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Effect of Aqueous Extract of *Aframomum*melegueta (Alligator Pepper) on some Haematological and Biochemical Profile of Albino Wistar rat.

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ARTICLE INFO ARTICLE INFO In the yuletide, plant extracts, animal fluids, roots, stems and leaves have been consulted by man in his vide to overcome hunger, illness and ignorance. Alligator is one of those plants whose seeds are of research interest in this study. Its effect on some of the biochemical and hematological indices of albino Wister rats were investigated. Twenty four (24) Albino Wistar rats weighing 120 - 150g were divided into two (2) groups. Group A (12 rats) served as control while group B (12 rats) served.

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Twenty four (24) Albino Wistar rats weighing 120 - 150g were divided into two (2) groups. Group A (12 rats) served as control while group B (12 rats) served as the test rats. The extraction of the aqueous extract was done by cold maceration of pulverized seeds in absolute sterile water for 24 hours. The extract was filtered and concentrated in rotary evaporators at 40° C.

The central group was given normal rat feed and water. While the test rats received 1 ml of the extract. Both groups were given rat feed and water ad libitium for 28 days. At the end of the feeding, blood samples were collected from the animals by cardiac puncture and $2.0^{\rm ml}$ each were placed in anticoagulant bottles containing E DTA (Ethylene diamine tetracetic acial for hematological tests while 3.0 ml were placed in plain bottles and left to clot and the serum extracted for biochemical analysis. Results from the analysis showed a significance increase in white blood cell counts (PLO. 05) and reduced hemoglobin levels and packed cell volume (PLO .25) in the test rats, there is no effect in platelet counts, creatinin, urea and the electrolyte. It could be deduced that aqueous extract of Aframomum melegueta (Alligator pepper) has adverse effect on haemopoiesis and no effect on the kidney using the safe dose.

INTRODUCTION

Aframomum melegueta (Alligator pepper) is a perennial plant native to swampy habitats along the West African coast. It is a specie in the ginger family, zingiberaceae. It is a flowery plant. Its trumpet shaped purple flowers develop into 5 to 7cm long pods containing numerous, reddish – brown seeds.

The spice commonly known as ossame, grains of paradise, meleguet a pepper, alligator pepper, quinea grains or guinea pepper is obtained from the ground seeds, it imparts a pungent, peppery flower with hint of citrus. The pungent peppery taste of seeds is caused by aromatic ketones. The stem at times can be short and usually share signs of scars and fallen leaves. The fruits contain numerous small, golden red brown seeds. It is a common ingredient in West African cuisine; where it imparts both pungency and spicy aroma to classic West African soup (stews) (Celt net and katzer's 2007) .The paucity of information on the reports of effect on blood cells prompted this study. Hence the research on the aqueous extract of A. melequeta is to find out its effect on some hematological and biochemical profiles of male albino Wister rats.

Alligator pepper seeds are an excellent source of phytonutrients as it contains terpenoides, alkaloids, flavenoids, and tannins cardiac glycosides, saponins and phenolic compounds. It is a popular spice that is normally snacked upon especially by elders and sometimes youths. In our locality in Nigeria, alligator pepper is used in conjunction with kola nuts and groundnuts butter during Baby naming ceremonies, traditional marriages, burial ceremonies and town meetings. It also has anti- inflammatory properties, its phytonutrients such as terpenoid, alkaloids, flavenoids and tannins scavenge for free radicals and offer protections against viruses, allergies, microbes, tumors, Ulcers and hepatoxins (chemical liver damage) in the body. The seed extracts of the alligator pepper can be used for treating gastrointestiment disorders such as stomach pain, unspecific stomach disturbances, and Ulcer and intestine worms. Due to the constituents of phenolic compounds, it is normally used to prepare disinfectants. The seed of alligator pepper can ease digestion of food thereby preventing constipation and blockage despite all the above nutrimental medicinal and health benefits of alligator pepper, pregnant women in their early trimesters are advised not to consume high dose of alligator pepper as research carried out in an experiment by Inegbenobor et al, 2009 showed that high dosage of alligator pepper administration to pregnant rats led to termination of their first trimester pregnancies. Alligator pepper seed can also be used in preparing herbal remedy for treating infections, skin diseases such as measles, chicken pox and small pox. The aqueous extract of the plants is analgesic in nature and as such can be used for receiving and alleviating pains such as joint pain, toothache, stomach pain, arthritic pain and rheumatoid pain.

MATERIALS AND METHODS

Twenty four (24) male albino Wister rats were randomly selected and kept in a metal cage with iron netting in a laboratory environment. They were kept in the animal house for 14 days (2 weeks) for acclimatization to the environment before commencing an acute feeding with the extract for 28 days.

EXPERIMENT DESIGN:

Twenty four male albino Wister rats (120 – 180g) were selected and divided into two groups. Group A rats (12) served as central while group B rats served as test animals. Group A rats were fed with normal rat feed and water ad libitum. Group B rats received rat feed in addition to the oral administration of I.0 ml of the seed extract of *Aframomum melegueta* once daily, normal rat feed and water also.

PREPARATION OF EXTRACT

The method of extraction was as described by Ugochukwu et al, 2003. The fruit containing seeds of A. melegueta were purchased from Afor Egbu Market, a local market around the campus. The botanical identification and authentic action was confirmed by a botanist from the department of Biological Science, Chukwuemeka Odumegwu Ojukwu University. (Former Anambra State University) Uli Campus. The seeds were dehulled dried in the sun for 48 hours, and later grinded to powder. The powdered seeds were stored in a glass bottle with a screw cap and kept in a refrigerator (4°C). Later the powdered seeds were homogenized with distilled water and kept for 12hrs. The mixtures were filtered with Whatman No .1 filter paper. The filtrates were concentrated in one tenth $(^{1}/10)$ of the original volume at 38 -40°C, using a rotating evaporator. 5.0g of the powdered was re - suspended in 100ml of distilled water before being given to the rats. 1. 0ml of the suspension was administered to the rats daily for 28 days using blunt needle and syringe.

TOXICITY STUDIES (LD50).

The LD_{50} of the extract in albino mice was determined using Lorke's method (1983). Mice (60- 80g) were fasted overnight for 22hrs and doses of the extract of *Aframomum melegueta* groups of the mice (n=3) and observed for another 24 – 48hrs. The mice that served as central received normal saline only. The LD50 for the extract was calculated by geometric mean of the dose killing none of the three mice in the group and dose killing all the animals in the group.

LD50 = Dose killing all animals in the group x Dose killing none of the animals in the group.

PHYTOCHEMICAL STUDIES OF A. MELEGUETA:

The aqueous extract of the *Aframomum melegueta* (Alligator pepper) was screened for the presence or absence of metabolites using standard phytochemical screening tests as described by Harbourme (1973), Trease and Evans (1996). The extract was tested for carbohydrates, steroids, sugars, saponins, alkaloids, flavenoids, resins, calcium glycosides, steroids, acidic compounds, fats and oils.

DETERMINATION OF SOME HAEMATOLOGICAL AND BIOCHEMICAL PROFILES:

Blood samples were collected into anticoagulant bottles and plain bottles from the animals by cardiac puncture. The samples were distributed into 2.0ml for

anticoagulant (EDTA) bottles and 5.oml into plain bottles. The hematological tests were done within hours of the blood sample collection while the biochemical analysis was carried out after separating the serum samples from the collected whole blood.

The packed cell volume and hemoglobin concentration were done by the method described by Alexander and Griffiths (1993). The total white blood cell count and platelet counts were done according to the visual method of Decline and Levis (1993). The biochemical analysis was determined using mindrel and analytical machine.

STATISTICAL ANALYSIS:

The results obtained in the study were represented as mean and standard deviation (mean + S.D), while the students s'-t- test was used to compare the result of the contrail and the test rats. A P value of less than (PL0.05) is statically significant.

RESULTS:

The result obtained from this research study is presented in tables below:

TABLE 1: The phytochemical analysis of *Aframomum melegueta*:

	Alka loids	Flave noids	Tan nins	Carbo hydrat e	Calcium	Saponins	Terpenoi ds	Reducing resins	Cardiac glycosides
Degree of concentration.	Н	+	+	-	+	Н	Н	-	+

KEY:

- Negative, + present in small concentrations
- ++ present in moderate concentrations
- + H present in high concentrations.

TABLE 2: Indicates the effect of *A. melegueta* on some haematological indicates of male albino rats after 28 days feeding. (PLO.05).

Groups	Hbg /100ml -+ SD	PCV -* S.D	NBC Per Mm³ -†S.D	Platelet Count x 10 ⁹ /L- ⁺ S.D
Group A Control n = 12	14. 2 - ⁺ 0.6	42.6 - ⁺ 1.8	6,420 - ⁺ 31	160 - +20
. Test rats (Group B) before extract feed n = 12	14.3 - ⁺ 0.6	43 - ⁺ 0.6	6,510 - ⁺ 23	162 - +25
Test rats (Group B) 28 days After extract feed.	9.5 - ⁺ 0.2	28.5 - ⁺ 0.6	8,412 + 40	165 - 20
P value	P < 0 05	P< 0.05	P< 0.05	P > 0.05

TABLE 3: The effect of *A. melegueta* on the serum electrolyte, urea and creatinine levels in rats after 28 days feeding on the extract.

Groups	Sodium meqIL - ⁺ S.D	Potassium meq IL - ⁺ S.D	Chloride MeqIL - ⁺ S.D	Bicarbonate meqIL - ⁺ S.D	Urea - ⁺ S.D	Creatinine Mmel I L -+ S.D
Control Group A n= 12	141-+23	3.8-* 0.7	104 - 18	26 - +0.4	6.1-+0.89	75.4- ⁺ 1.6
Test rats Group B n= 12 Before extract.	143 - ⁺ 28	3.7 - + 0.5	105 - 12	27 - + 0.2	6.0 - + 0.44	75.4- ⁺ 1.5
Test rats Group B After extract n= 12	148 -* 26	4.8 - 0.8	110 - 16	32 - + 0.8	6.9 - †0.42	80.2 - 1.2
P value	P > 0.05	P > 0.05	P > 0.05	P > 0.05	P > 0.05	P > 0.05

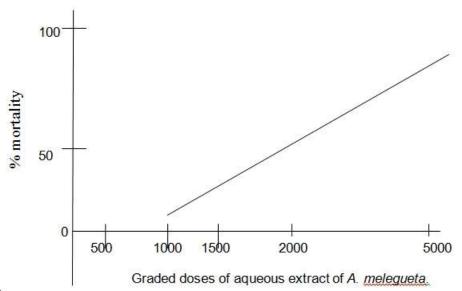


FIG 1. Lethality studies medicating the effect of administering graded doses (500- 5000 mg IU) on mice on aqueous extract of *A. melegueta*. $LD_{50} = 4,000.00$.

DISCUSSION

The effect of *Aframomum melegueta* on some hematological and biochemical indices of male albino Wister rats has been demonstrated.

The lethality dose used in this research was for the safe up keep of the animals throughout the period of research. The 1. 0 ml volume of the extract administered to the test rats in group B was adequate for their health, hence the acute toxicity study (<D 50) showed that *A. melegueta* extract was non - toxic as shown in fig. 1. (< > 50 of 4000 mg kg. The administration of *A. melegueta* to the rats showed a negative effect in the hemoglobin concentration (Group A HB: 14.3 - + 0.2, Grp B. HB: 9.5 - + 0.2)

Giving a low concentration in test animals, Group B compared to their corresponding control.

This observed effect could be attributed to the feint that *A. melegueta* tacks protein in its content, but contains carbohydrates. Proteins and carbohydrates are for energy, they form the structural material of muscles, tissues and organs and are equally regulates of function as enzymes and hormones. The packed cell percentage concentration was also reduced. (Table 1.) . The packed cell volume in test rats was 28.5 - 0.6% in Group A. compared to their corresponding controls given as 42.3- 0.2%. However the white blood cell count was elevated in the test rats Group B. The result was recorded as 8,412 - 40 /mm³.

This slight increase though within normal range could be a physiological response to the pepperish nature of *A. melegueta* seed. Certain conditions such as acute bacterial infections are capable of producing a variation in the white cell count, leucocytes is the term used to describe an increase in white cell count that is above 11.0x10³/mm³. (Bakers et al, 1985).

This pepperish taste could cause reduction in appetite to eat, in the rats so resulting to decreased blood hemoglobin concentration. The platelet count, and all the biochemical indices studied were not affected by the extract feed hence the kidney function was not affected. The result of the biochemical analysis showed that though the kidney is the main organ by which drug and their metabolites are eliminated from the body and the target of drug toxicity, it was not affected by the extent.

A. melegueta demonstrated a pure plant characteristics by the presence of Tannins, saponins, terpenoids, alkaloids and flavonoids in its physiochemical content.

It could be deduced that high consumption of *Aframomum melegueta* may possibly have an adverse negative effect in hematopoiesis.

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