Original Article **Open Access**

Weight Reducing Effect of Moringa Oleifera Leaves Against Induced Obesity in Rabbits

Syed Saqib Khalid, Tabassum Zehra, Fizzah Ali, Fizza Batool

ABSTRACT

Objectives: To study the weight-reducing effect of M. Oleifera Leaves against induced obesity in rabbits.

Study Design and Settings: It was an animal study conducted in the Department of Pharmacology, Liaquat National Hospital & Medical College, Karachi.

Methodology: The study period was 6 months, and random sampling was done. Twenty four Healthy rabbits of either sex, weighing 1–2 kg and aged 1-3 years, were included in the study. Sick rabbits weighing less than 1 kg or more than 2 kg and aged less than 1 year or more than 3 years were excluded from the study.

Results: The weight of Group B animals fed with a high-fat diet (HFD) for 28 days increased significantly from 1.28±0.07 to 1.88 ± 0.17. While the weight of Group C and D rabbits which were fed with M. oleifera extract(300mg/kg and 600mg/kg) along with a HFD for 28 days, increased slightly from 1.38±0.07 to 1.53± 0.08 and from 1.25±0.13 to 1.38± 0.14 respectively. P-value of weight on Day 28 also became significant with a P-value of (0.000), which was (0.176) on Day 0, proving the anti-obesity effects of M. oleifera leaves.

Conclusion: In our study, we found out that there was a significant weight-reducing effect of M. oleifera leaves against obesity induced rabbits as well as skin fold thickness was also decreased. This study also indicates that the given dose of M. oleifera leaves extracts are devoid of any side effects.

Keywords: Body mass index, High-fat diet, Moringa oleifera, Obesity, World Health Organization

How to cite this Article:
Khalid SS, Zehra T, Ali F, Batool F. Weight Reducing Effect of Moringa Oleifera Leaves Against Induced Obesity in Rabbits. J Bahria Uni Med Dental Coll. 2023;13(4):266-70 DOI: https://doi.org/10.51985 /JBUMDC2022143

INTRODUCTION:

Today, obesity has become a serious health concern and a possible risk for many diseases worldwide. World Health Organization (WHO) defined obesity as having a body mass index of 30 kg/m2 or more. A Body Mass Index over the normal range of 18 to 25 kg/m^2 is typical in Western societies. It has been connected to inactive lifestyles and Western food intake (rich in saturated fat and calories).[1]

The National Heart, Lung, and Blood Institute's "Pharmacological Management of Obesity: An Endocrine Society Clinical Practice Guideline" states that moderate-intensity exercise combined with suitable lifestyle and dietary adjustments should be the preferred method of weight loss treatment. However, epidemiological and clinical research has demonstrated how difficult it is to sustain a long-term healthier lifestyle. Therefore, much research has been done on natural supplement items that primarily aid people in their fight against obesity.[2]

Reports indicate that the biological activity of M. oleifera includes prevention of gastric ulcers, reducing blood glucose levels, decreasing blood pressure, and anti-inflammatory qualities. Moringa oleifera leaves protect against a wide range of potentially dangerous illnesses, including bacterial growth, cancer, oxidative stress, inflammation, hepatic fibrosis, liver damage, and hypercholesterolemia. It has also been shown to improve regulation of hormones in the kidneys, liver, and thyroid.[3]

Moringa oleifera also have beneficial effects on brain as one study shows that it can protect male wistar rats against ketamine-induced memory problems, and NMDA receptors may play a role in this protective action of M. oleifera.[4]

Research also shows that mice performed better during reproduction when they consumed Moringa oleifera leaves. Dietary Moringaoleifera leaves lowered serum

Syed Saqib Khalid *(Corresponding Author)*
Lecturer, Department of Pharmacology
Liaquat National Hospital and Medical College, Karachi
Email: saqibkhalid32@gmail.com

Tabassum Zehra
Professor & HOD, Department of Pharmacology,
Liaquat National Hospital and Medical College, Karachi
Email: drtabassum.zehra@lnh.edu.pk

Fizzah Ali
Assistant Professor, Department of Pharmacology,
Liaquat National Hospital and Medical College, Karachi
Email: dr.fizzah.ali@lnh.edu.pk

Fizza Batool
Lecturer, Department of Pharmacology,
Liaquat National Hospital and Medical College, Karachi
Email: fizza.batool@lnh.edu.pk

Received: 03-11-2022
Accepted: 12-09-2023

Malondialdehyde, but more significantly, decreased the frequency of defective sperm.[5]

In mice, the polysaccharide from Moringaoleifera leaves has been shown to modulate the intestinal microbial composition by increasing the abundance of bacteria linked to weight reducing effects, short chain fatty acid generation, and lactic acid production. These results suggest that the polysaccharide found in M. oleifera leaves may be a useful prebiotic with health-enhancing properties.[6]

M. oleifera also has garnered significant attention as a chemotherapeutic approach due to possible anti-inflammatory and antioxidant properties, and higher flavonoid content compared to other vegetables and fruits.[7]

The major metabolic enzymes that are linked to obesity are pancreatic lipase, α-amylase, and α-glucosidase, which were strongly suppressed by M. oleifera leaves. Therefore, M. oleifera leaves may be considered a readily available health supplement source for reducing obesity.[8]

This study outlined an anti-obesity function of M. oleifera leaves powder in experimentally induced obesity in rabbits, a species that has recently been studied for hypocholesterolemic action of M. oleifera.

The purpose of this study was to determine whether M. oleifera leaf had any therapeutic value for treating obesity in rabbits.

Some pharmacological drugs are used to treat obesity, but each drug has their undesirable side effects and slow onset of action, and some are expensive. If the weight-reducing effects of M. Oleifera leaves are established, they can replace the pharmacological drugs for treating obesity.

METHODOLOGY:

It was an animal study performed in the Department of Pharmacology along with Animal House, Liaquat National Hospital & Medical College, Karachi. The study duration was six months from 1st January 2022 to 30th June 2022 after approval from the research and ethical committee. Ethical permission for the current study was taken by the Ethical Review Committee of Liaquat National Hospital & Medical College, Karachi. (Ref: App # 0658-2021 LNH – ERC). Data that were obtained during the study was kept extremely confidential.

Sample size was 24 Rabbits. Sample size was calculated by using below formula.

Minimum numbers of animals per group:

Minimum n = 10/k + 1

n = number of subjects per group and k = number of groups

n = 10/4 + 1

n = 2.5 + 1 = 3.5

Total Minimum numbers of animals required:

Minimum N = Minimum n x k

Minimum N = 3.5 x 4= 14

Random sampling was done and 24 Healthy rabbits of either sex, Weighing 1 – 2 kg and Aged 1 - 3 years, were included in the study. Sick Rabbits Weighing less than 1 kg or more than 2 kg and aged less than one year or more than three years were excluded from the study. Rabbits were divided into groups A, B, C, and D, with six animals in each group.

Group A: represent the standard control in which the rabbits were given a regular diet and free access to water for 28 days.

Group B: represent a negative control in which the rabbits were given a high-fat diet for 28 days.

Group C: represent a test treatment in which rabbits were given M. oleifera extract(300mg/kg) along with a high-fat diet for 28 days.

Group D: represents test treatment in which rabbits were given M.oleifera extract (600mg/kg) along with a high-fat diet for 28 days.[10]

Previous reports of an acute toxicity investigation using a single dosage of orally given 2 g/kg extracts of M. oleifera(leaf) indicate that these concentrations which are used in our study are safe to use.

The body weight, skin fold thickness, and body temperature were recorded on day 0 and then weekly consecutively for 28 days.

Fine powder of M. oleifera leaves (Stock Number: 124876300_PK-1282732171) was brought from Moringa Powder Store Pakistan and stored in a clean, sterile glass container. 300mg and 600 mg of M. oleifera leaf powder/rabbit/day were administered with a high-fat diet according to the experimental design.

Rabbits that were fed with a regular diet include hay which was brought from the local market on an everyday basis.

High-fat diet induced obesity in rabbits is considered a reliable tool for evaluating anti-obesity activity. A high-fat diet includes 10% fat (2/3 corn oil and 1/3 coconut oil) added to the regular rabbit diet.

Statistical analysis: Data was analyzed on SPSS version 22.0 (IBM, incorporation, USA). The results were expressed as means ± standard deviation. One way ANOVA test is applied. Statistical significance was taken at $P<0.05$.

RESULTS:

Body weight, skin fold thickness and temperature of animals, of all groups were analyzed and compared on Day 0, 7, 14, 21 and 28.

The weight of Group B animals fed with a high fat diet for 28 days increased significantly from 1.28±0.07 to 1.88 ± 0.17. While the weight of Group C and D rabbits which were fed with M. oleifera extract(300mg/kg and 600mg/kg) along with a HFD for 28 days, increased slightly from

1.38±0.07 to 1.53± 0.08 and from 1.25±0.13 to 1.38± 0.14 respectively. P-value of Weight on Day 28 also became significant with a P-value of (0.000), which was (0.176) on Day 0, proving the anti-obesity effects of M. oleifera leaves.

Skin fold thickness also increased significantly from 1.08±0.07 to 1.45 ±0.20 in Group B animals fed with a HFD for 28 days. There was only a slight increase in Skin fold thickness from 1.16±0.10 to 1.23± 0.10 and from 1.16±0.08 to 1.25± 0.08, respectively, in Group C and D animals fed with M. oleifera extract(300mg/kg and 600mg/kg) along with HFD for 28 days. The P-value of Skin fold thickness on Day 28 was close to significant with a P-value of (0.061), which was (0.411) on Day 0.

There was a noteworthy drop in Body temperature from 39.08±0.33 to 37.18± 0.22 in Group B animals fed with a HFD for 28 days, while there was a rise in Body temperature from 38.41±0.16 to 39.18.± 0.17 and from 38.63±0.31 to 39.28± 0.09 in Group C and Group D animals fed with M. oleifera extract(300mg/kg and 600mg/kg) along with a HFD for 28 days. P-value of Body temperature on Day 28 also became significant with a P-value of (0.000), which was (0.006) on Day 0, indicating that intake of M. Oleifera leaves can increase body temperature slightly.

DISCUSSION:

Obesity has been declared a worldwide epidemic by WHO.[13] It has become a serious health concern and a possible risk for many diseases worldwide. When a person's amount of fat tissue increases to the point that it negatively impacts their mental and physical well-being and shortens their life span, they are said to be obese. Recent studies showed that interaction of genetic, environmental, psycho-behavioral, hormonal, metabolic, cultural, and socioeconomic elements might contribute to the development of obesity. The incidence of the world's main severe illnesses and primary causes of death is dramatically increased by obesity. Severe obesity is often associated to a number of other serious medical conditions. These include high cholesterol, high blood pressure, diabetes type II, gallstones, fatty liver disease, pulmonary hypertension, sleep apnea, and even certain forms of cancer.[14]

A study of the available literature reveals that several different herbal plants are used in the treatment of obesity. These include fucus vesiculosus, citrus aurantium, yacon syrup, curcumin, nigella sativa, camellia synensis, green tea, and black Chinese tea.[1]

Vitamins, phenolic acids, flavonoids, isothiocyanates, tannins, and saponins are some of the many bioactive components found in substantial concentrations throughout different M. oliefera plant parts. They've all been researched for their health benefits.[15]

This study outlined an anti-obesity function of M. oleifera leaves powder in experimentally induced obesity in rabbits, a species that has recently been studied for hypocholesterolemic action of M. oleifera.

Study conducted by (Nahar et al., 2016) revealed that Body weight, thoracic (TC) and abdominal (AC) circumferences, and body mass index, all increased significantly in rats fed a high-fat diet. A comparable research found that HFD animals gained much more weight than pellet-fed animals (Bais, Singh, & Sharma, 2014). Our findings, in which Group B animals given an HFD for 28 days had their weight grow dramatically from 1.280.07 to 1.880.17 kgs, are supported by these investigations.

Research from (Nahar et al., 2016) also found that both single-dose and repeated-dose administrations of M. oleifera leaf powder successfully decreased body mass index (BMI) in obese participants which is in accordance with our study in which Groups C and D treated with M. oleifera leaves

Table 1: Mean differences among different group with respect to weight, skin fold thickness and body temperature

Parameters		Group A	Group B	Group C	Group D	P-value
Weight	Day 0	1.28±0.11	1.28±0.07	1.25±0.13	1.25±0.13	0.176
	Day 7	1.33±0.15	1.38±0.13	1.28±0.11	1.28±0.11	0.496
	Day 14	1.38±0.13	1.45±0.13	1.31±0.11	1.31±0.11	0.150
	Day 21	1.38± 0.13	1.65±0.19	1.33±0.15	1.33±0.15	0.006
	Day 28	1.45±0.13	1.88± 0.17	1.38±0.14	1.38±0.14	0.000
Skin fold thickness	Day 0	1.18±0.16	1.08±0.07	1.16±0.08	1.16±0.08	0.411
	Day 7	1.21±0.11	1.20±0.08	1.23±0.10	1.23±0.10	0.927
	Day 14	1.25±0.12	1.28±0.11	1.26±0.08	1.26±0.08	0.363
	Day 21	1.26±0.12	1.31±0.17	1.28±0.07	1.28±0.07	0.564
	Day 28	1.28±0.14	1.45±0.20	1.25±0.08	1.25±0.08	0.061
Body temperature	Day 0	38.78±0.31	39.08±0.33	38.63±0.31	38.63±0.31	0.006
	Day 7	39.13±0.20	39.46±0.45	38.90±0.24	38.90±0.24	0.02
	Day 14	39.16±0.26	38.96±0.67	39.10±0.15	39.10±0.15	0.437
	Day 21	38.90±0.46	38.06±0.16	39.03±0.08	39.03±0.08	0.000
	Day 28	38.86±0.43	37.18±0.22	39.28±0.09	39.28±0.09	0.000

extract at 300 and 600mg dosages, respectively, had only a modest rise in weight in comparison to an obese control group in which the weight increase was highly significant.

Additionally a similar study reported that the use of a HFD result in a considerablerise in body weight. The percentage of gain in weight of body of rats fed with the HFD supplemented with extract of M. oleifera leaves was notably lesser than that in the HFD group, which coincides with our study in which P-value of Weight at Day 28 becomes significant (0.000) which was (0.176) at Day 0 proving anti-obesity effects of M. oleifera leaves.[16]

The study performed by Irfan et al., 2016 evaluated that the diabetic rats fed with M. oleifera leaf extract produced an utmost loss in body weight on the 7th day of study. Moreover, body weight loss at the completion of oral doses was progressively declining, which supports our study.[17]

Another study conducted by Madkhali H et al., 2019 revealed when HFD was given to animals, it resulted in a considerable rise in body weight. On the other hand, body weight was considerably decreased in HFD-induced obese rats that were given M. oleifera leaves extract in a dose-dependent manner, which is in accordance with our study.[18]

The study conducted by Othman A et al., 2019 support our study and provides evidence that HFD rats demonstrate a marked rise in weight of body compared to animals given normal diet, and two weeks of M. oleifera extract therapy noticeably decreases weight of body compared to HFD supplemented rats.[10]

Kilany O et al., 2020 observed that administration of M. oleifera seed oil extract to rats significantly decreased body weight; in contrast, in our study, M. oleifera leaves extracts were used instead of M. oleifera seed oil.[19]

The HFD group showed statistically significant weight gain in a research by Dhungel et al., 2009. According to their findings, those who followed the HFD gained about 24% more weight than those who followed the Low Fat Diet, and those on the High Fat Diet also had increases in skinfold thickness (SFT) in the fourth and tenth weeks of the research. Consistent with these findings, we found that skin fold thickness increased significantly from 1.08 ±0.07 to 1.45 ±0.20 in Group B animals given the high-fat diet.

Research done by Bais S et al., 2014found that rats getting a HFD showed a significant decrease in rectal body temperature observed on Day 49 while giving M. oleifera extract (200 and 400mg/kg) dose-dependently increased the body temperature. Our study is in accordance with this study, and there was a considerable drop in Body temperature from 39.08±0.33 to 37.18± 0.22 in Group B animals fed with a HFD while there was a rise in Body temperature from 38.41±0.16 to 39.18.± 0.17 and 38.63±0.31 to 39.28± 0.09 in Group C and Group D animals, respectively, fed with M. oleifera extract(300mg/kg and 600mg/kg).[20]

Further, studies are carried out to determine the active principle of this plant, followed by the identification of the mechanistic approach of M. oleifera leaf powder that helps in weight management.

CONCLUSION:

Many pharmaceutical methods have been explored for treating obesity, but only a few are safe, and the majority have undesirable side effects. The search for plant-based anti-obesity medications is, therefore, an option. Based on our findings, M. oleifera leaves significantly reduced body weight and skin fold thickness in obese rabbits. This research also shows that the recommended dose of M. oleifera leaves extract is safe and without side effects.

Authors Contribution:
Syed Saqib Khalid: developed the protocol of study and carried out data collection.
Tabassum Zehra: conducted literature review.
Fizzah Ali: assisted in data collection.
Fizza Batool: contributed in statistical analysis.

REFERENCES:

1. Nahar S, Faisal F, Iqbal J, Rahman M, Yusuf DMA. Antiobesity activity of Moringa oleifera leaves against high fat diet-induced obesity in rats. International Journal of Basic and Clinical Pharmacology. 2016:1263-8. DOI: https://doi.org/10.18203/2319-2003.ijbcp20162427
2. Aziza S, Elsenosi Y, Ghada H, El S, Hussein S. Moringa Oleifera improve lipid metabolic disorders in obesity induced oxidative stress in rats. Benha Veterinary Medical Journal. 2019;35. https://doi.org/10.21608/BVMJ.2018.96443
3. Vergara-Jimenez M, Almatrafi MM, Fernandez ML. Bioactive Components in Moringa Oleifera Leaves Protect against Chronic Disease. Antioxidants (Basel, Switzerland). 2017;6(4). doi: 10.3390/antiox6040091
4. Afrin S, Hossain A, Begum S. Effects of Moringa oleifera on working memory: an experimental study with memory-impaired Wistar rats tested in radial arm maze. BMC Research Notes. 2022;15(1):314. DOI: 10.1186/s13104-022-06219-5
5. Zeng B, Luo J, Wang P, Yang L, Chen T, Sun J, et al. The beneficial effects of Moringa oleifera leaf on reproductive performance in mice. Food Science & Nutrition. 2019;7(2):738-46. doi: 10.1002/fsn3.918
6. The Beneficial Effects of a Polysaccharide from Moringa oleifera Leaf on Gut Microecology in Mice. Journal of Medicinal Food. 2019;22(9):907-18. DOI: 10.1089/jmf .2018.4382
7. Lin M, Zhang J, Chen X. Bioactive flavonoids in Moringa oleifera and their health-promoting properties. Journal of Functional Foods. 2018;47:469-79. https://doi.org/10.1016/j.jff.2018.06.011
8. Swamy GM, Meriga B. Therapeutic effect of Moringa oleifera leaf extract s on oxidative stress and key metabolic enzymes related to obesity. Internationa l Journal of Pharmaceutical Sciences and Research. 2020;11(4):1949-57. DOI: 10.13040/IJPSR.0975-8232.11(4).1949-57
9. Arifin WN, Zahiruddin WM. Sample Size Calculation in Animal Studies Using Resource Equation Approach. The Malaysian journal of medical sciences : MJMS. 2017;24(5):101-5. DOI: 10.21315/mjms2017.24.5.11

10. Othman A, Amer M, Basos A, El-Missiry MA. Moringa oleifera leaf extract ameliorated high-fat diet-induced obesity, oxidative stress and disrupted metabolic hormones. Clinical Phytoscience. 2019;5. DOI https://doi.org/10.1186/s40816-019-0140-0

11. Jacob J, Amalraj A, Divya C, Janadri S, Manjunatha PM, Gopi S. Oral toxicity study of sports nutritional powder in Wistar rats: A 90 day repeated dose study. Toxicology reports. 2018;5:497-503. https://doi.org/10.1016/j.toxrep.2018.04.001

12. Dhungel S, Sinha R, Sinha M, Paudel BH, Bhattacharya N, Mandal M. High fat diet induces obesity in British Angora Rabbit: A model for experimental obesity. Indian journal of physiology and pharmacology. 2009;53:55-60.

13. El-Shehawi AM, Alkafafy M, El-Shazly S, Sayed S, Farouk S, Alotaibi S, et al. Moringa oleifera leaves ethanolic extract ameliorates high fat diet-induced obesity in rats. Journal of King Saud University-Science. 2021;33(6):101552. DOI https://doi.org/10.1186/s40816-019-0140-0

14. Ahmed H, Metwally F, Rashad H, Zaazaa A, Ezzat S, Salama M. Moringa oleifera offers a multi-mechanistic approach for management of obesity in rats. 2014;29:98-106.

15. Vergara-Jimenez M, Almatrafi MM, Fernandez ML. Bioactive Components in Moringa Oleifera Leaves Protect against Chronic Disease. Antioxidants. 2017;6(4):91. DOI: 10.3390/antiox6040091

16. Mabrouki L, Rjeibi I, Taleb J, Zourgui L. Cardiac Ameliorative Effect of Moringa oleifera Leaf Extract in High-Fat Diet-Induced Obesity in Rat Model. BioMed Research International. 2020;2020:1-10. DOI: 10.1155/2020/6583603

17. Irfan H, Abdullah M, Khan N, Sadikun A. Effect of Ethanolic Extract of Moringa oleifera Lam. Leaves on Body Weight and Hyperglycemia of Diabetic Rats. Pakistan Journal of Nutrition. 2016;15:112-7. DOI: 10.3923/pjn.2016.112.117

18. Madkhali H, Alharthy M, Asiri M, Ganaie M, Ansari M, Rehman NU, et al. Moringa oleifera Lam. (family Moringaceae) leaf extract attenuates high-fat diet-induced dyslipidemia and vascular endothelium dysfunction in Wistar albino rats. 2019;18:2597-604. DOI: 10.4314/tjpr.v18i12.20

19. Kilany O, Abdelrazek H, Aldayel T, Abdo S, M.A.Mahmoud M. Anti-obesity potential of Moringa olifera seed extract and lycopene on high fat diet induced obesity in male Sprauge Dawely rats. Saudi Journal of Biological Sciences. 2020;27. https://doi.org/10.1016/j.sjbs.2020.06.026

20. Bais S, Singh G, Sharma R. Antiobesity and Hypolipidemic Activity of Moringa oleifera Leaves against High Fat Diet-Induced Obesity in Rats. Advances in Biology. 2014;2014:1-9. https://doi.org/10.1155/2014/162914