

Antiobesity effects of seedlac and shellac in rats fed with a high-fat diet

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Abstract

Background and purpose: Obesity is a global health problem and also a well-known risk for many diseases. Although some synthetic drugs have been marketed for the treatment of obesity, natural remedies may be considered as safe and cost-effective alternatives. Lac (*Kerria lacca* Kerr) is a product from animal origin and is sold as seedlac or shellac. This drug is very famous among Unani practitioners for its antiobesity effects. The aim of the present study was to evaluate the antiobesity potential of lac in rats.

Experimental approach: The effect of lac on rats fed with a high-fat diet (HFD) was investigated through determination of the changes in body weight, and serum levels of leptin. In addition, the effect of lac on total cholesterol, triglyceride (TG), low-density lipoprotein-cholesterol (LDL-C) and high-density lipoprotein-cholesterol (HDL-C) was studied. Male Wistar rats (170-220 g) were divided into eight groups; a control group with normal diet, the HFD group received a HFD, and the experimental groups received the HFD containing 0.1, 0.2, and 0.4% (w/w) of seedlac or 0.1, 0.2, and 0.4% (w/w) of shellac for 12 weeks. The body weight of each rat was measured once a week. At the end of the experiment, animals were sacrificed and serum concentrations of cholesterol, TG, low-density lipoprotein-cholesterol, high-density lipoprotein-cholesterol, and leptin were determined.

Results: The study showed that seedlac and shellac significantly prevented increasing body weight and the levels of serum leptin were decreased in treated groups compared with HFD group. Also, shellac decreased TG level and both shellac and seedlac exerted a significant increase in HDL-C concentration.

Conclusion and implications: Lac had weight-reducing properties and could be a promising alternative for controlling obesity.

Keywords: Leptin; Obesity; Seedlac; Shellac.

INTRODUCTION

Studies have shown that overweight is one of the most important risk factors for metabolic diseases, such as type 2 diabetes, hypertension, non-alcoholic fatty liver, dyslipidemia, and cardiovascular diseases (1,2). Obesity can be a risk of estrogen dependent breast cancer after menopause (3). The ultimate goal of treating obesity is to reduce the risk or severity of diseases,

functional disorders and limitations. Some of the common ways to treat obesity include diet changes, increased activity and exercise, drug use, and weight loss surgery (4,5). One of the most commonly used drugs for obesity is orlistat.

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DOI: 10.4103/1735-5362.278715

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This drug changes digestion and absorption of fat by inhibition of the pancreatic lipase enzyme, so that fatty hydrolysis is not performed completely, and the defecation of fecal fats increases (6). Major side effects of orlistat include gastrointestinal disorders such as cramp and abdominal bloating, defecation incontinence, and removal of fatty spots (7). Acute kidney damage caused by oxalate has also been observed following administration of this drug. Therefore, it should not be used in people with a history of oxalate stones (8). Serotonin reduces appetite in humans and animals. Therefore, serotonin receptor agonists can be effective drugs for weight loss. Loracasin is one of these drugs and its side effects include headaches and dizziness, upper respiratory tract infections, nasal pharyngitis, and nausea (9). Other medications include sympathetic agents which reduce the intake of food through the induction of early satiation. Sympathetic drugs such as phentermine, diethylpropion, and benzaphtamine have been approved only for short-term treatment (maximum 12 weeks) of obesity. These drugs are contraindicated in people with coronary artery disease, hypertension, and hyperthyroidism (10).

Today, plant compounds have been widely used in the treatment of some metabolic disorders (11). Some of the most commonly used herbs in the treatment of obesity are *Garcinia cambogia*, *Camellia sinensis*, and *Citrus aurantium* (12-17). Some studies have shown that long-term use of *Garcinia cambogia* can lead to inflammation of the liver and collagen accumulation in the liver (12). Green tea (*Camellia sinensis*) contains caffeine and catechin which prevent lipid absorption and also affect fat metabolism in the liver (14-16,18).

Iranian sources of traditional medicine have recommended the use of laxatives, reduced food and sleep, and the use of hot and dry medicines to treat obesity. Also traditionally, lac has been one of the most commonly used formulations for controlling obesity (19-22).

Kerria lacca or *Laccifer lacca* Kerr. is the scientific name of an insect which parasitically lives on several trees including

Zizyphus mauritiana (Lam.), *Butea monosperma* (Lam.), and *Scheichera oleosa* (Lour.) that are abundant in some Asian countries like Thailand, China, and India. The insect secretes a resin compound known as lac. Shellac is a relatively pure form of lac that is used as a sealing and coating compound in pharmaceutical industries (23-25).

Shellac is composed of resin (70-80%), wax (6-7%), coloring agents (4-8%) , and other substances (15-25%) such as debris, obtained by refining sticklac (the material collected directly from the plant). After washing the sticklac, most of the water-soluble material (e.g. laccic acids) is removed and seedlac is obtained. Seedlac is refined more by melting filtration process, the product obtained is called shellac or common shellac (23-24,26).

Lac has been used for medical purposes in India for a long time. So an example in Ayurveda lac has been used to treat skin burns, wound healing, bone fracture, fever, inflammation, and obesity (23).

The aim of the present investigation was to study the antiobesity potential of lac in a high-fat diet (HFD) induced model of obesity in rats.

MATERIALS AND METHODS

Lac was purchased from Isfahan medicinal herb market and identified by an expert in Museum of Entomolgy (Iranian Research Institute of Plant Protection, Tehran, Iran) and a specimen (No. 2785) was deposited there for future reference.

Male Wistar rats (170-220 g) were purchased from animal house of School of pharmacy and Pharmaceutical Sciences at Isfahan University of Medical Sciences, Isfahan, I.R. Iran. Rats were housed in cages with standard conditions. Animal experiments were performed according to the Guidelines for the Care and Use of Laboratory Animals of Iranian National Committee for Ethics in Biomedical Research (IR.TUMS.VCR.REC.1396.4027). Possible efforts were made to decrease animal number and distress.

Following one week of adaptation with a pelletized commercial diet, 48 rats were randomly divided into 8 groups (n = 6). Rats in control group were fed with standard laboratory pellet chow. Animals in HFD group received HFD (fat 45%, carbohydrate 36%, protein 15%, fiber 2%, calcium 0.6 %, phosphorus 0.7%, other elements and vitamins 0.7% w/w) for 12 weeks. The rats of the experimental group received HFD containing 0.1, 0.2, and 0.4% (w/w) seedlac, and 0.1, 0.2, and 0.4% (w/w) shellac for 12 weeks. The body weight of each rat was measured at the beginning of the study and once a week thereafter.

At the end of the experiment fasting blood samples were collected and centrifuged at 3200 rpm. The serum was separated, aliquoted, and stored at -70 °C for further analysis. The concentrations of serum triglycerides (TG), total cholesterol, low-density lipoprotein- cholesterol (LDL-C), and high-density lipoprotein-cholesterol (HDL-C) were measured spectrophotometrically, using Hitachi 902 Biochemistry Autoanalyzer (Switzerland) by commercial kits from Pars Azmun Co. (Tehran, I.R. Iran). The serum leptin levels were determined using Zell Bio rat leptin ELISA kit (Zell Biotech, Germany).

Statistical analysis

Results are expressed as mean \pm SD. One-way analysis of variance (ANOVA)

with Tukey as the post hoc test was used to evaluate the differences between the groups. SPSS version 13 was used for performing statistical analyses. P value < 0.05 was considered as statistically significant.

RESULTS

Determination of body weight gain and food intake

Body weight change and food intake were determined weekly. Initial body weight of all animals was measured before they were fed with either normal diet or HFD. At the end of each week, the weight gain (%) was calculated as follow:

$$\text{Weight gain (\%)} = \frac{W_1 - W_0}{W_0} \times 100$$

where, W_0 and W_1 are initial weight and weight after treatment, respectively.

At the end of the study the body weight gains of the rats fed with HFD was 57.4%, whereas rats fed with the normal diet only gained 30% weight. The percentage of body weight gain was higher in the HFD group comparing to that of control group.

Body weight gain was decreased in shellac groups. The percentage body weight gains of rats fed with the HFD supplemented with both 0.2 and 0.4% of shellac was significantly ($P < 0.05$) lower compared to rats fed with the HFD alone (Fig. 1).

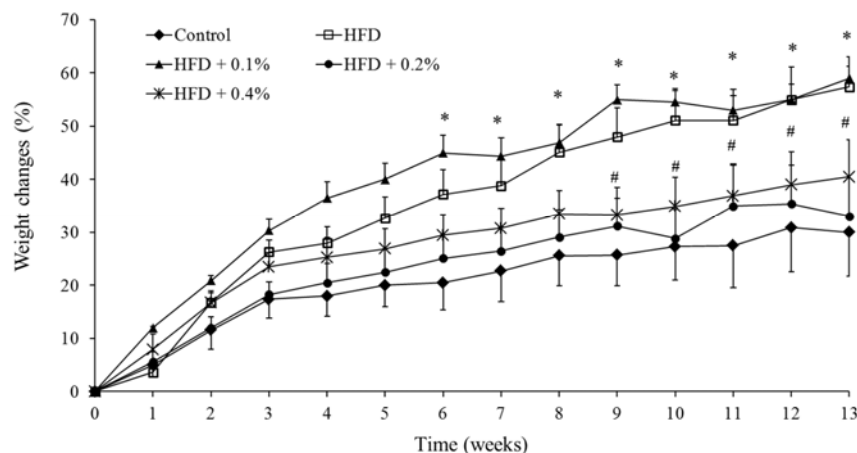


Fig. 1. Effect of shellac (0.1, 0.2, and 0.4%) on animal weight changes. All values are expressed as mean \pm SD. n = 6 rats/group. * $P < 0.05$ indicates significant differences between HFD vs control group and # $P < 0.05$ shows differences between HFD + 0.2% shellac or HFD + 0.4% shellac compared with HFD group. HFD, high fat diet.

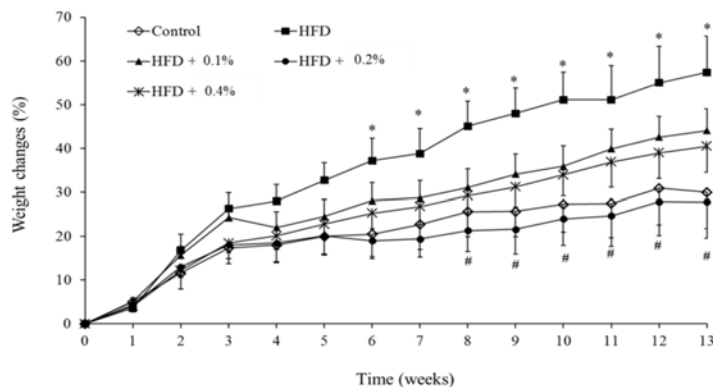


Fig. 2. Effect of seedlac (0.1, 0.2, and 0.4%) on animal weight changes. All values are expressed as mean ± SD. n = 6 rats/group. **P* < 0.05 HFD shows significant differences compared to control group and #*P* < 0.05 indicates HFD + 0.2% seedlac or HFD + 0.4% seedlac differ significantly from HFD group.

Table 1. Effect of seedlac or shellac on total food intake.

Diet	Total food intake (g)
HFD	11400
HFD + 0.1% seedlac	10200
HFD + 0.2% seedlac	9800
HFD + 0.4% seedlac	9200
HFD + 0.1% shellac	10400
HFD + 0.2% shellac	9400
HFD + 0.4% shellac	9600

HDF, High fat diet.

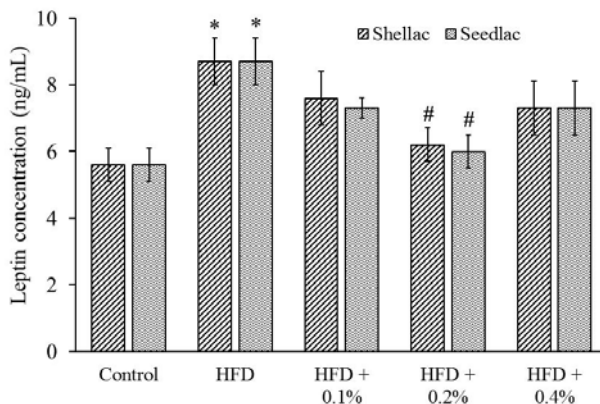


Fig. 3. Effects of shellac and seedlac (0.1, 0.2, and 0.4%) on serum concentration of leptin. Values are mean ± SD. n = 6 rats/group. **P* < 0.05 indicates differences between HFD and control group and #*P* < 0.05 shows differences HFD + 0.2% shellac or seedlac in comparison with HFD alone. HDF, High fat diet.

In seedlac groups, body weight gains in HFD + 0.2 and 0.4% seedlac was significantly (*P* < 0.05) decreased compared with the HFD (Fig. 2).

The addition of seedlac or shellac to the HFD decreased food intake compared with the HFD group. Thus, the highest reduction in food intake was in HFD + 0.4%

seedlac group (9200 g) and the lowest effect was in HFD + 0.1% shellac group (10400 g) (Table 1).

Effects of seedlac and shellac on plasma leptin level

Figure 3 shows that the serum concentration of leptin was significantly

($P < 0.05$) increased in HFD group compared with control group. In addition, the levels of plasma leptin were significantly ($P < 0.05$) decreased in HFD + 0.2% seedlac or shellac groups compared with HFD alone.

Effects of seedlac and shellac on plasma lipid profiles of rats

Figure 4 clearly shows that supplementation of HFD with shellac or seedlac has no significant effect on serum levels of total cholesterol.

HFD did not change serum concentrations of HDL-C. Addition of 0.1% shellac or 0.2% seedlac to HFD significantly ($P < 0.05$)

increased serum levels of this lipoprotein when compared with control or HFD group (Fig. 5).

Fig. 6 shows that shellac or seedlac could not produce any significant change in serum levels of LDL-C in HFD-fed rats.

In the HFD group, the levels of serum TG increased significantly compared with those of the control group (Fig. 7). The levels of serum TG was significantly ($P < 0.05$) decreased in HFD + shellac groups compared with HFD and it was more prominent in HFD + 0.4% group (Fig. 7A). The serum levels of TG in seedlac groups did not show any significant difference with the HFD-fed rats (Fig. 7B).

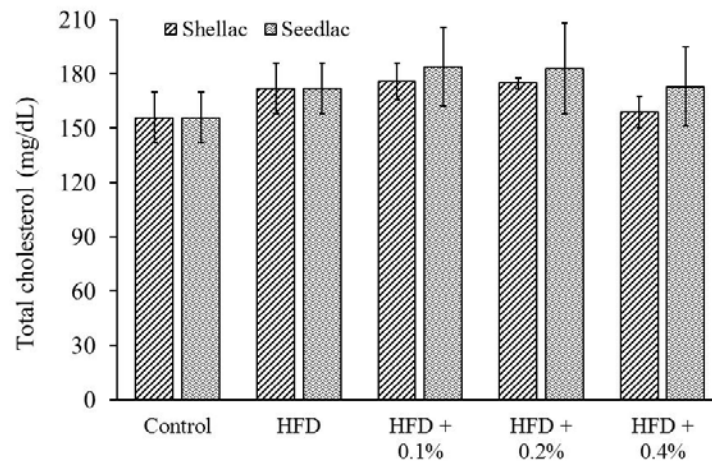


Fig. 4. Effects of shellac or seedlac (1, 2, and 4%) on serum concentration of total cholesterol. All values represent mean \pm SD. n = 6 rats/group. HFD, High fat diet.

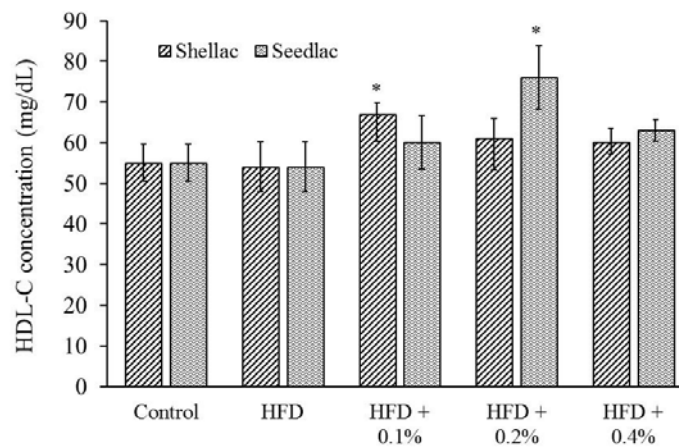


Fig. 5. Effect of shellac or seedlac (1, 2, and 4%) supplementation on serum concentration of HDL-C. Data are expressed as mean \pm SD of serum HDL. n = 6 rats/group. * $P < 0.05$ indicates significant differences compared to control and HFD groups. HDL-C, High-density lipoprotein-cholesterol; HFD, high fat diet.

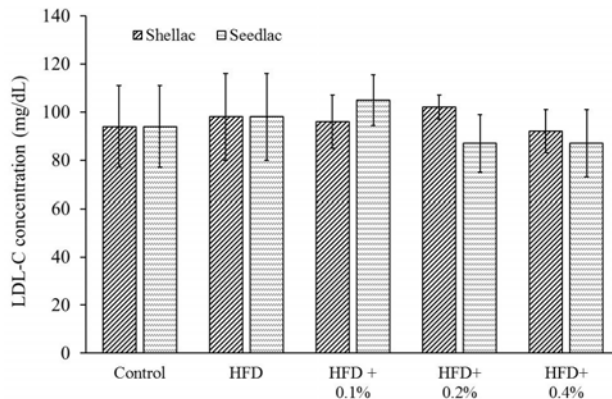


Fig. 6. Effect of shellac or seedlac (1, 2, and 4%) on serum concentration of LDL in HFD-fed rats. All values are mean \pm SD. n = 6 rats/group. LDL-C, Low-density lipoprotein-cholesterol; HFD, high fat diet.

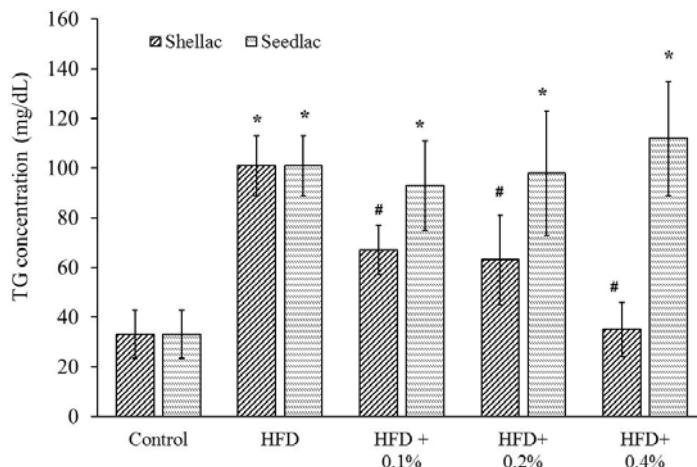


Fig. 7. Effects of shellac or seedlac (1, 2, and 4%) on serum concentration of TG. Values show mean \pm SD. n = 6 rats/ group. * $P < 0.05$ indicates significant differences in comparison with control group and # $P < 0.05$ shows significant differences when compared with HFD alone. TG, Triglyceride; HFD; high fat diet.

DISCUSSION

Diet control and regular exercise have been recommended as safe approaches to manage obesity. Generally, reduction of food intake and increase of energy expenditure are known as beneficial methods to achieve desirable weight loss. However, they are difficult and bothersome ways for long periods and most subjects do not have a good adherence to these programs (27). On the other hand, a recently published article states that the success of physical activity in management of obesity is controversial (28). Therefore, a combination of drug therapy and lifestyle modification is considered more popular approach. Although several

drugs have been introduced as anti-obesity agents, fenfluramin, dexfenfluramine, rimonabant, and sibutramine were withdrawn from the market because of serious adverse effects (29). Currently the US Food and Drug Administration, f have approved five anti-obesityantiobesity medications including orlistat, lorcaserin, naltrexone/bupropion, phentermine/topiramate, and liraglutide have been approved by the US Food and Drug Administration (FDA) for obese patients (30).

Parallel with studies on chemical drugs, many investigators have focused on natural and herbal remedies for weight control. The present study targeted lac that there are some claims about its usefulness in obesity management.

Results of our study showed that body weight was decreased in seedlac groups compared with the HFD group. This decrease was significant in HFD + 0.2 and 0.4% seedlac groups. Also the percentage body weight gains of rats fed the HFD supplemented with both HFD + 0.2 and 0.4% of shellac was significantly lower compared to rats fed with the HFD alone. In a clinical study, lac was given in a dose of 3-5 g twice daily for 60 days and resulted in reduction of body weight and skin fold thickness in 40% of cases, and decreased serum cholesterol and TG levels (31). Our results indicated that seedlac and shellac decreased appetite and food intake and it might explain its weight controlling property.

In the present investigation, the serum level of leptin was also measured. Leptin is a hormone, which is mainly secreted by adipocytes and has a crucial role in regulation of appetite and energy balance (32). Consistent with the previous literature (13) our findings clearly showed that HFD increased the serum level of this hormone. It has been reported that in obesity leptin resistance occur and thereby satiety is not detected by hypothalamus and results in more food intake. Supplementation of HFD with 0.2% of seedlac or shellac decreased weight gain and serum leptin level.

It means that at least a part of their weight reducing effects might be through modulation of leptin release and prevention of HFD-induced leptin resistance.

Obesity is mostly associated with dyslipidemia and it has been reported that obese individuals have higher levels of TG and cholesterol and lower HDL levels and these alterations are more considerable and consistent with TG and HDL (33,34).

Our results showed that shellac could reduce TG. Reduction is significant compared to the HFD group. Decrease in serum TG concentration might be due to increased activity of lipoprotein lipase enzyme activity or the prevention of its biosynthesis (35) and more studies are needed to elucidate a definite mechanism of action.

In the seedlac groups, TG did not change much in comparison with the HFD group

and the impurities present in it seem to have covered its useful effect.

Considering the results, both seedlac and shellac did not change the level of the LDL and total cholesterol but they increased the serum level of HDL. Based on these findings, it seems that lac has weight reducing effect as well as beneficial effects on serum lipid pattern.

In a clinical study on hyperlipidemia, lac was administered in a dose of 3 g twice a day for 3 months. Results showed reduction of serum cholesterol, serum TG, and total lipids at rates of 23, 38, and 12%, respectively. It was also reported that lac did not cause any side effect (36). Our results also demonstrated that the effect of lac was more prominent on TG than other lipids.

CONCLUSION

Our findings clearly demonstrated that both seedlac and shellac are effective to prevent weight gain induced by HFD. They also reduced leptin and increased HDL-C serum concentration. The results confirmed antiobesity activity of lac mentioned in Unani medicine (37) and both seedlac and shellac are promising agents to control weight and treat some lipid disorders.

ACKNOWLEDGEMENTS

This work was financially supported by Research Council of Tehran University of Medical Sciences (Tehran, I.R. Iran) Grant No. 9321427003. We are also thankful to Prof. Gholamreza Amin and Dr. Mohsen Amin for their technical support and Dr. Moghadam in Museum of Entomolgy (Iranian Research Institute of Plant Protection, Tehran, I.R. Iran) for identification of lac.

CONFLICT OF INTEREST STATMENT

The authors declare no conflict of interest for this study.

AUTHORS' CONTRIBUTION

All authors contributed equally in this work.

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