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Effect of *Mimosa Himalayana* Extract on Potassium Oxonate Induced Hyperuricemia in Rats

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ABSTRACT

Objective: To determine the hypouricemic effect of *Mimosa himalayana* extract on potassium oxonate induced hyperuricemia in rats.

Study Design: Laboratory-based Experimental study.

Place and Duration of Study: Pharmacology Department, University of Health Sciences Lahore Pakistan, from Jan 2017 to Dec 2018.

Methodology: Forty-eight healthy male Wistar rats were placed randomly in 6 Groups (n=8). Group-I served as negative control while hyperuricemia was induced in Groups II, III, IV, V and VI with potassium oxonate 250 mg/kg intraperitoneally on day 1st, 3rd and 7th. Group-II served as positive control while Group III received allopurinol 5 mg/kg body weight orally once daily for seven days. Groups IV, V and VI were treated with *Mimosa himalayana* extract 100, 200 and 400 mg/kg body weight by gavage once daily for 7 days respectively. Oral acute toxicity was observed in eighteen rats for three days with a dose of 1000 and 2000 mg/kg body weight. On day seven, all animals were sacrificed, and blood was collected for serum separation. Serum uric acid was measured with the help of chemistry analyzer.

Results: *Mimosa himalayana* extract showed no signs of acute toxicity in rats with 1000 mg/kg and 2000 mg/kg oral dose. Hyperuricemia was successfully induced with potassium oxonate. Treatment with allopurinol (5 mg/kg) markedly reduced serum uric acid levels (p<0.05). *Mimosa himalayana* extract (100mg/kg, 200mg/kg and 400mg/kg) also reduced serum uric acid levels significantly (p<0.05).

Conclusion: *Mimosa himalayana* extract possesses significant hypouricemic effect.

Keywords: Hyperuricemia, *Mimosa himalayana* extract, Potassium oxonate, Uric Acid.

How to Cite This Article: *Shaheen B, Fatima J, Qammar SM, Malik T, Batool S, Noureen S. Effect of Mimosa Himalayana Extract on Potassium Oxonate Induced Hyperuricemia in Rats. Pak Armed Forces Med J 2024; 74(4): 984-988. DOI: https://doi.org/10.51253/pafmj.v74i4.6441*

INTRODUCTION

Hyperuricemia (HU) is abnormally high serum urate (SUA) levels. The upper normal limit of SUA levels in males is approximately 7 mg/dl and in females, 6 mg/dl.[1] SUA levels and prevalence of HU have been increased in the past 40 years both in developed and underdeveloped countries.[2] Hyperuricemia is a common metabolic disease which can remain asymptomatic or can precede to accumulation of monosodium urate (MSU) crystals in the joints predisposing to gout and nephrolithiasis in kidneys.[3] A positive association has been found between SUA levels and various disorders like hypertension, congestive cardiac failure, type-2 diabetes mellitus, chronic renal disease and metabolic syndrome.[4]

Human beings and higher primates cannot oxidize uric acid because of nonsense mutation of the enzyme uricase, resulting in higher urate levels in humans.[5] Human serum uric acid levels are maintained within normal limit by the balance between synthesis within the liver and elimination via gut and kidneys. Any imbalance in this results in hyperuricemia. Major causes of hyperuricemia include purine rich sea food, overproduction of purines, tumor lysis syndrome, under excretion of uric acid and various drugs (low dose salicylates, thiazide and loop diuretics, pyrazinamide and ethambutol). Xanthine oxidase inhibitors (allopurinol, febuxostat) and uricosuric drugs are the mainstay of treatment but adverse effects and drug interactions associated with them limit their use.[6]

Botanical products have long been used because of their safety and cost effectiveness. *Mimosa himalayana* (*M. himalayana*) from Leguminosae family, is a large deciduous shrub. It has been utilized in traditional medicine for the treatment of leukoderma, rheumatism, chronic diarrhea and fungal infections. Previous studies showed that *M. himalayana* possesses anti-fungal, antibacterial and anti-neoplastic activity.[7]

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Received: 10 Mar 2021; revision received: 05 Jan 2023; accepted: 10 Jan 2023

According to phytochemical analysis of MHE it is rich in flavonoids like quercetin (65.38%) and luteolin (2.5%).[8] These flavonoids possess xanthine oxidase inhibitory effect.[9] With limited drugs available for hyperuricemia, it was planned to see the effect of M. himalayana on potassium oxonate induced hyperuricemia in rats.

METHODOLOGY

The laboratory-based study was conducted at the Pharmacology Department of University of Health Sciences (UHS), Lahore Pakistan after approved from Ethical Review Committee for Medical and Biomedical Research, UHS, (No.UHS/Education/126-16/1040) from January 2017 to December 2018.

Inclusion Criteria: Adult healthy male Wistar albino rats were included.

Exclusion Criteria: Rats with any obvious injury and disease were excluded.

Fresh flowering shoots were obtained from the surroundings of Quaid-e-Azam University, Islamabad on 24th February 2017. The whole plant was thoroughly washed, and shade dried. Later on, it was macerated in ethyl alcohol 95% for 3 days. The filtrate was concentrated under reduced pressure at 34˚C temperature with the help of rotary evaporator and freeze dried at - 44oC using lyophilizer.[11] *Mimosa himalayana* extract was labeled as MHE and stored at 4˚C temperature till further utilization.

Sixty-six adult healthy male Wistar albino rats were collected from the Animal Facility of University of Health Sciences, Lahore, and placed in Experimental Research Laboratory under controlled conditions.[9] Animals were kept in conventional cages with woodshed bedding. To identify the animals each cage was properly labeled. Environment of rats was maintained with temperature ranged 25±5˚C and twelve hours day and night cycle. Animals were given standard rodent chow and free access to water during the whole study. Sixty-six animals were further divided in two Groups with 18 animals in one Group for oral acute toxicity and 48 rats in second Group for in vivo study. Eighteen adult healthy male rats were randomly placed in three Groups (n=6) for oral acute toxicity. Group-A received 1 ml distilled water while Group B and C received MHE 1000 mg/kg and 2000 mg/kg body weight once by gavage respectively. The animals were closely observed for first 3 hrs, then at an interval of 6 hrs. during next 48 hrs for any obvious change in behavior with other animals, alertness, food intake, change in body weight, consistency of fecal matter and mortality.[10]

Forty-eight, adult, healthy male albino rats were placed randomly in 6 Groups (n=8). To observe the hypouricemic effect of MHE in animals, hyperuricemia was induced with the help of potassium oxonate (PO), which is a uricase inhibitor. All animals except Group-I received PO 250 mg/kg dissolved in distilled water intraperitonially 1 hour before oral dose of test compounds on day 1, 3 and 7 of the study.[11,12] Test samples of MHE and allopurinol were freshly prepared in distilled water each time before administration.

Group-I (Negative control) get distilled water (1ml) orally one time a day for seven days. Group-II animals (Hyperuricemic control) were given 1ml pure water by gavage one time a day for seven days in addition to PO intraperitoneally on day 1st, 3rd and 7th. Group III animals received allopurinol 5 mg/kg body weight orally for seven days while intraperitoneal injections of PO were given on 1st, 3rd and 7th days of the study.[13] Group IV, V and VI received MHE 100 mg/kg, 200 mg/kg and 400 mg/kg body weight once daily by gavage for seven days in addition to PO intraperitoneally on day 1st, 3rd and 7th.[14]

The tail veins of rats were used for blood sampling on day 0, 1 and 3 of the study. At the end of study on seventh day, rats were anesthetized three hours after the last dose and blood was obtained through intra cardiac puncture. Sera was separated and stored at –20˚C till further measurements of uric acid.[15]

Uric acid concentration of the blood was checked by Dry chemistry analyzer on day 0, 1 and 3 of the study. On seventh day, serum urate was measured by chemistry analyzer, using a commercially available diagnostic kit compatible to the instrument.

Statistical Package for Social Sciences (SPSS) version 22.0 was used for the data analysis. Quantitative variables with normal distribution were expressed as Mean±SD and qualitative variables were expressed as frequency and percentages. One-way analysis of variance (ANOVA) was applied to gauge the mean differences among the groups. The group differences were calculated using Post Hoc test (Tukey HSD).

RESULTS

In the present study, we initially observed oral acute toxicity of MHE in rats. Group-I was negative

control and received distilled water only. Groups II and III received MHE as 1000mg/kg and 2000mg/kg body weight respectively. We observed changes in behavior, alertness, food intake, consistency of fecal matter, body weight and mortality of the animals before and after treatment. None of the animals included in the study revealed signs and symptoms of acute toxicity. Only male rats were taken for the study. Average weight of animals in each Group was comparable with mean weight of 181g. Animals in Group-I had 178.8±12.2 g weight before treatment while 179.3±12.8g after treatment. Animals in Group-II had 183.8±12.7 and 184.3±12.5 g weight before and after treatment consecutively. Similarly in Group-III average weight of the animals was not significantly changed before and after treatment. MHE had no adverse effects on the behavior and food intake of the animals in Group-II and Group-III receiving 1000 mg/kg and 2000 mg/kg body weight MHE. No animal showed change in consistency of fecal matter in all Groups. Mortality with MHE was zero even at 2000 mg/kg dose. The expected oral lethal dose 50% (LD_{50}) was more than 2000 mg/kg. Results of oral acute toxicity are expressed in Table-I.

Table-I: Effect of *Mimosa Himalayana* Extract (MHE) on Various Parameters during Oral Acute Toxicity in Rats (n=48)

Parameters	Group-I (Negative control)		Group-II (MHE 1000 mg/kg)		Group-III (MHE 2000 mg/kg)	
	Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment
Behavior with other animals	Normal	Unchanged	Normal	Unchanged	Normal	Unchanged
Alertness	Active	Active	Active	Active	Active	Active
Food intake	35 g	34 g	33 g	30 g	37 g	33 g
Body weight (g)	178.8±12.2	179.3±12.8	183.8±12.7	184.3±12.5	181.7±10.7	182±10.4
Consistency of fecal matter	Pellets	Unchanged	Pellets	Unchanged	Pellets	Unchanged
Mortality	Alive	None	Alive	None	Alive	None

** results of body weight were presented as Mean±S.D (n=6)*

Table-II: Effect of *Mimosa Himalayana* Extract on Serum Urate Levels in Hyperuricemic Rats Induced by Potassium Oxonate (n=48)

Days	Group-I (Negative Control Group)	Group-II (Positive Control Group)	Group-III (Standard drug)	Group-IV (MHE 100 mg/kg)	Group-V (MHE 200 mg/kg)	Group-VI (MHE 400 mg/kg)	*p*-value
Zero day	2.36±0.19	2.46±0.2	2.46±0.2	2.4±0.18	2.47±0.17	2.47±0.24	<0.01
1st day	2.37±0.19	9.52±1.6#	4.35±0.81*	4.72±0.8 *	4.32±0.64 *	4.12±0.8 *	<0.01
3rd day	2.3±0.15	10.05±1.9#	3.64±1.4 *	3.9±0.69 *	4.25±0.79 *	4.17±0.54 *	<0.01
7th day	2.3±0.18	9.25±1.5#	3.21±0.8 *	4.57±0.5 *	3.65±0.8 *	3.47±0.8 *	<0.01

*Values were presented as Mean±Standard Deviation (n=8). Where # indicated p-value < 0.001 as compared to Group-I whereas * indicated p-value <0.001 as compared to Group- II*

Table-III Comparison of Serum Uric Acid Level Among Various Groups, (n=48)

Group Comparison	Group-1 Vs. Group-2	Group-2 Vs. Group-3	Group-2 Vs. Group-4	Group-2 Vs. Group-5	Group-2 Vs. Group-6
Serum uric acid level (mg/dl)	<0.001	<0.001	<0.001	<0.001	<0.001

The effect of various doses of MHE and allopurinol in hyperuricemic rats is shown in Table-II. In negative control Group (Group-I), SUA levels on day zero was 2.36±0.19 (Mean±S.D.) mg/dl. In Group-II (Positive Control-Group), SUA levels significantly raised after potassium oxonate injection (p<0.001). Mean±S.D of SUA level was 10.05±1.9 and 9.25±1.5 mg/dl on 3rd and 7th days of the experiment respectively in Group-II. Allopurinol significantly reduced SUA levels to 3.21±0.8 mg/dl on day seven (p<0.001). In Group-IV, mean SUA was 4.57±0.5 mg/dl which was remarkably lower as compared to Group II (p<0.001). Mean SUA of Group-V animals (3.65±0.8 mg/dl) significantly decreased on last day of experiment as compared to hyperuricemic animals (p<0.001). On day seven, average SUA level was 3.47±0.8 mg/dl in Group VI, that was significantly decreased value in comparison with hyperuricemic animals (Group II) (p<0.001). The comparison of serum urate levels among various Groups is tabulated in Table-III.

DISCUSSION

Hyperuricemia i.e. elevated serum urate levels is responsible for the development of various prevailing disorders like gout, nephrolithiasis and metabolic syndrome. Overproduction of uric acid by xanthine oxidase or under excretion of urate through kidneys lead to hyperuricemia.[13,14] Xanthine oxidase inhibitors play a pivotal role in the treatment of hyperuricemia

and gout. Allopurinol and febuxostat are currently available xanthine oxidase inhibitors with multiple adverse effects which limit their use.[15] Literature review depicts that polyphenolic compounds including flavonoids show xanthine oxidase inhibitory activity.[16-18] In our study, potassium oxonate 250 mg/kg was used intraperitoneally on 1st,3rd and 7th days to induce hyperuricemia. This results in 100% induction of hyperuricemia. In Group I (Negative control Group), 2.3±0.19 mg/dl was the mean serum uric acid level on seventh day. After potassium oxonate injections in Group II (Positive control Group), 75% increment of urate levels were observed as compared to negative control healthy rats on seventh day. Allopurinol in Group III decreased serum urate levels to about 65% (four times less) as compared to Group II. A similar study done by Park *et al*. showed results comparable to ours.[19] In Group IV (MHE 100mg/kg), 49% decrease of serum urate levels occurred as compared to hyperuricemic animals. Serum urate levels were 60% decreased in Group V (MHE 200 mg/kg) as compared to Group II. Group VI (MHE 400 mg/kg) showed 62% lower serum urate levels than positive control Group (Group II). This is suggestive to use plants rich in polyphenols in the cure of hyperuricemia and its related problems. Our results are supported by the work done by Chen *et al*. in 2019.[20] They used curcumin, a natural polyphenol, to attenuate potassium oxonate induced hyperuricemia. A similar study done by Oh *et al*. revealed the hypouricemic effects of Chondro-T in hyperuricemic mice which are consistent with our results.[21]

Mimosa himalayana is rich in various flavonoids like quercetin and luteolin.[22] Many flavonoids are recognized to retain pharmacological effects like anti-oxidative, anti-mutagenic and anti-bacterial activities. Moreover, they are powerful inhibitors of various enzymes including, xanthine oxidase, lipoxygenase, cyclooxygenase and phosphoinositide 3-kinase.[23] These facts explain the abovementioned hypouricemic effect of MHE. Further studies can be done to find out which particular constituent of MHE is responsible for xanthine oxidase inhibition and to explore uricosuric effect of MHE.

LIMITATIONS OF STUDY

Our study was limited in its application as it was based on animal models of human diseases. Further research is needed to determine the practical applications of these findings in clinical practice. This study may have beneficial outcomes regarding the treatment of hyperuricemia and gout if done on human participants as part of randomized clinical trials.

ACKNOWLEDGEMENTS

We are highly obliged to the Department of Pharmacology, Department of Physiology and Resource Laboratory of University of Health Sciences, Lahore, Pakistan, for providing research facilities and technical assistance for present study.

CONCLUSION

Oral acute toxicity of *Mimosa Himalayana* extract showed no obvious change in weight, behavior of animals, food intake and alertness. *Mimosa Himalayana* extract lowered serum urate levels significantly as compared to hyperuricemic animals with results comparable to allopurinol.

Conflict of Interest: None.

Authors' Contribution

Following authors have made substantial contributions to the manuscript as under:

BS & JF: Data acquisition, data analysis, data interpretation, critical review, approval of the final version to be published.

SMQ & TM: Study design, drafting the manuscript, critical review, approval of the final version to be published.

SB & SN: Conception, data acquisition, drafting the manuscript, approval of the final version to be published.

Authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

REFERENCES

1. Walker BR, Colledge NR, Ralston SH, Penman ID. In: Davidsons Principles & Practice of Medicine 22nd ed. Elsevier Churchill Livingstone Sydney 2014
2. Ali N, Perveen R, Rahman S, Mahmood S, Rahman S, Islam S, et al. Prevalence of hyperuricemia and the relationship between serum uric acid and obesity: a study on Bangladeshi adults. PLoS One 2018; 13(11): 1-12. https://doi.org/10.1371/journal.pone.0206850
3. Sunarni T, Levian F, Fidriann YI, Iwo MI, Wirasutisna KR. Anti hyperuricemic effect of four plants annonaceae using hyperuricemic rats and enzymatic assay. Asian J Pharm Clin Res 2015; 8(6): 250-253.
4. Wang H, Zhang H, Sun L, Guo W. Roles of hyperuricemia in metabolic syndrome and cardiac-kidney-vascular system diseases. Am J Transl Res 2018; 10(9): 2749–2763.
5. Aziz N, Jamil RT. Biochemistry, Xanthine Oxidase. In: StatPearls. Treasure Island (FL): Stat Pearls Publishing; 2021.
6. Chen C, Lü JM, Yao Q. Hyperuricemia-Related Diseases and Xanthine Oxidoreductase (XOR) Inhibitors: an overview. Med Sci Monit 2016; 22(1): 2501-2512. https://doi.org/10.12659/msm.899852
7. Majeed I, Rizwan K, Ashar A, Rasheed T, Amarowicz R, Kausar H, et al. A comprehensive review of the ethnobotanical uses and biological and pharmacological potential of the genus Mimosa. Int J Mol Sci 2021; 22(14): 7463. https://doi.org/10.3390/ijms22147463

8. Khan AM, Qureshi RA, Ullah F, Shinwari Z, Khan J. Flavonoids distribution in selected medicinal plants of Margalla hills and surroundings. Pak J Bot 2012; 44(4): 1241-1245.
9. Pauff JM, Hille R. Inhibition studies of bovine xanthine oxidase by luteolin, silibinin, quercetin, and curcumin. J Nat Prod 2009; 72(4): 725-731. https://doi.org/10.1021/np8007123
10. Hazarika I, Geetha KM, Sundari PS, Madhu D. Acute oral toxicity evaluation of extracts of Hydrocotyle sibthorpioides in wister albino rats as per OECD 425 TG. Toxicol Rep 2019; 6: 321-328. https://doi.org/10.1016/j.toxrep.2019.04.001
11. Valle Jr DL, Andrade JI, Puzon JJM, Cabrera EC, Rivera WL. Antibacterial activities of ethanol extracts of Philippine medicinal plants against multidrug-resistant bacteria. Asian Pac J Trop Biomed 2015; 5(7): 532-540. https://doi.org/10.1016/j.apjtb.2015.04.005
12. Haidari F, Mehmood RR, Keshavarz SA, Mahboob SA, Eshraghian MR, Shahi MM. Effect of onion on serum uric acid levels and hepatic xanthine dehydrogenase / xanthine oxidase activities in hyperuricemic rats. Pak J Biol Sci 2008; 11(14): 1779–1784. https://doi.org/10.3923/pjbs.2008.1779.1784
13. Haidari F, Keshawarz SA, Shahi MM, Mahboob SA Rashidi MR. Effects of parsley (Petroselinum crispum) and its flavonol constituents, kaempferol and quercetin, on serum uric acid levels, biomarkers of oxidative stress and liver xanthine oxidoreductase activity in oxonate-induced hyperuricemic rats. Iran J Pharm Res 2011; 10(4): 811−819.
14. Nandipati MC, Sumalath G, Baburao C, Babu JR, Sridevi C. Antitumor activity of Mimosa rubicaulis lam against ehrlich ascites carcinoma in Swiss albino mice. Intern J Pharm Sci Res 2014; 5(4): 1514-1524. https://doi.org/10.13040/IJPSR.0975-8232.5(4).1514-24
15. Hanafi MA, Modawe G, Abdrabo AA. Serum sodium, potassium and proteins levels in protein energy malnutrition disorder. Sch J App Med Sci 2016; 4(1B): 96-98.
16. Stewart DJ, Langlois V, Noone D. Hyperuricemia and hypertension: links and risks. Integr Blood Press Control 2018; 12(1): 43–62. https://doi.org/10.2147/IBPC.S184685
17. Sarvaiya VN, Sadariya KA, Pancha PG, Thaker AM, Patel AC, Prajapati AS, et al. Evaluation of antigout activity of Phyllanthus emblica fruit extracts on potassium oxonate-induced gout rat model. Vet World 2015; 8(10): 1230-1236. https://doi.org/10.14202/vetworld.2015.1230-1236
18. Al-Azzawie HF, Abd SA. Effect of some plant extracts on serum uric acid levels, and xanthine oxidase activity in vitro and in oxonate-induced hyperuricemic rats. Eur J Pharm Sci 2015; 2(6): 55-61.
19. Park JE, Yeom Z, Park KT, Han EH, Yu HJ, Kang HS, et al. Hypouricemic effect of ethanol extract of Aster glehni leaves in potassium oxonate-induced hyperuricemic rats. Clin Nutr Res 2018; 7(2): 126-135. https://doi.org/10.7762/cnr.2018.7.2.126
20. Chen Y, Li C, Duan S, Yuan X, Liang J, Hou S. Curcumin attenuates potassium oxonate-induced hyperuricemia and kidney inflammation in mice. Biomed Pharmacother 2019; 118(1): 109195. https://doi.org/10.1016/j.biopha.2019.109195
21. Oh DR, Kim JR, Choi CY. Effects of ChondroT on potassium oxonate-induced hyperuricemic mice: downregulation of xanthine oxidase and urate transporter 1. BMC Complement Altern Med 2019; 19(10): 1-8. https://doi.org/10.1186/s12906-018-2415-2
22. Khan AM, Qureshi RA, Ullah F, Shinwari Z, Khan J. Flavonoids distribution in selected medicinal plants of Margalla hills and surroundings. Pak J Bot 2012; 44(4): 1241-1245.
23. Huang J, Wang S, Zhu M, Chen J, Zhu X. Effects of genistein, apigenin, quercetin, rutin and astilbin on serum uric acid levels and xanthine oxidase activities in normal and hyperuricemic mice. Food Chem Toxicol 2011; 49(9): 1943-1947. https://doi.org/10.1016/j.fct.2011.04.029