981, 1982) reported that *O. ficus indica* cladodes have a high mucilage and pectin content. Several studies have confirmed the presence of dietary fibers (Kurasawa et al., 1992; Sanchez-Castillo et al., 1995; Burkitt et al., 1985). The high percentage of pectins and fibers can increase fecal mass and intestinal motility (Rosado & Diaz, 1995), which in turn affect cholesterol and glucose plasma levels (Fernández et al., 1992, 1994; Frati et al., 1990; Camacho-Ibanez et al., 1983).

O. ficus indica cladodes also contain several active ingredients: sitosterol, vitamins, flavonoids, ascorbic acid, and carotenoids (Abramovitch et al., 1968; Burrett et al., 1982; Endo et al., 1987; El-Moghazy et al., 1984; Pinto & Avecedo, 1983; Salt et al., 1987; Sawaya et al., 1983; Teles et al., 1994).

Materials and methods

Plant material

Cladodes (modified stems in cacti) of *O. ficus indica* were harvested from the S. Cono cultivation (Sicily). The cladodes, with epidermis and glochides removed, were homogenized by Ultra-Turrax for 5 min and then lyophilized.

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Address correspondence to: E.M. Galati, Pharmaco-Biological Department, Vill. SS. Annunziata, 98168 Messina, Italy.
E-mail: emgalati@unime.it

The lyophilized cladodes, dissolved in an aqueous medium, were administered in the morning, by gavage, at a dose of 1 g/kg, in a volume of 1.5 ml/100 g b.w.

Animals

Adult male Wistar rats (180–200 g) were used in all experiments. They were kept in standard conditions (temperature $22 \pm 2^\circ\text{C}$; humidity $60 \pm 4\%$; natural light) and water was provided *ad libitum*.

The rats were divided into 5 groups of 10 animals each. At the end of the treatment period, the animals were fasted overnight. Blood was collected under light diethyl-ether anesthesia in heparinized tubes from the carotid artery of the rats. The plasma was immediately centrifuged and kept at -20°C until assayed.

Total cholesterol (Allain et al., 1974), HDL (high density lipoproteins), LDL (low density lipoproteins) (Vikari, 1976; Izzo et al., 1981; Grillo et al., 1981) and triglycerides (Wahlefeld, 1962) were assayed by colorimetric and enzymatic methods (Boehringer Mannheim Diagnostica).

Statistical analysis

Data were expressed as the mean \pm S.E. of 10 determinations. The results were statistically analyzed by Student's *t*-test; $P < 0.05$ versus control was taken as significant.

Normolipidemic rats

Group I was a normolipidemic control group and so the rats were fed a standard diet only (S. Morini, Mil rat GLP). Group II was also maintained on a standard diet, but these animals received in addition the lyophilized cladodes by gavage daily for a period of 30 days. Blood samples were collected from both groups at the end of the treatment period.

Diet-induced hypercholesterolemic rats

Group III was a high-fat diet control group and so the rats were fed with a hypercholesterolemic diet only (cholesterol 2%, sodium cholate 2%, vitamin mixture 2%, oligoelements 0.2%, salt mixture 5.8%, coconut oil 20%, cellulose 4%, sucrose 44%, casein 5%, drakettprotein 15%), supplied by Altromin-Rieper (Bolzano, Italy), for 30 days. Group IV was also maintained on a hypercholesterolemic diet for 30 days, but these animals received in addition the lyophilized cladodes (1 g/kg) once a day in the morning, from day 15 to day 30. Blood samples were collected after 30 days.

In order to further test the hypothesis that cladodes of *O. ficus indica* have a hypocholesterolemic effect, the lyophilized cladodes were administered daily together with a hypercholesterolemic diet for the whole period of 30 days to another group of animals (Group V). Blood samples were collected after 30 days.

Results

There were no significant differences in body weight between the animals of the different dietary groups.

Plasma cholesterol was 13.7% lower in the normolipidemic rats treated with lyophilized cladodes (Group II), when compared to the control group (Group I). Triglyceride levels were also 51.6% lower in Group II, while HDL and LDL levels were unaffected (Fig. 1).

Total cholesterol, LDL and triglyceride plasma levels were very high in the rats that were fed only the hypercholesterolemic diet (Group III). These levels were significantly lower in the rats of Group IV, which were treated from day 15 with the lyophilized cladodes: their cholesterol, triglyceride and LDL levels were 41.2, 44.3 and 43.4% lower, respectively (Fig. 1).

The rats of Group V, fed the hypercholesterolemic diet and simultaneously treated with lyophilized cladodes for the full 30 days, showed a 49% reduction in cholesterol levels, a 44.3% drop in triglycerides and a 52.6% reduction in LDL as compared to control Group III.

Conclusion

Cardenas et al. (1998) reported that the administration of dehydrated *O. ficus indica* cladodes to normolipidemic rats did not affect their total cholesterol and LDL plasma levels. In our experimental conditions, daily oral administration of lyophilized cladodes to normal and hyperlipidemic rats was associated with a reduction in plasma cholesterol, LDL and triglyceride levels, without affecting HDL levels. This reduction was especially evident in hypercholesterolemic rats.

These effects might be due to the soluble fiber contained in the dry cladodes (Rosado & Diaz, 1995). Previously reported data suggest that *O. ficus indica* (prickly pear) pectin might interfere with lipid absorption, and so reduce the quantity of cholesterol delivered to the liver (Fernandez et al., 1992). The pectin might also include bile acids which would stimulate cholesterol catabolism (Ulrich, 1987).

Moreover, prickly pear pectin may have a hypocholesterolemic effect because of the short-chain fatty acids which are produced by the fermentation of the fiber in the colon. In experimental animals, these mainly propionic and acetic acids stimulate insulin secretion and reduce glucose plasma levels (Brockman, 1982). However, pectin intake in rats fed with a hypercholesterolemic diet has been shown to produce negligible effects on short-chain fatty acids (Koseki et al., 1991). Fernandez et al. (1994) reported that prickly pear pectin reduces plasma cholesterol in guinea pigs without affecting cholesterol absorption. The pectin of *O. ficus indica* cladodes probably generates hypocholesterolemic effects in rats by means of a mechanism that alters hepatic cholesterol homeostasis.

Moreover, pectin reduces plasma LDL levels by increasing apolipoprotein B/E receptor expression and receptor-

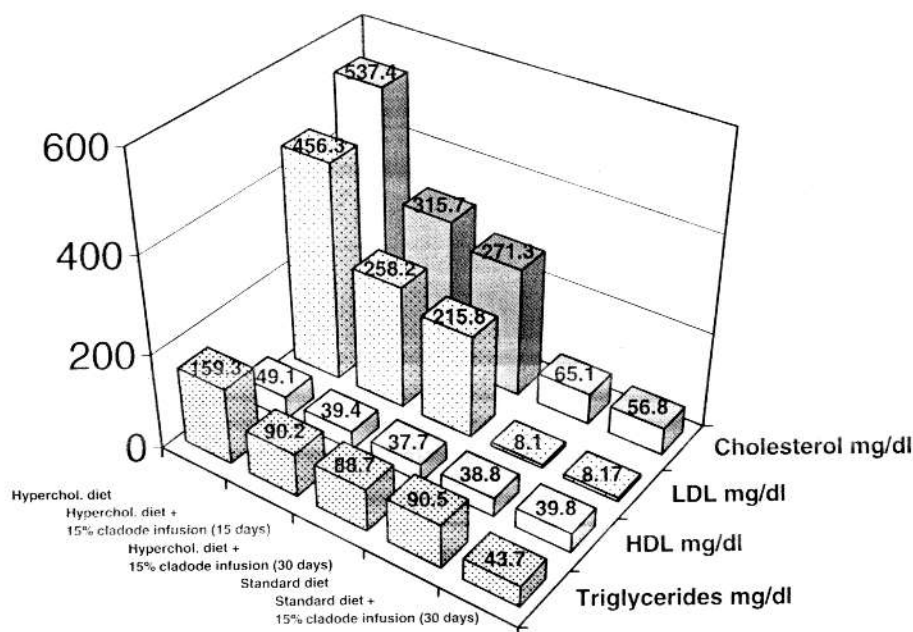


Figure 1. Cholesterol, HDL, LDL and triglyceride plasma levels in normolipidemic rats and in rats with diet-induced hypercholesterolemia, after 15- and 30-day treatment with *O. ficus indica* 15% cladode infusion (1 g/kg).

mediated LDL turnover (Fernandez et al., 1990, 1992), and so the hypolipidemic effect of the cladodes could also be due to this mechanism. However, it is possible that some of the other active ingredients present in *O. ficus indica* cladodes could have hypolipidemic effects and be acting in conjunction with the pectins. Some of these active ingredients (β -sitosterol, flavonoids, vitamins) exercise a protective influence against several diseases (e.g., atherosclerosis, coronary heart disease) in which aetiology the oxidative stress plays an important role. It is well known that flavonoids influence lipid metabolism and many authors have reported that flavonoids lower serum cholesterol (Cody et al., 1986; Monforte et al., 1995).

Ascorbic acid can influence cholesterol levels by a mechanism which involves hydroxylation reactions. Such reactions transform the cholesterol into bile acid, and constitute a major excretion route for cholesterol (Ginter, 1975). β -sitosterol has also been shown to reduce serum cholesterol in different animal species (Behar & Anthony, 1955; Gould, 1955; Shipley et al., 1968; Bhattacharyya & Lopez, 1979; Malini & Vanithakumari, 1990). These substances may offer some degree of protection against the onset of hypercholesterolemia and against the damage this condition can inflict (Goldstein & Brown, 1989; Manninen et al., 1992; Watts et al., 1992).

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