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RESEARCH ARTICLE

Effect of *Pterocarpus marsupium* in animal model of high carbohydrate diet-induced metabolic syndrome

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ABSTRACT

Background: Metabolic syndrome (MS) is a group of interconnected disorder linked to obesity, insulin resistance hypertension, and dyslipidemia. It is a risk factor for the development of Type II diabetes and cardiovascular diseases. Thus, there is a need to search for therapeutic options for the treatment of MS. **Aims and Objective:** The present study was conducted to evaluate the protective effect of *Pterocarpus marsupium* (Bijasar) in animal model of high carbohydrate diet-induced MS. **Materials and Methods:** Adult male Wistar rats were divided into three groups. All the animals were given 20% sucrose in drinking water ad libitum for 8 weeks for the induction of MS along with the following treatment. Group I (Control): Distilled water; Group II (PM group): Aqueous extract of *P. marsupium* (200 mg/kg/day); and Group III (Standard treatment): Metformin 100 mg/kg/day + Atorvastatin 10 mg/kg/day. Body weight, abdominal circumference (AC), blood glucose, and serum triglycerides were evaluated at the end of 4 weeks and 8 weeks. **Results:** The aqueous extract of *P. marsupium* at a dose of 200 mg/dl caused a significant reduction ($P < 0.05$) in the body weight, AC, blood sugar and serum triglyceride levels when compared to the control group. Similar results were seen in the standard treatment group. **Conclusion:** PM was effective in the treatment of major components of MS in rat model of high carbohydrate diet-induced MS.

KEY WORDS: Metabolic Syndrome, *Pterocarpus Marsupium*, Insulin Resistance, Obesity, Heartwood

INTRODUCTION

Metabolic syndrome (MS) is a collection of interconnected disorders linked to central obesity, insulin resistance, dyslipidemia, and hypertension. The term "MS" was coined by a Swedish physician Kylin in 1920 who demonstrated that the association of these symptoms have an increasing risk of diabetes mellitus and atherosclerotic cardiovascular disease, which ultimately leads to high mortality and morbidity in terms of long-term complications.[1]

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In the United States, the prevalence of the MS in the adult population was estimated to be more than 25%. Similarly, the prevalence of MS in seven European countries was approximately 23%. The prevalence of metabolic disorders in India is alarming and is increasing exponentially both in the urban and rural areas. Escalation in the prevalence of MS in different parts of India is from 11% to 41%.[2]

Various agencies have put forward the defining criteria for MS; however, the definition of MS by the International Diabetes Foundation in 2005 is the most widely accepted definition.[3] Gender- and ethnicity-specific central obesity is the essential defining criteria for MS along with any two of the four parameters, i.e., raised triglycerides, reduced high-density lipoprotein (HDL) cholesterol, raised blood plasma fasting glucose, and raised blood pressure.

Changing lifestyle and diet have been an important causative factor for the development of MS. Consumption of high

caloric foods containing sugars such as fructose and sucrose has been implicated in the pathogenesis of metabolic disorders. Sucrose has been shown to produce leptin resistance which contributes to the onset and maintenance of obesity.[4,5] Sucrose-rich diet also increases the level oftriglycerides, glucose, and free fatty acids and also impairs glucose tolerance. Sucrose feeding in rats leads to an insulin-resistant state and hyperglycemia.[5]

The term MS may sound simple but itself carries a cluster of metabolic defects and complication. The uncontrolled blood sugar is associated with microvascular and macrovascular complications together with raised blood pressure, and dyslipidemia forms the largest cause of preventable cardiovascular events.

The difficulty lies in the treatment of MS because there is no recognized method to treat the whole syndrome effectively; however, most treatment guidelines aim to treat each component of MS separately, laying more emphasis on the components that are easily modifiable with drugs.

Lifestyle modification primarily by dietary restriction, weight loss, and exercise are first-line treatment considerations. The pharmacotherapy of MS focuses on treating the individual components of MS.

Therapeutic interventions target the treatment of obesity, atherogenic dyslipidemia, hypertension, and elevated fasting plasma glucose. Metformin along with statins and anti-hypertensive medications such as ACE inhibitors and ARB is recommended by the American Diabetes Association as the primary drugs for the treatment of MS. The combined long-term pharmacotherapy is associated with multiple adverse effects and untoward drug interactions.[6] The patient compliance is also poor as there is frequent and multiple dosing schedule. Therefore, there is a need to explore newer molecular entities for the treatment of this syndrome.

Pterocarpus marsupium Roxb. (Fabaceae) is a medium-to-large, deciduous tree that can grow up to 30 m tall. It is native to India, Nepal, and Sri Lanka. In India, it is found in parts of the Western Ghats and in Karnataka and Kerala.[7] *P. marsupium* is widely used in Ayurveda as "Rasayana" for the management of various metabolic disorders such as obesity and diabetes. Recent research has proposed that it possesses blood glucose lowering properties. It is also shown to restore normal insulin secretion by reversing the damage to the beta cells and by repopulating the islets.[8]

The flavonoid phytoconstituents of *P. marsupium* mainly marsupin, pterosupin, and liquiritigenin have shown to have anti-hyperlipidemic effect. The experimental observations proved that the extract was able to reduce serum triglyceride, total cholesterol, low-density lipoproteins, and low-density lipoproteins -cholesterol without any significant effect on the level of HDL cholesterol.[9]

Thus, the present study was conducted to evaluate the protective effect of *P. marsupium* on the body weight, abdominal circumference (AC), blood sugar, and serum triglycerides in animal model of high carbohydrate diet-induced MS.

MATERIALS AND METHODS

Animals

Wistar rats (*Rattus norvegicus*) of either sex weighing 150–250 g and aged about 6–8 weeks were used in the study. They were housed in the Institutional Animal House in polycarbonate cages under temperature, humidity, and light and dark cycle-controlled environment [24 ± 2°C, 60%–70%, 12 h cycle]. Animals were fed on standard pellet diet and water *ad libitum* before the start of experiment.

All the experiments were performed as per the guidelines of Animal Care by Committee for the Purpose of Control and Supervision of Experiments on Animals after the approval from the Institutional Animal Ethics Committee of Era's Lucknow medical college, Lucknow (Approval No: ELMC/PHAR/10).

Collection and Authentication of Plant Extract

The heartwood of *P. marsupium* was collected from a local market of Lucknow, Uttar Pradesh, India. It was dried under shade at a temperature of 35–38°C. The dried sample was authenticated by a botanist at the National Botanical Research Institute, Lucknow.

Extract of *P. marsupium* [10]

The heartwood of PM was grounded in an electric grinder. The powder was soaked in an equal amount of water and stirred intermittently and then left for 36 h. The macerated pulp was then filtered through a fine muslin cloth, and the filtrate was evaporated at 60°C for 2 h to yield a semisolid extract.

The dried aqueous extract was packed in an airtight container and stored at room temperature for further studies.

The dose of *P. marsupium* was 200 mg/kg body weight.[8]

Drugs and Chemical

Sucrose (AsiaChem Pvt. Ltd., Lucknow) was used for the induction of MS. Metformin (Glucophage, Sun Pharma Ltd., Mumbai) and atorvastatin (Aztor, Sun Pharma Ltd., Mumbai) were used as standard treatment. All the drugs and chemicals were obtained from local market.

Induction of MS[11]

Wistar Rats were fed with standard pellet diet along with free access to drinking water containing 20% sucrose *w/v* ad libitum for 8 weeks for the induction of MS. The animals were given normal diet throughout the experiment.

Experimental Protocol

Wistar rats (n = 6 per group) were randomly divided into three groups. Group I (Control group): Animals were given water containing 20% sucrose *ad libitum* for the induction of MS along with distilled water for 8 weeks. Group II (Pterocarpus Group): Animals were given 20% sucrose along with aqueous extract of *P. marsupium* 200 mg/kg/day for 8 weeks. Group III (Standard treatment): Animals were given 20% sucrose along with metformin 100 mg/kg/day and atorvastatin 10 mg/kg/day for 8 weeks.

Estimation of Study Parameters

Weight and AC were measured on day 0, week 4, and weeks 8, with the help of weighing balance and measuring tape, respectively. AC was assessed on the largest zone of the rat abdomen using a plastic non-extensible measuring tape with an accuracy of 0.1 cm.

Animals were fasted overnight, and blood samples were withdrawn by retro-orbital puncture under mild ether anesthesia and centrifuged at 3000 rpm for 15 min, at 4°C in cooling centrifuge. Glucose in serum was estimated by glucose oxidase and peroxidase kit method.

Serum triglycerides was measured by GPO (Glycerol-3-phosphate) method. The intensity of the red color was measured in auto analyzer (Erba Mannheim EM 360).

Statistical Analysis

All values were expressed as mean ± standard error of mean. The data obtained were subjected to statistical analysis using one-way ANOVA followed by Dennett's multiple comparisons test for the control and multiple test groups. All statistical analysis was performed using GraphPad Prism version 6.01. $P < 0.05$ was considered to be statistically significant.

RESULTS

Extract of *P. marsupium*

The extract of *P. marsupium*, prepared as per the method of Rajasekharan *et al*., 1976, gave a yield of 23% (11.52 g from 50 g sun-dried powder). The extract so obtained was brown in color and had a characteristic smell.

Effect on Body Weight

No significant difference in body weight in any of the groups was observed at day 0 ($P = 0.601$). There was a significant increase in the body weight in the control group at week 4 ($P < 0.05$) and at week 8 ($P < 0.0001$).

A significantly lower body weight was observed in the PM group and the standard treatment group after 8 weeks of treatment when compared to the control ($P < 0.01$) [Table 1].

Effect on AC

No significant difference in AC in any of the groups was observed at day 0 ($P = 0.389$). There was an increase in the AC in the control group at week 8 ($P < 0.0001$). There was no significant increase in the AC in the PM group or standard treatment group at week 4 or week 8 when compared to day 0.

A significantly lower AC was observed in the PM group and standard treatment group at 8 weeks of treatment when compared to the control [Table 2].

Effect on Blood Glucose and Triglycerides

All the groups were similar at baseline, and there was no significant difference in blood sugar and serum triglyceride

Table 1: Effect of ***P. marsupium*** on body weight in animal model of MS

Group (n=6 per group)	Day 0		Week 4		Week 8		ANOVA
	Mean	95% CI	Mean	95% CI	Mean	95%CI	
Control (Distilled water)	195.00	167.03–222.97	265.00*	235.50–294.50	328.33*	310.25–346.41	F=44.56 P<0.0001
P. marsupium	203.33	166.57–240.09	223.33	187.18–259.49	226.67#	204.99–248.35	F=1.009 P=0.3879
Standard treatment (Metformin+Atorvastatin)	208.33	183.13–233.54	221.67	200.24–243.09	231.67#	200.24–243.09	F=1.855 P=0.1907
ANOVA	F=0.635 P=0.601		F=3.048 P=0.052		F=34.499 P<0.001		

*P<0.05 comparison to Day 0, #P<0.05 comparison to control. CI: Confidence interval, MS: Metabolic syndrome, *P. marsupium: Pterocarpus marsupium*

levels in any of the groups at baseline ($P = 0.905$). There was an increase in the blood sugar level in the control group at week 4 and week 8 ($P < 0.0001$). No statistically significant increase in the blood sugar was observed in the PM group at weeks 4 and 8 ($P = 0.1019$) when compared to the baseline [Table 3].

Baseline levels of serum triglycerides were similar in all the groups, no significant difference was observed in any of the groups ($P = 0.800$) There was increase in the serum triglycerides in the control group at week 4 ($P < 0.05$) and at week 8 ($P < 0.0001$). There was no significant increase in the serum triglycerides in the PM group or standard treatment group at week 4 or week 8 when compared to the baseline. A significantly lower serum triglyceride levels were observed in the PM group and standard treatment group after 4 and 8 weeks of treatment [Table 4].

DISCUSSION

In the study, we found that the animals fed with water containing 20% sucrose *ad libitum* developed an increase in the body weight, AC, increased blood sugar, and triglyceride levels after 8 weeks of treatment. The animals treated with aqueous extract of *P. marsupium* showed a significant reduction in all the above parameters, and similar results were seen in animals treated with metformin and atorvastatin. These results were in accordance to study done by Sato-Mito *et a*l., 2009[12]

Diet rich in carbohydrates also contributes to the regulation of lipid metabolism partially by acting as inducers of sterol regulatory element-binding protein 1c (SREBP1c) and stearoyl-CoA desaturase.[12] Obesity is associated with an increase in the oxidative stress, and the possible mechanisms contributing to this is increased oxygen consumption through mitochondrial respiratory chain, increased fat deposition, and cell injury causing increased rates of radicals and reactive oxygen species formation such as H_20_2.[13]

Marsupin is the active constituent of *Pterocarpus marsupium* and has been reported to downregulate the adipogenesis-related transcriptional factors PPAR-γ, C/EBP-α, and SREBP-1 and to inhibit adipocyte differentiation during the early stage.[14]

Pterocarpus is also shown to increase lipolysis and triglyceride hydrolysis to diminish fat stores, thereby combating obesity. Marsupin and other phytoconstituents of PM have been shown to act in the gut lumen by forming a covalent bond with the active serine site of gastric and pancreatic lipases by forming the covalent bond, thus inhibiting these lipases from

Table 2: Effect of ***P. marsupium*** on AC in animal model of MS

Group (*n*=6 per group)	Day 0		Week 4		Week 8		ANOVA
	Mean	95% CI	Mean	95% CI	Mean	95% CI	
Control (Distilled water)	12.500	12.031–12.969	13.083	13.096–14.204	14.467*	14.130–14.803	F=30.21 *P*<0.0001
P. marsupium	12.466	11.747–13.187	12.866	12.289–13.444	12.916#	12.460–13.374	F=1.137 *P*=0.3469
Standard treatment (Metformin+Atorvastatin)	12.800	12.094–13.506	12.933	12.391–13.475	12.950#	12.287–13.613	F=0.1089 *P*=0.8976
ANOVA	F=1.058 *P*=0.389		F=2.444 *P*=0.094		F=11.130 *P*<0.0001		

*$P<0.05$ comparison to Day 0, #$P<0.05$ comparison to control. CI: Confidence interval, MS: Metabolic syndrome, *P. marsupium: Pterocarpus marsupium*

Table 3: Effect of ***P. marsupium*** on blood sugar in animal model of MS

Group (*n* = 6 per group)	Day 0		Week 4		Week 8		ANOVA
	Mean	95% CI	Mean	95% CI	Mean	95% CI	
Control	83.167	62.983–103.35	208.67*	146.02–271.32	280.17*	239.13–321.20	F = 32.78 *P* < 0.0001
P. marsupium	87.667	70.297–105.04	132.33	93.588–171.08	120.67#	73.926–167.41	F = 2.669 *P* = 0.1019
Standard treatment (Metformin + Atorvastatin)	88.500	69.218–107.78	103.83	87.622–120.05	133.50*#	98.025–168.97	F = 5.482 *P* = 0.0163
ANOVA	F = 0.185 *P* = 0.905		F = 8.365 *P*= 0.001		F = 34.499 *P*< 0.0001		

*$P < 0.05$ comparison to Day 0, #$P < 0.05$ comparison to control. CI: Confidence interval, MS: Metabolic syndrome, *P. marsupium: Pterocarpus marsupium*

Table 4: Effect of Nigella sativa and ***P. marsupium*** on serum triglyceride in animal model of MS

Group (*n*=6 per group)	Day 0		Week 4		Week 8		ANOVA
	Mean	95% CI	Mean	95% CI	Mean	95% CI	
Control	98.500	86.667–110.33	171.00*	133.64–208.36	174.33*	156.01–192.65	F=19.46 *P*<0.0001
P. marsupium	107.67	85.875–129.46	116.67#	96.459–136.87	116.33#	101.41–131.26	F=0.4668 *P*=0.6358
Standard treatment (Metformin+Atorvastatin)	101.00	85.361–116.64	103.67#	91.060–116.27	101.50#	91.221–111.78	F=0.0782 *P*=0.9251
ANOVA	F=0.335 *P*=0.800		F=11.282 *P*=0.0001		*P*=28.098 *P*<0.0001		

*$P<0.05$ comparison to Day 0, #$P<0.05$ comparison to control. N. sativa: Nigella sativa, CI: Confidence interval, MS: Metabolic syndrome, *P. marsupium: Pterocarpus marsupium*

hydrolyzing the ingested fat into absorbable free fatty acids and monoglycerides.[15]

The potential antidiabetic activity of *P. marsupium* is attributed to one of its active constituents (-), epicatechin, which is shown to have a beta cell restorative and regenerative activity.[16]

The antidiabetic effect of PM may be due to its strong *in vitro* antioxidant activity. It leads to both restoration of activity of antioxidant enzyme levels and reduction in the lipid peroxidation. *Pterocarpus marsupium* leads to inactivation of free radical system without causing free radical scavenging.[17]

The mechanism related to anti-hyperlipidemia effect of PM is not fully understood. Reduction in the levels of triglyceride by downregulating lipid accumulation and upregulating adiponectin expression in the 3T3-L1 adipocyte cells has been proposed.

It has been suggested that PM suppresses adiposity and affects the expression of lipid metabolism genes, especially hepatic expression of the lipid catabolism genes, acyl coenzyme A oxidase 1, palmitoyl, acyl coenzyme-A dehydrogenase, c-4 to c-12 straight chain, and peroxisome proliferator-activated receptor alpha.[18]

The study demonstrated the potential therapeutic benefits of *P. marsupium* in the treatment of various components of MS. The limitations of the study were that, due to the time constraint, we could not discuss the potential therapeutic benefits of *P. marsupium* on the chronic complications of MS.

CONCLUSION

The study concluded that high sucrose diet may lead to the development of various changes associated with MS. There was a reduction in body weight, AC, blood sugar, and serum triglycerides in animals treated with *P. marsupium*. The effect of drug on long-term sequel and complications of MS is yet to be explored before the drug could be used therapeutically in humans.

REFERENCES

1. Reaven GM. The metabolic syndrome: Is this diagnosis necessary? Am J Clin Nutr 2006;83:1237-47.
2. Sawant A, Mankeshwar R, Shah S, Raghavan R, Dhongde G, Raje H, *et al*. Prevalence of metabolic syndrome in Urban India. J Cardiovasc Dis Res 2012;3:204-11.
3. Thresher JS, Podolin DA, Wei Y, Mazzeo RS, Pagliassotti MJ. Comparison of the effects of sucrose and fructose on insulin action and glucose tolerance. Am J Physiol Regul Integr Comp Physiol 2000;279:R1334-40.
4. IDF Worldwide Definition of the Metabolic Syndrome. International Diabetes Federation; 2017.
5. Shapiro A, Mu W, Roncal C, Cheng KY, Johnson RJ, Scarpace PJ, *et al.* Fructose-induced leptin resistance exacerbates weight gain in response to subsequent high-fat feeding. Am J Physiol Regul Integr Comp Physiol 2008;295:R1370-5.
6. Grundy SM, Hansen B, Smith SC Jr., Cleeman JI, Kahn RA, American Heart Association. *et al.* Clinical management of metabolic syndrome: Report of the American heart association/National heart, lung, and blood institute/American diabetes association conference on scientific issues related to management. Arterioscler Thromb Vasc Biol 2004;24:e19-24.
7. Devgan M, Nanda A, Ansari SH. Comparative evaluation of the anti-diabetic activity of *Pterocarpus marsupium* roxb. Heartwood in alloxan induced diabetic rats using extracts obtained by optimized conventional and non-conventional extraction methods. Pak J Pharm Sci 2013;26:973-6.
8. Vats V, Grover JK, Rathi SS. Evaluation of anti-hyperglycemic and hypoglycemic effect of *Trigonella foenum-graecum* Linn, *Ocimum sanctum* Linn and *Pterocarpus marsupium* Linn in normal and alloxanized diabetic rats. J Ethnopharmacol 2002;79:95-100.
9. Jahromi MA, Ray AB. Antihyperlipidemic effect of flavonoids from A*Pterocarpus marsupium.* J Nat Prod 1993;56:989-94.
10. Rajasekharan S, Tuli SN. Vijayasara, *Pterocarpus marsupiumin* the treatment of madhumeha (diabetes mellitus)-a clinical trial. J Res Indian Med Yoga Homeo 1976;11:9-14.
11. Pagliassotti MJ, Prach PA, Koppenhafer TA, Pan DA. Changes in insulin action, triglycerides, and lipid composition during sucrose feeding in rats. Am J Physiol 1996;271:R1319-26.
12. Sato A, Kawano H, Notsu T, Ohta M, Nakakuki M, Mizuguchi K, *et al.* Antiobesity effect of eicosapentaenoic acid in high-fat/high-sucrose diet-induced obesity: Importance of hepatic lipogenesis. Diabetes 2010;59:2495-504.

13. Miyazaki M, Dobrzyn A, Man WC, Chu K, Sampath H, Kim HJ, *et al*. Stearoyl-coA desaturase 1 gene expression is necessary for fructose-mediated induction of lipogenic gene expression by sterol regulatory element-binding protein-1c-dependent and -independent mechanisms. J Biol Chem 2004;279:25164-71.
14. Begriche K, Massart J, Robin MA, Borgne-Sanchez A, Fromenty B. Drug-induced toxicity on mitochondria and lipid metabolism: Mechanistic diversity and deleterious consequences for the liver. J Hepatol 2011;54:773-94.
15. Chien PJ, Chen YC, Lu SC, Sheu E. Dietary flavonoids suppress adipogenesis in 3t3-l1 preadipocytes. J Food Drug Anal 2005;13:168-75.
16. Cannon B, Nedergaard J. Brown adipose tissue: Function and physiological significance. Physiol Rev 2004;84:277-359.
17. Chakravarthy BK, Saroj G, Gambhir SS, Gode KD. Pancreatic beta cell regeneration–a novel antidiabetic mechanism of *Pterocarpus marsupium* roxb. Indian J Pharmacol 1980;12:123.
18. Manjunatha HD. Evaluation of antimicrobial activity and antioxidant activity by electrochemical method of ethanolic extract of *Pterocarpus marsupium* Roxb bark. J Biotechnol Biomater 2014;4:1.

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