

Evaluation of Chios Mastic Gum on Lipid and Glucose Metabolism in Diabetic Mice

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ABSTRACT Chios mastic gum (MG), a resin produced from *Pistacia lentiscus* var. *Chia*, is reported to possess beneficial cardiovascular and hepatoprotective properties. This study investigated the effect of crude Chios MG on metabolic parameters in diabetic mice. Streptozotocin-induced diabetic 12-week-old male C57bl/6 mice were assigned to three groups: NC ($n=9$) control; LdM ($n=9$) animals receiving low dose mastic for 8 weeks (20 mg/kg body weight [BW]); and HdM ($n=9$) animals receiving high dose mastic (500 mg/kg BW) for the same period. Serum lipid and glucose levels were determined at baseline, at 4 and 8 weeks. Serum total protein, adiponectin, and resistin levels were also measured at the end of the experiment. Histopathological examination for liver, kidney, aorta, and heart lesions was performed. After 4 weeks, MG administration resulted in decreased serum glucose and triglyceride levels in both LdM and HdM, whereas BW levels were reduced in LdM group compared with controls. At the end of the experiment, LdM presented significantly lower serum glucose, cholesterol, low-density lipoprotein cholesterol, and triglyceride levels and improved high-density lipoprotein cholesterol levels compared with control group. HdM group had ameliorated serum triglyceride levels. Hepatic steatosis observed in control group was partially reversed in LdM and HdM groups. MG administered in low dosages improves glucose and lipid disturbances in diabetic mice while alleviating hepatic damage.

KEY WORDS: • Chios mastic gum • diabetes • dietary herbal supplement • hepatoprotection • lipid metabolism • mice

INTRODUCTION

CHIOS MASTIC GUM (MG) is a resin produced from *Pistacia lentiscus* var. *Chia*, an evergreen shrub of the Anacardiaceae family. The variety *Chia* is uniquely cultivated in the Greek island of Chios, in the Aegean Sea. Its resin is obtained as an exudate in the form of tears or droplets, practically by longitudinal incisions of the trunk and branches of the tree.

The beneficial, healing properties of MG are known since antiquity.^{1,2} Since 1997 mastic extracts have been characterized as Products of Protected Origin by the European Union.³ Among MG pharmacological activities, the eradication of bacteria and fungi that may cause peptic ulcer, tooth plaque formation, and malodor of the mouth are reported.⁴ A few years ago, MG was specifically reported to be effective against *Helicobacter pylori* *in vitro*.^{5,6} However, clinical trials remain controversial with MG administration

failing to prove the eradication of *H. pylori* infection in the stomach compared with antibiotics.⁷

MG has been reported to aid in the amelioration of symptoms of autoimmune diseases by inhibiting the production of pro-inflammatory substances by activated macrophages and the production of cytokines in patients with active Crohn's disease.⁸ Other anti-inflammatory and antioxidant properties are documented in *in vitro* studies.³

The major constituents of MG as determined by Gas chromatography–mass spectrometry method are α -pinene, β -pinene, β -myrcene, limonene, and β -caryophyllene.⁹ The anti-inflammatory activity of MG can be mainly attributed to a variety of compounds such as triterpenes of the oleanane, euphane, and lupine type.^{10,11} Further, it contains α -tocopherol and polyphenols, which have been previously associated with the hypotensive effects of mastic.¹¹ MG anti-bacterial activity appears to arise from verbenone, α -terpineol, and linalool.^{11,12}

MG has been associated with cardiovascular protection by effectively lowering the levels of serum cholesterol and inhibiting human low-density lipoprotein (LDL) oxidation *in vitro*.⁴ A study by Petersen *et al.* reported that the oleanonic acid, an extract of MG, is a peroxisome proliferator-activated receptor γ (PPAR γ) agonist exerting antidiabetic

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properties.¹³ Recently, Vallianou *et al.* showed that the hypocholesterolemic and hypolipidemic activities of the essential oil from Chios MG were associated with one of its constituents, a bicyclic monoterpene namely camphene and suggested that this effect is associated with a mechanism different from that of statins.¹⁴

In this study, the biological activity of MG on lipid and glucose metabolism in diabetic mice was investigated with a particular focus on diabetic control and amelioration of lipid profile. This is a novel research study focusing on the impact of crude MG on diabetes mellitus and on the diabetes-associated lipid disturbances in mice.

MATERIALS AND METHODS

Twenty-seven male mice C57bl/6 (*Mus Musculus*), 12-week-old and weighing 25–30 g each were used. The animals were obtained from Alexander Fleming Institute (Vari, Greece) and were acclimatized for 1 week before the experiment started. The animals were housed under standard laboratory conditions ($22 \pm 2^\circ\text{C}$) and humidity (60%) with 12 h light and dark cycle. The experimental protocol was reviewed and approved by the Ethics Committee of the Medical School of the University of Athens and by the Veterinary Directorate of Athens Prefecture Committee, in accordance with the ethical recommendations of the European Communities Council Directive of November 24, 1986 (86/609/EEC).

The mice were separated into the following three groups: NC ($n=9$) control mice; LdM ($n=9$) animals that received low dose MG for 8 weeks (20 mg/kg Body Weight [BW]); HdM ($n=9$) animals that received high dose MG for the same period (500 mg/kg BW). In all groups normal chow (4FR25; Mucedola, Milan, Italy) was administered during the entire experiment. Food and water were given *ad libitum*. Because MG is highly insoluble in water, its administration in the drinking water of the mice was not feasible. Thus, the crude MG was ground to fine powder and then mixed with normal chow. Water and food intake of mice was recorded daily. Mice BWs were recorded once a week.

The crude of MG was supplied by the Chios MG Growers Association, which is the exclusive worldwide producer of the mastic resin. The MG Growers Association receive and elaborate those productions of crude MG that are of good quality and purity to ensure thus that the resin is free of other compounds or pesticides.

All mice became diabetic with intraperitoneal injection of streptozotocin (40 mg/kg BW), confirmed by fasting serum glucose measurements 2–3 days after the administration of streptozotocin, where glucose levels were found to be >200 mg/dL. In those mice where fasting glucose levels were not >200 mg/dL, a second injection of streptozotocin was given.

Blood samples were collected from mice at the establishment of diabetes (Baseline), at 4 weeks (T_1) and at the end of the experimental period (T_2 ; 8 weeks). All blood samples were obtained in the morning after a 12-h fasting period, using capillary tubes introduced into the medial retro-orbital venous plexus under light ether anesthesia. The

serum was separated by centrifugation at 3000 rpm (1358 g) for 10 min. Serum concentrations of total cholesterol and of triglycerides were determined using the enzymatic PAP commercial kit (“biosis”-Biotechnological Applications, Athens, Greece) and high-density lipoprotein (HDL) cholesterol was determined with a cholesterol enzymatic photometric method. LDL cholesterol was determined by the mathematic model “LDL cholesterol = Total Cholesterol – (HDL cholesterol + Triglycerides/5).” Serum total protein levels were determined with commercially available kits (Biosis Biotechnological Applications, Athens, Greece).

Measurements of adiponectin and resistin

The measurements of adiponectin and resistin in serum samples obtained at the 8th week of the experiment were performed with Enzyme-Linked Immunosorbent Assay (ELISA) using commercial available kits (Adiponectin: Biovendor, MOUSE Adiponectin Elisa, Cat. No RD293023100R; Resistin: Biovendor (MOUSE Resistin Elisa, Cat. No RD293016100R).

Histopathological staining

At the end of the 8-week period, animals were euthanized under ether anesthesia. The liver, kidneys, aorta, and heart of the animals were dissected immediately for histopathological examination. The tissues were fixed in 10% formalin at room temperature. The tissues were then embedded in paraffin, sectioned, and mounted on glass microscope slides. The sections were stained with hematoxyllin–eosin and blindly examined by two independent researchers using light microscopy. Specifically, the liver was evaluated as had been previously described by the Pathology Committee of non-alcoholic steatohepatitis Clinical Research Network.¹⁵ The histological features in liver were divided into five broad categories: steatosis, ballooning, portal inflammation, focal necrosis, and lobular activity. A score from 0 (absence) to 3 (severe lesion) was assigned to each parameter.

Statistical analysis

Data were expressed as mean \pm 1 standard deviation for continuous variables. The normality of the distributions was assessed with Kolmogorov–Smirnov test and graphical methods. Comparisons between more than two groups were performed with Analysis of Variance (ANOVA) using Benjamini and Hochberg’s False Discovery Rate (FDR) to assess between-group differences, and to control familywise error to <0.05 . Kruskal–Wallis’s test was utilized as a non-parametric test for multiple group comparisons, using Mann–Whitney’s U test and FDR for *post hoc* multiple testing. Comparisons between more than two measurements of the same group were performed using Repeated Measures ANOVA for normal distributions, or Friedman’s test and Wilcoxon’s signed rank test for non normal distributions, with FDR, as *post hoc* tests. Normalization of continuous variables for covariates and subsequent comparisons were

TABLE 1. BODY WEIGHT, SERUM LIPID, AND GLUCOSE LEVELS

<i>t</i>	<i>Parameter</i>	<i>NC (n=9)</i> <i>Mean ± SD</i>	<i>LdM (n=9)</i> <i>Mean ± SD</i>	<i>HdM (n=9)</i> <i>Mean ± SD</i>	<i>Significant differences between groups</i>
Baseline	Glucose (mg/dL)	224.67 ± 43.02	215.78 ± 10.87 ^{2,3}	244.00 ± 24.88 ^{2,3}	NS
	Body weight (g)	27.33 ± 2.45	25.78 ± 2.90 ^{2,3}	27.11 ± 2.67	NS
	Total cholesterol (mg/dL)	86.67 ± 11.51 ³	81.11 ± 4.88	90.22 ± 4.26	NS
	LDL cholesterol (mg/dL)	35.75 ± 10.14	40.17 ± 4.97 ^{2,3}	37.00 ± 6.86	NS
	HDL cholesterol (mg/dL)	37.11 ± 1.90 ²	30.56 ± 3.24 ^{2,3}	38.11 ± 5.18	NC vs. LdM*, LdM vs. HdM*
	Triglycerides (mg/dL)	69.00 ± 13.43	51.89 ± 6.64	75.56 ± 11.61	NC vs. LdM*, LdM vs. HdM*
T ₁	Glucose (mg/dL)	232.88 ± 24.99	167.89 ± 7.80 ¹	176.44 ± 19.45 ¹	NC vs. LdM*, NC vs. HdM*
	Body weight (g)	28.89 ± 2.67	24.89 ± 2.76 ¹	27.33 ± 2.00	NC vs. LdM*
	Total cholesterol (mg/dL)	87.00 ± 6.84 ³	84.33 ± 7.35 ³	86.00 ± 24.41	NS
	LDL cholesterol (mg/dL)	28.96 ± 7.35	24.40 ± 11.42 ^{1,3}	37.28 ± 18.44	NS
	HDL cholesterol (mg/dL)	36.33 ± 2.34 ^{1,3}	49.33 ± 8.24 ¹	38.78 ± 5.23	NC vs. LdM*, LdM vs. HdM*
	Triglycerides (mg/dL)	108.56 ± 44.36	53.00 ± 14.18 ³	60.56 ± 16.84	NC vs. LdM*, NC vs. HdM*
		<i>Mean ± SD</i>	<i>Mean ± SD</i>	<i>Mean ± SD</i>	
T ₂	Glucose (mg/dL)	231.50 ± 35.03	160.33 ± 14.21 ¹	194.33 ± 43.86 ¹	NC vs. LdM*
	Body weight (g)	28.33 ± 3.67	24.56 ± 2.79 ¹	27.11 ± 3.02	NC vs. LdM*
	Total cholesterol (mg/dL)	96.83 ± 10.15 ^{1,2}	78.11 ± 5.21 ²	95.44 ± 15.00	NC vs. LdM*, LdM vs. HdM*
	LDL cholesterol (mg/dL)	35.50 ± 9.65	19.78 ± 9.76 ^{1,2}	44.04 ± 11.65	NC vs. LdM*, LdM vs. HdM*
	HDL cholesterol (mg/dL)	37.33 ± 2.06 ²	49.44 ± 8.20 ¹	38.89 ± 5.04	NC vs. LdM*, LdM vs. HdM*
	Triglycerides (mg/dL)	120.00 ± 30.45	44.56 ± 11.10 ²	62.56 ± 25.15	NC vs. LdM*, NC vs. HdM*

Body weight levels (g), glucose (mg/dL), total cholesterol (mg/dL), LDL cholesterol (mg/dL), HDL cholesterol (mg/dL), and triglyceride levels (mg/dL) in serum at baseline, at the 4th (T₁) and at the 8th week (T₂) of the experimental period.

**P* < .05.

¹*P* < .05 versus Baseline.

²*P* < .05 versus T₁.

³*P* < .05 versus T₂.

NC, Control group; LdM, Low dose mastic group; HdM, High dose mastic group; HDL, high density lipoprotein; LDL, low density lipoprotein; NS, non significant.

performed with Analysis of Covariance (ANCOVA) and Multivariate Analysis of Variance and Covariance (MANOVA and MANCOVA). Baseline total cholesterol, triglyceride, and HDL cholesterol measurements were selected as covariates. All tests were two-sided. Differences were considered as statistically significant if the null hypothesis could be rejected with >95% confidence (*P* < .05).

RESULTS

Body weight measurements and serum lipid profile

At baseline, no differences were observed in BW levels, serum total cholesterol, and LDL cholesterol levels, among the groups. Baseline levels of serum HDL cholesterol and triglycerides differed between the studied groups (Table 1).

At T₁, BW levels were significantly decreased in LdM group as compared with the control group (*P* = .006). LdM group exhibited a significant increase in mean HDL cholesterol concentration as compared with the NC group while both LdM and HdM groups had significantly lower triglyceride levels in comparison with the untreated mice (LdM vs. NC and HdM vs. NC; *P* < .001 and *P* = .003 respectively). HDL levels were significantly increased in LdM in comparison to HdM group (*P* = .0015).

At T₂, the LdM group presented significantly reduced BW (*P* = .033), serum total cholesterol levels (*P* < .001), LDL cholesterol levels (*P* = .0045), and triglyceride levels (*P* < .001) and increased HDL cholesterol levels (*P* = .015) compared with the control group. Concerning HdM group, the only significant difference in comparison to NC group

TABLE 2. TOTAL PROTEIN, ADIPONECTIN, AND RESISTIN LEVELS

<i>t</i>	<i>Parameter</i>	<i>NC (n=9)</i> <i>Median</i> <i>(interquartile range)</i>	<i>LdM (n=9)</i> <i>Median</i> <i>(interquartile range)</i>	<i>HdM (n=9)</i> <i>Median</i> <i>(interquartile range)</i>	<i>Significant differences between groups</i>
T ₂	Total proteins (mg/dL)	5.40 (0.20)	5.20 (0.20)	5.30 (0.10)	NS
	Adiponectin (µg/mL)	8.51 (7.32)	21.35 (19.43)	15.80 (7.76)	NS
	Resistin (ng/mL)	3.39 (2.98)	6.24 (6.41)	3.00 (4.75)	NS

Serum total protein (mg/dL), adiponectin (µg/mL), and resistin (ng/mL) levels at the end of the experimental period (T₂).

**P* < .05.

was detected in serum triglyceride levels ($P = .006$). Statistical analysis showed elevated HDL cholesterol levels and reduced LDL cholesterol levels in LdM group compared with HdM group ($P = .0225$ and $P = .0045$ respectively).

Total cholesterol levels in NC group were significantly increased at the end of the 8-week experimental period compared with baseline and T₁ levels (Baseline vs. T₁ and Baseline vs. T₂; $P = .0495$, $P = .045$, respectively).

In LdM group, BW levels were lower at T₁ and T₂ compared with the initial levels ($P = .0135$ in all cases). Total cholesterol levels were significantly decreased in T₂ in comparison to T₁ ($P = .033$), whereas LDL levels were decreased in a significant way at the 4th and 8th week of the experiment compared with baseline (Baseline vs. T₁ and Baseline vs. T₂; $P = .021$ and $P = .018$, respectively). On the contrary, HDL cholesterol levels in LdM group were increased with time (Baseline vs. T₁, Baseline vs. T₂; $P < .001$, $P < .001$, respectively). Serum triglyceride levels in this group were also lower at the 8th as compared with the 4th week of the experiment ($P < .001$). In HdM group, no differences were recorded in serum lipid and BW levels during time (Table 1).

Serum glucose and total protein levels

Baseline serum glucose levels did not show any statistically significant difference between groups (Table 1). At T₁, serum glucose levels were lower in a statistically significant way in both LdM and HdM groups compared with the control group (LdM vs. NC and HdM vs. NC; $P < .001$ and $P < .001$, respectively). At the 8th week of study, statistical analysis revealed decreased serum glucose levels in LdM group in comparison with controls ($P < .001$).

No statistically significant differences were recorded in serum glucose levels of control mice among the three different time points. Both low and high dose MG-treated animals presented decreased, in a statistically significant way, serum glucose levels in T₁ and T₂ as compared with baseline (LdM group: Baseline vs. T₁ and Baseline vs. T₂; $P < .001$ and $P < .001$, respectively, HdM group: Baseline vs. T₁ and Baseline vs. T₂; $P = .003$ and $P = .006$, respectively) (Table 1). Serum total protein levels did not differ between groups at T₂ (Table 2).

All statistically significant differences between the three groups at week 4 and 8 excepting total cholesterol remained significant even after normalizing for baseline total cholesterol, HDL cholesterol, and triglyceride levels.

Serum adiponectin and resistin levels

Serum adiponectin and resistin levels did not present significant differences in LdM and HdM groups compared with the NC group at the end of the experimental period (Table 2).

Tissue histopathology

Hematoxylin-eosin stained liver samples obtained from all three groups showed signs of hepatic steatosis and focal necrosis (Fig. 1). NC group exhibited the highest scores in

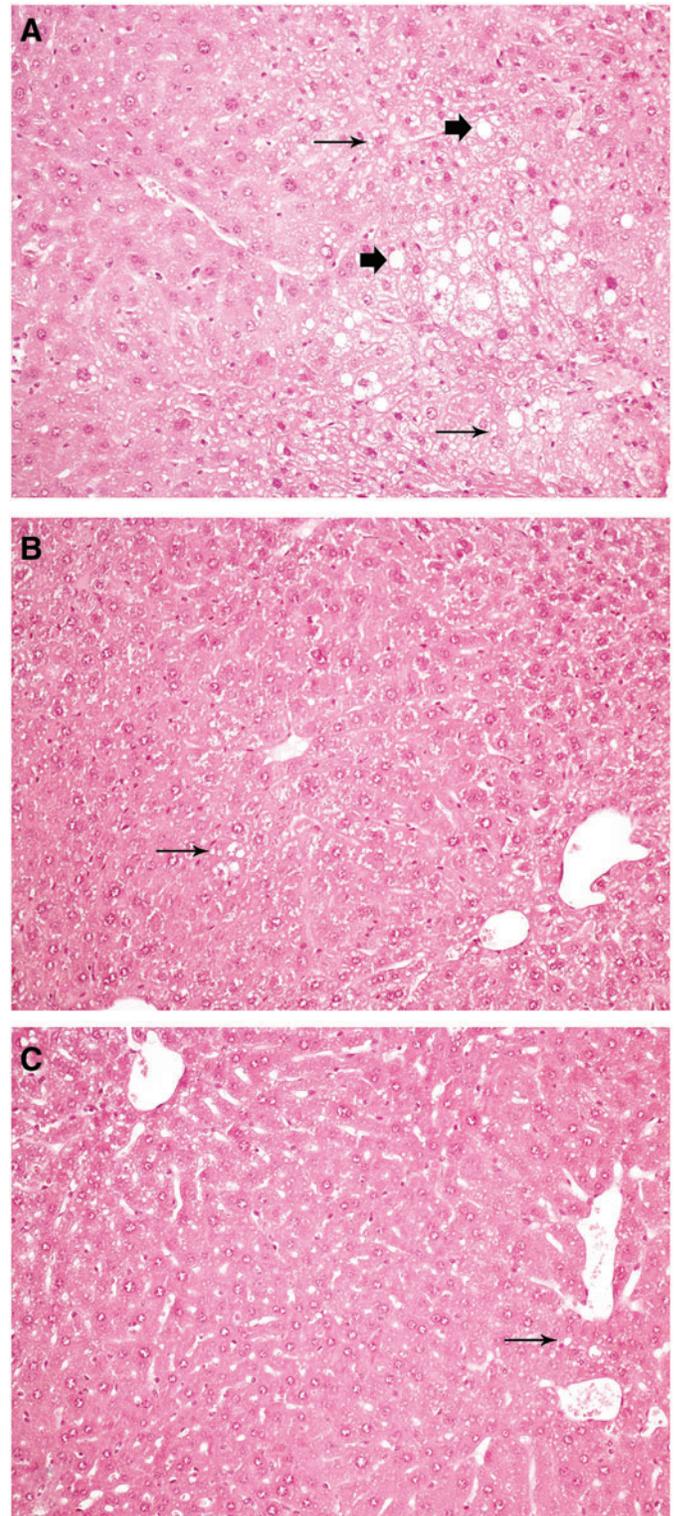


FIG. 1. Representative figures, eosin-hematoxylin stain, 200 \times original magnification. (A) demonstrates moderate steatosis of liver parenchyma, both macrovesicular (thick arrows) and microvesicular (thin arrows). (B) and (C) represent snapshots from the treated groups. Mild steatosis in both groups (thin arrows) was observed, although no significant difference existed between groups. Color images available online at www.liebertpub.com/jmf

TABLE 3. HISTOLOGICAL OBSERVATIONS OF HEPATIC TISSUE AT T₂ (END OF THE EXPERIMENTAL PERIOD)

t	Parameter	NC (n=9)		LdM (n=9)		HdM (n=9)		Significant differences between groups
		Median (interquartile range)	Mean ± SD	Median (interquartile range)	Mean ± SD	Median (interquartile range)	Mean ± SD	
T ₂	Liver steatosis	1.00 (1.00)	1.44 ± 0.53	1.00 (1.00)	0.56 ± 0.53	0.00 (1.00)	0.45 ± 0.73	NC vs. LdM*, NC vs. HdM*
	Liver focal necrosis	0.00 (1.00)	0.55 ± 0.73	0.00 (1.00)	0.34 ± 0.50	0.00 (0.00)	0.11 ± 0.33	NS
	Overall	2.00 (1.00)	2.00 ± 0.71	1.00 (2.00)	1.00 ± 0.87	0.00 (1.50)	0.56 ± 0.88	NC vs. LdM*, NC vs. HdM*

3, severe; 2, moderate; 1, mild; 0, no changes in histology (scores).

* $P < .05$.

steatosis and overall results compared with LdM and HdM group (Steatosis: NC vs. LdM and NC vs. HdM, $P = .0105$ in all cases; Overall: NC vs. LdM and NC vs. HdM, $P = .036$ and $P = .015$, respectively) (Table 3).

The grade of fatty liver disease was considered as “mild” in five of the nine mice in LdM group. In nine mice of the HdM group, the grade was “moderate” in one and “mild” in two mice. Finally, the grade of fatty liver was “moderate” in four and “mild” in five of the nine mice of the control group.

No statistically significant differences were detected in the grade of focal necrosis between groups. In addition, no statistically significant differences were observed in the grade of hepatic steatosis ($P = .514$) and in the overall hepatic histopathology ($P = .249$) between LdM and HdM experimental groups.

Hepatocellular degeneration (ballooning) of grade 1 was confirmed in one animal of LdM group; portal inflammation in one animal of NC group and lobular activity was not observed in any group. No pathological findings were detected in the other examined tissues, such as aorta, heart, and kidneys, of all experimental mice (data not shown).

DISCUSSION

Currently, there is an increased interest and an ongoing search for natural agents with antidiabetic potential but without the detrimental side effects presented in various antidiabetic drugs. The present study investigated the impact of MG administration on a murine diabetic model.

The administration of low dose of MG in our study exhibited beneficial effects regarding serum lipid levels. Both doses of MG led to a decrease in serum triglyceride levels, independent of the treatment period. MG powder administration in high dosages has been previously shown to result in decreased serum total cholesterol and LDL cholesterol levels, total cholesterol/HDL ratio, and atherogenic apolipoprotein B levels in healthy human subjects.⁴

Vallianou *et al.* reported that the intraperitoneal administration of MG oil in normolipidemic and dyslipidemic rats ameliorated their serum lipid levels in a dose-dependent manner.¹⁴ The hypolipidemic activity of MG oil correlated with one of its minor constituents, named camphene. However, it was shown that camphene on its own did not achieve similar levels of serum lipid improvement as compared with the co-administration of camphene along with

the five major components of MG oil. The hypolipidemic properties of camphene have been attributed to its potent action as a lipoprotein lipase activator. A similar pathway could explain the hypolipidemic action of MG observed in the diabetic mice in our study.

Although we observed a marked improvement in triglyceride levels after both low and high dose MG administration, the beneficial activity of MG regarding serum total, LDL, and HDL cholesterol levels was not detected in the high dose MG administration. We could assume that the hypocholesterolemic activity of MG is dose-dependent.

Adiponectin is an adipokine that has been recognized as a key regulator of insulin sensitivity and tissue inflammation. Previous studies indicated that diabetes mellitus and obesity are linked to reduced levels of adiponectin.^{16–21} Adiponectin also exhibits anti-inflammatory activity, enhances the production of anti-inflammatory cytokines,²² whereas it has been used as marker of insulin resistance and lipid profile.^{23,24} In this experiment the modifications in the serum levels of this adipokine were recorded but no significant differences were detected in adiponectin serum levels during the MG administration period. Further investigations are required since this could be due to the relatively short period of the MG administration or due to small sample size.

Regarding serum glucose levels, a significant reduction after the 4-week treatment period for both high and low dose MG groups and after 8 weeks of MG treatment for LdM group, was recorded. In addition, different measurements of the same group in time, showed an improvement in serum glucose levels in the two MG crude supplemented groups. PPARs control a variety of genes in several pathways of lipid metabolism,²⁵ while PPAR γ agonists have insulin-sensitizing effects reducing serum glucose levels in patients with Type 2 diabetes.²⁶ The presence of the oleanonic acid in MG, which acts as a PPAR γ agonist may be implicated in its hypoglycemic activity. However, other mechanisms including the antioxidant and anti-inflammatory properties of Chios MG may be also related to both its hypolipidemic and hypoglycemic function.²⁷

In accordance with our findings, diabetic status induces non-alcoholic fatty liver disease in patients with Type 1 diabetes.²⁸ Serum lipid aberrations are consistent with the fat accumulation observed in the hepatic sections. The administration of MG, independently of the dose, partially

reversed the grade of hepatic steatosis, improving hepatic architecture. The PPAR γ agonist activity of the oleanonic acid in MG may display a role in the amelioration of hepatic histology.²⁹

One possible confounder that must be taken into account is that animals were housed in groups of three per cage, which did not enable the determination of the exact daily food and MG consumption by each individual animal.

The crude resin used in this study contains a high percentage (30%) of an insoluble and sticky polymer (poly- β -myrcene) that is postulated to interfere with oral administration and bioavailability of active compounds present in mastic resin. Thus, various studies of MG have utilized preparations of total mastic extract without this polymer. However, contrary to the findings of prior research, the activity of compositions comprising polymeric myrcene has been found useful to the induction of neuronal cell differentiation, and to the rejuvenation of a large number of cells and tissues.³⁰ In our opinion, future research studies should investigate *in vitro* and *in vivo* all the different constituents of MG separately, and examine their bioavailability in terms of selected target tissues or organs.

The different beneficial actions of MG-oil, besides its traditional uses, should also be investigated with the help of molecular biology techniques. For example, a recent study performed microarray gene expression profiling and bioinformatic analyses on lung adenocarcinoma cells, following mastic-oil treatment.³¹ The researchers identified potential novel target molecules and pathways underlying mastic oil inhibitory actions on tumor cell growth and survival. Such high-throughput studies can help us understand further the pleiotropic actions of MG and its constituents.

In conclusion, Chios MG administered in low dosages appears effective in improving the disturbed glucose and lipid levels of the experimentally diabetic mice. Further, hepatic damage is alleviated during MG administration. Chios MG should be further investigated in future studies for its medicinal effects as a potent natural antidiabetic agent.

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AUTHOR DISCLOSURE STATEMENT

No competing financial interests exist. No conflicts of interest declared.

REFERENCES

- Moussaieff A, Fride E, Amar Z, Lev E, Steinberg D, Gallily R, Mechoulam R: The Jerusalem Balsam: from the Franciscan Monastery in the old city of Jerusalem to Martindale 33. *J Ethnopharmacol* 2005;101:16–26.
- Kolliarios G: Chios Mastic. Chios mastic from antiquity to today. In: *Tradition and Current Practice*. Acta of the International Symposium held in Chios on October 3–5, 1997; Athens, Greece, 1997, pp. 242–243.
- Triantafyllou A, Bikineyeva A, Dikalova A, Nazarewicz R, Lerakis S, Dikalov S: Anti-inflammatory activity of Chios mastic gum is associated with inhibition of TNF-alpha induced oxidative stress. *Nutr J* 2011;10:64.
- Triantafyllou A, Chaviaras N, Sergentanis TN, Protopapa E, Tsaknis J: Chios mastic gum modulates serum biochemical parameters in a human population. *J Ethnopharmacol* 2007;111:43–49.
- Marone P, Bono L, Leone E, Bona S, Carretto E, Perversi L: Bactericidal activity of Pistacia lentiscus mastic gum against Helicobacter pylori. *J Chemother* 2001;13:611–614.
- Huwez FU, Thirlwell D, Cockayne A, Ala'Aldeen DA: Mastic gum kills Helicobacter pylori. *N Engl J Med* 1998;339:1946.
- Loughlin MF, Ala'Aldeen DA, Jenks PJ: Monotherapy with mastic does not eradicate Helicobacter pylori infection from mice. *J Antimicrob Chemother* 2003;51:367–371.
- Dimas KS, Pantazis P, Ramanujam R: Review: Chios mastic gum: a plant-produced resin exhibiting numerous diverse pharmaceutical and biomedical properties. *In Vivo* 2012;26:777–785.
- Koutsoudaki C, Krsek M, Rodger A: Chemical composition and antibacterial activity of the essential oil and the gum of Pistacia lentiscus Var. chia. *J Agric Food Chem* 2005;53:7681–7685.
- Assimopoulou AN, Papageorgiou VP: GC-MS analysis of penta- and tetra-cyclic triterpenes from resins of Pistacia species. Part I. Pistacia lentiscus var. chia. *Biomed Chromatogr* 2005;19:285–311.
- Sanz MJ, Terencio MC, Paya M: Isolation and hypotensive activity of a polymeric procyanidin fraction from Pistacia lentiscus L. *Pharmazie* 1992;47:466–467.
- Magiatis P, Melliou E, Skaltsounis AL, Chinou IB, Mitaku S: Chemical composition and antimicrobial activity of the essential oils of Pistacia lentiscus var. chia. *Planta Medica* 1999;65:749–752.
- Petersen RK, Christensen KB, Assimopoulou AN, Fretté X, Papageorgiou VP, Kristiansen K, Kouskoumvekaki I: Pharmacophore-driven identification of PPAR γ agonists from natural sources. *J Comput Aided Mol Des* 2011;25:107–116.
- Vallianou I, Peroulis N, Pantazis P, Hadzopoulou-Cladaras M: Camphene, a plant-derived monoterpene, reduces plasma cholesterol and triglycerides in hyperlipidemic rats independently of HMG-CoA reductase activity. *PLoS One* 2011;6:e20516.
- Korou LM, Agrogiannis G, Pantopoulou A, Vlachos IS, Iliopoulos D, Karatzas T, Perrea DN: Comparative antilipidemic effect of N-acetylcysteine and sesame oil administration in diet-induced hypercholesterolemic mice. *Lipids Health Dis* 2010;9:23.
- Stefan N, Vozarova B, Funahashi T, Matsuzawa Y, Weyer C, Lindsay RS, Youngren JF, Havel PJ, Pratley RE, Bogardus C, Tataranni PA: Plasma adiponectin concentration is associated with skeletal muscle insulin receptor tyrosine phosphorylation, and low plasma concentration precedes a decrease in whole-body insulin sensitivity in humans. *Diabetes* 2002;51:1884–1888.
- Yang B, Brown KK, Chen L, Carrick KM, Clifton LG, McNulty JA, Winegar DA, Strum JC, Stimpson SA, Pahel GL: Serum

- adiponectin as a biomarker for *in vivo* PPAR γ activation and PPAR γ agonist-induced efficacy on insulin sensitization/lipid lowering in rats. *BMC Pharmacol* 2004;4:23.
18. Kadowaki T, Yamauchi T: Adiponectin and adiponectin receptors. *Endocr Rev* 2005;26:439–451.
 19. Kim JY, van de Wall E, Laplante M, Azzara A, Trujillo ME, Hofmann SM, Schraw T, Durand JL, Li H, Li G, Jelicks LA, Mehler MF, Hui DY, Deshaies Y, Shulman GI, Schwartz GJ, Scherer PE: Obesity-associated improvements in metabolic profile through expansion of adipose tissue. *J Clin Invest* 2007;117:2621–2637.
 20. Rasouli N, Yao-Borengasser A, Miles LM, Elbein SC, Kern PA: Increased plasma adiponectin in response to pioglitazone does not result from increased gene expression. *Am J Physiol Endocrinol Metab* 2006;290:E42–E46.
 21. Bodles AM, Banga A, Rasouli N, Ono F, Kern PA, Owens RJ: Pioglitazone increases secretion of high-molecular-weight adiponectin from adipocytes. *Am J Physiol Endocrinol Metab* 2006;291:E1100–E1105.
 22. Carbone F, La Rocca C, Matarese G: Immunological functions of leptin and adiponectin. *Biochimie* 2012;94:2082–2088.
 23. Kern PA, Di Gregorio GB, Lu T, Rassouli N, Ranganathan G: Adiponectin expression from human adipose tissue: relation to obesity, insulin resistance, and tumor necrosis factor- α expression. *Diabetes* 2003;52:1779–1785.
 24. Lara-Castro C, Fu Y, Chung BH, Garvey WT: Adiponectin and the metabolic syndrome: mechanisms mediating risk for metabolic and cardiovascular disease. *Curr Opin Lipidol* 2007;18:263–270.
 25. Desvergne B, Wahli W: Peroxisome proliferator-activated receptors: nuclear control of metabolism. *Endocr Rev* 1999;20:649–688.
 26. Schoonjans K, Auwerx J: Thiazolidinediones: an update. *Lancet* 2000;355:1008–1010.
 27. Paraschos S, Mitakou S, Skaltsounis AL: Chios gum mastic: a review of its biological activities. *Curr Med Chem* 2012;19:2292–2302.
 28. Targher G, Bertolini L, Padovani R, Rodella S, Zoppini G, Pichiri I, Sorgato C, Zenari L, Bonora E: Prevalence of non-alcoholic fatty liver disease and its association with cardiovascular disease in patients with type 1 diabetes. *J Hepatol* 2010;53:713–718.
 29. Seo YS, Kim JH, Jo NY, Choi KM, Baik SH, Park JJ, Kim JS, Byun KS, Bak YT, Lee CH, Kim A, Yeon JE: PPAR agonists treatment is effective in a nonalcoholic fatty liver disease animal model by modulating fatty-acid metabolic enzymes. *J Gastroenterol Hepatol* 2008;23:102–109.
 30. Zadik H: Therapeutic uses of mastic gum fractions. Australia. Patent AU2010220056. March 4, 2010.
 31. Moulos P, Papadodima O, Chatziioannou A, Loutrari H, Roussos C, Kolis FN: A transcriptomic computational analysis of mastic oil-treated Lewis lung carcinomas reveals molecular mechanisms targeting tumor cell growth and survival. *BMC Med Genomics* 2009;2:68.