



The Effect of Aqueous Extract of *Ocimum gratissimum* On Monosodium Glutamate Induced Biochemical Changes in Albino Rats

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ABSTRACT

The plant *Ocimum gratissimum* is used in folklore for the treatment of some diseases such as cancer, blood disorders and hepatitis. This study investigated the impact of *Ocimum gratissimum* leaf extract on monosodium glutamate-induced biochemical changes in albino rats. Fresh leaves of *Ocimum gratissimum* were procured in Oyingbo market in Lagos. The leaves were air-dried and the aqueous extract was prepared and used for the study. Acute toxicity of the extract was investigated by Lorke's method. Twenty-five albino rats were randomized into 5 groups of five animals each. Group 1 was used as control, group 2 received no treatment while groups 3, 4 and 5 served as the treatment groups. Groups 2-5 were orally administered with 200 mg/kg body weight of monosodium glutamate for 14 days. This was followed with administration of 100, 200 and 300 mg/kg of *Ocimum gratissimum* leaf extract for groups 3, 4 and 5, respectively. The animals were sacrificed after 14 days of treatment. Blood samples were obtained in all the groups and analyzed for renal and hepatic functions indices. The median lethal dose (LD₅₀) of the extract was found to be 3800 mg/kg body weight. There was increase in weight of the rats after treatment, hepatic functions indices of alanine transferase (ALT), aspartate amino transferase (AST), alkaline phosphatase (ALP), total protein, albumin and renal function indices of urea and creatinine serum levels normalized after treatment. *Ocimum gratissimum* aqueous extract affected some biochemical parameters induced by monosodium glutamate.

Keywords: *Ocimum gratissimum*, Monosodium glutamate, Hepatic function, Renal function.

Introduction

Herbal medicine is an indispensable old practice in different part of the globe for both preventive and curative purposes. Lately the use of herbs for medical reasons is gaining global acceptability. A greater percentage of the world population rely on botanical preparations to meet their health needs. Many nations have monitoring agencies responsible for addressing the concerns and worries often entertained about toxicity and dosage of herbal medicine.

Research has shown that herbal medicine is not without some challenges with regards to their adverse side effects that could be caused by herbal combinations which are expected to be more potent when used as remedies. The increase in demand for herbal medicine together with the erroneous impression by the people that herbal products are natural and as such less harmful to the body¹ has brought concern and fear over the quality, effectiveness and safety of some of the available natural herbs.² There have been confirmed cases of renal failure and liver diseases associated with the consumption of herbal medicine.^{3,4}

The plant *Ocimum gratissimum* (*O. gratissimum*) belongs to the family Lamiaceae. It has been found to possess nematocidal and

insecticidal properties.⁵ The phytochemical composition of the leaf extract of *O. gratissimum* revealed the presence of alkaloids, saponins, tannins, anthraquinones, steroids, terpenoids, and cardiac glycosides.⁷⁻⁹ This may account for its wide use in folk medicine as curative for many diseases especially in Africa and Asia.

The essential oil of *O. gratissimum* contains eugenol believed to possess antibacterial activity^{10,11} as well as food preservative property.¹² It is known to be toxic to leishmania. The oil has also been shown to contain anti-diabetic agent.¹³ Monosodium glutamate (MSG) is the sodium salt of glutamic acid, and glutamic acid is a non-essential amino acid¹⁴ present in tomatoes, grapes, cheese, mushrooms and other foods.¹⁵ MSG is added to food either as pure monosodium salt or as a part of a mixture of amino acids and little peptide. It is produced as a direct salt of hydrolysis of polypeptides.¹⁶ When added into food in minute quantities, the taste of the food is enhanced.¹⁷ There is substantial evidence that the reasons for this effect is that MSG stimulate leptin-hypothalamus sensory cells of taste.¹⁸ However, in Nigeria, dry cleaners and individuals often use MSG in laundry. This attribute suggests it could be toxic to tissues and organs of the body when ingested.

Previous investigation revealed that MSG is toxic to some organs.¹⁹ There are reports of its toxic effect on the pancreas^{19,20} and kidney.²¹ There are reports that MSG induced or caused changes in the metabolic rate of glucose utilization and reduced radical scavenging properties of some agents. It can also produce reactive oxygen species which is known to cause damage to DNA, lipid and protein²² in different cells of the body.

This work determined the impact of varying doses of aqueous extract of *O. gratissimum* on biochemical changes in albino rats treated with monosodium glutamate.

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Materials and Methods

Plant collection and authentication

Fresh leaves of *Ocimum gratissimum* were obtained from Oyingbo in Ebute Meta, Lagos and it was identified and authenticated by Mr. Nodza George at Botany Department, University of Lagos, Nigeria. A voucher sample reference number LUH 8550 was retained in the herbarium.

Preparation of Plant Materials

Ocimum gratissimum leaves were plucked, cleaned and dried for 14 days. The dried leaves were ground to fine powder using a mechanical grinder.

A 66-gram portion of the powdered leaf sample was suspended in 480 mL of distilled water for 4 hours at room temperature and filtered. The filtrate was then poured into stainless plate and the extract was dried at 30°C in the oven for 2 days. The concentrated extract was stored in labelled container until needed for the study.

Experimental animals

Fifteen albino mice aged 6-7 weeks, average weight of 20 gram and twenty-five albino rats of both sexes aged 8-9 weeks, average weight of 110 gram were used for the study. The animals were obtained from the animal Laboratory of University of Lagos, Idi Araba. They were kept in a cage with free access to standard rat pellet and water. Acclimatization of the animals was done for 14 days prior to commencement of the experiment.

The study was in conformity with the NIH guidelines for animal care in studies involving experimental animals.^{23,24} Ethical permission was granted by Research and Experimental Ethics Committee of Yaba College of Technology.

Acute toxicity study

Acute toxicity was investigated using Lorke method.²⁵ This was determined in 2 phases using 15 mice. In phase one 12 mice were randomized into 4 groups of 3 rats each. Group 1 served as control while groups 2, 3 and 4 received 10, 100 and 1000 mg/kg body weight of *Ocimum gratissimum* leaf extract, respectively. In phase two 3 mice were randomized into groups 5, 6 and 7 with one mouse per group. Each received 1600, 2900 and 5000 mg/kg body weight of *Ocimum gratissimum*, respectively.

Biochemical studies

The rats were randomly divided into five groups of 5 rats each separating the males from the females to avoid reproductive activities. MSG at 200 mg/kg body weight were orally administered daily for 14 days in groups 2-5 followed by 100, 200 and 300 mg/kg body weight of the extract in groups 3-5. The weights of the rats were taken before and after administration of MSG and the plant extract.

After 14 days of administration of the plant extract, the rats were fasted overnight and blood sample was taken from each rat through cardiac puncture into sterile plain sample bottles and serum was collected after spinning.

Statistical Analysis

Tests were conducted in triplicate and data was subjected to statistical analysis using SPSS 15. Results were expressed as mean \pm standard error of mean. Level of statistical significance difference between test and control was at $P < 0.05$.

Results and Discussion

In the first phase of acute toxicity no death was recorded after 24 hours and no sign of toxicity was observed among the mice. However, in the second phase one death was recorded at 5000 mg/kg body weight. The median acute toxicity of *O. gratissimum* was calculated to be 3800 mg/kg body weight as shown in Table 1. Administration of varying doses of *O. gratissimum* leaf extract to rats treated with MSG

caused an increase of 17%, 20% and 26% in the weight of rats in groups 3, 4 and 5, respectively as shown in Table 2. *O. gratissimum* at doses of 100 and 200 mg/kg body weight in MSG treated albino rats did not cause significant change ($P > 0.05$) in serum levels of ALT, AST, total protein and albumin but at 300 mg/kg body weight, serum level of Alkaline phosphatase reduced significantly ($P < 0.05$) compared to control as shown in Table 3. Furthermore, administration of varied doses of *Ocimum gratissimum* leaf extract did not cause significant change ($P > 0.05$) on renal function indices (urea and creatinine levels) on MSG treated rats compared to the control group (Table 4).

Determination of lethal dose (LD₅₀) is nowadays regarded as a major parameter in the measurement of acute toxicity and an initial step for screening chemicals and pharmacological active agent for toxicity.²⁶

The LD₅₀ of aqueous extract of *O. gratissimum* was calculated to be 3800 mg/kg body weight as shown in table 1. The extract at this dose is classified as being slightly toxic.²⁷ This value is slightly lower than 4242.64 mg/kg body weight earlier reported²⁸ where acute and sub-chronic studies were carried out. The weight of an animal is often used as an index to measure toxic effect of extract. Groups 3, 4 and 5 administered with 100, 200 and 300 mg/kg body weight of the extract caused 17%, 20% and 26% increase, respectively in weight compared to 23% in group 1 (control). This trend showed that *O. gratissimum* did not have any harmful effect on the animals.

The hepatic function indices of ALT, AST, total protein and albumin were not significantly affected by varied doses of *O. gratissimum* extract. This indicate that the liver is not adversely affected. AST, ALT and ALP enzymes are normally found in vital organs but are found largely in the liver. AST is released into serum when any of these tissues is damaged, an elevation or decrease of this liver enzyme in the serum might indicate liver injury. *O. gratissimum* trim the level of serum ALT, AST and ALP activities in the liver compared to rats in the Group 1. The results suggest that the daily dosage has no significant effect on the level of ALP, AST and ALT. The non-significant difference in the total protein and albumin values between the control group and all other groups suggest that there was no much depression in hepatic function and or degradation of protein, this trend conformed to earlier report.²⁹ In addition, renal function indices of urea and creatinine did not change significantly which indicated that the kidneys were not affected.

Table 1: Acute Toxicity of the aqueous extract of *Ocimum gratissimum* on mice.

Group	Dose mg/kg	Mortality (D/T) after 24 hours
Phase 1		
1	Control	0/3
2	10	0/3
3	100	0/3
4	1000	0/3
Phase 2		
5	1600	0/1
6	2900	0/1
7	5000	1/1

*D/T = Number of dead mice/Total number of mice

$$LD_{50} = \sqrt{D_0 \times D_{100}}$$

Where D₀ = Highest dosage that gave 0% mortality

D₁₀₀ = Lowest dosage that gave 100 % mortality

$$LD_{50} = \sqrt{2900 \times 5000} = 3800 \text{ mg/kg body weight.}$$

Table 2: The Effect of varied doses of *Ocimum gratissimum* extract on the weight of monosodium glutamate treated albino rats

Groups	Dose (mg/kg bw)	Weight before (g)	Weight After (g)	Weight gain (g)	% Weight gained
1	Control	123.88 ± 14.76	151.88 ± 12.09	28.00 ± 2.67	23
2	MSG not treated	108.43 ± 14.85	133.57 ± 25.57	25.44 ± 10.72	23
3	100	116.00 ± 15.93	135.25 ± 20.82	19.25 ± 4.89	17
4	200	102.43 ± 11.27	127.14 ± 16.65	20.71 ± 5.38	20
5	300	109.57 ± 21.05	138.29 ± 15.89	28.72 ± 5.16	26

Table 3: The effect of varied doses of *Ocimum gratissimum* extract on hepatic function Indices of MSG treated albino rats¹

Group	Dose mg/kg b.wt	ALT (iu)	AST (iu/)	ALP(iu)	Total Protein g/L	Albumin (g/L)
1	Control	20.67 ± 6.43	19.67 ± 4.73	99.00 ± 33.00	54.60 ± 4.67	40.73 ± 2.17
2	not treated	21.33 ± 2.89	20.00 ± 2.65	132.00 ± 57.16	54.90 ± 1.0	42.10 ± 1.71
3	100	23.00 ± 5.00	20.00 ± 6.08	121.00 ± 50.41	54.20 ± 2.14	42.10 ± 2.07
4	200	23.00 ± 5.00	24.33 ± 2.31	143.00 ± 19.0	56.60 ± 2.56	41.53 ± 1.44
5	300	24.67 ± 5.77	24.33 ± 6.11	21.00 ± 38.1*	55.90 ± 2.74	42.90 ± 2.07

¹Value represent mean ± SD of triplicate determinations. *p < 0.05 there is statistical significant difference between test and control
Key: ALT = Alanine transaminase, AST = Aspartate Transaminase, ALP = Alkaline Phosphatase.

Table 4: The effect of oral administration of varied doses of *Ocimum gratissimum* on some renal function indices of MSG treated albino rats¹.

Group	Dose (mg/kg bw)	Urea(mg/dL)	Creatinine (mg/dL)
1	Control	46.00 ± 3.46	1.73 ± 0.23
2	MSG not treated	47.40 ± 1.91	1.73 ± 0.23
3	100	42.90 ± 4.25	1.33 ± 0.61
4	200	48.50 ± 0.00	2.00 ± 0.00
5	300	44.17 ± 3.75	1.47 ± 0.61

¹Value represents mean ± SD (standard deviation) of triplicate determinations
P < 0.05 there is statistical significant difference between test and control.

Conclusion

Ocimum gratissimum leaf extract caused increase in weight in MSG treated rats, hepatic and renal functions indices of MSG treated rats were not significantly affected at low doses of the extract.

Conflict of interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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