



Effect of Cashew apple juice (*Anacardium occidentale* L.) on Hematology and Spleen of Gentamicin Induced Injury in Albino Rats

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Abstract

Aim/Introduction: The use of plant in treatment of human diseases is as old as man. Medicinal plants are often consumed locally without a graded dose or expected duration of use. Cashew is traditionally used for diabetes, high cholesterol, heart disease, stomach and intestinal (gastrointestinal) ailments, skin problems, and other conditions. The aim of this study is to determine protective effect of Cashew apple juice, *Anacardium occidentale*, on gentamicin induced injury on spleen of albino rats.

Method: A total of thirty rats (155–210 g) were used for this work. Group one served as the positive control receiving normal saline. Group two received normal saline for eight days, while group 3, 4 and 5 received 10, 20 and 40 ml/kg of the extract respectively. On the eighth (8th) day, animals in groups 2 – 5 received gentamicin 100 mg/kg bw orally. Twenty hours after the last administration, that is on the 9th day, all animals were weighed again and sacrificed under light diethylether vapour.

Result: *Anacardium occidentale* fruit juice extract caused significant increase ($P < 0.05$) in the level of RBC, PCV, Hb, platelet and monocyte when compared to the gentamicin group. There was no significant increase ($P < 0.05$) in the level of WBC. There was also significant increase ($P < 0.05$) in the body weight ratio of the rat's spleen. Histopathology observations agree with hematology study.

Conclusion: *Anacardium occidentale* has protective tendency on vital body organ. The fruit may also be useful in improving hematological parameters.

Introduction

The cashew tree (*Anacardium occidentale* L.) belongs to the family Anacardiaceae. It is a native plant of Brazil and the fruit consists of a cashew nut (the true fruit) and a cashew apple (pseudofruit), which has excellent nutritional and sensory properties.¹ The cashew culture is one of the main agronomic activities in Northeast Brazil and almost the whole production is concentrated in the states of Ceará, Piauí, and Rio Grande do Norte.² However, no more than 10% of potential cashew apple output is at present consumed or utilized in either fresh or processed form such as jam, syrup, chutney, beverage, ice creams, etc.^{3,4,5} The economic value of cashew apple juice has increased as the concentrated and processed (called cajuína) forms are among the most popular products in Northeast Brazil⁴.

In Brazil, it is also used to treat diabetes, weakness, muscular debility, urinary disorders, asthma, eczema, psoriasis, scrofula, dyspepsia, genital problems, bronchitis, cough, intestinal colic, leishmaniasis, venereal disease, as well as impotence, and syphilis-related skin disorders.⁶ In the sixteenth-century, Brazil cashew fruits and their juice were taken by Europeans to treat fever, sweeten breath, and ‘conserve the stomach’. It is taken for syphilis and as a diuretic, stimulant and aphrodisiac⁷. In addition to being delicious, cashew fruit is a rich source of vitamins, minerals and other essential nutrients. It has up to five times vitamin C than oranges and contains a high amount of mineral salts⁸. Because of its high amount of amount of vitamin C and mineral salts, cashew fruit is used as a catalyst in the treatment of premature ageing of the skin. Several clinical studies have shown that *anacardiac* acid is a component of cashew, with highest concentration in the nutshells curb the darkening effect of ageing by inhibiting tyrosinase activity, and that they are toxic to certain cancer cells⁹ reveal a unique function of *anacardic* acid in that, for dietary conditions enhancing body fat deposition that is consumption of a diet high in carbohydrates, dietary anacardic acid has the potential to decrease body fat deposition. In the last 20 years, the interest in medicinal plants has increased together with the number of investigations into their biological effects on human beings and animals.¹⁰ Although, poisonous plants are ubiquitous, herbal medicine is used by up to 80% of the population in the developing countries.¹¹

Cashew gum is used in pharmaceuticals and as substitute for gum Arabic. Extracts from roots, stems and fruits of *A. occidentale* leaf have been used by the Cameroonian and other countries’ folk medicine¹². In the traditional Nigerian and Brazilian pharmacopoeia, stem bark of *A. occidentale* Leaf is known for its anti-inflammatory effects⁷ The leaves and/or the bark is also used in Brazil for eczema, Psoriasis, Scrofula, Dyspepsia, genital problems, and venereal diseases, as well as for impotence, bronchitis, cough, intestinal colic, leishmaniasis and syphilis-related skin disorders.¹³ A strong antioxidant capacity was also observed against hepatocarcinogenesis induced by aflatoxin B1 in Wistar rat.¹⁰ Preclinical studies reveals that metabolites isolated from the bark of cashew tree demonstrated antipyretic action for *anacardic* acid⁹ realized mutagenic tests with vegetable oil derived from cashew nut, which presented mutagenicity with or without the activation of the S9 fraction in *Salmonella thyphimurium. H.*¹⁴ demonstrated that tannic acid, a compound present in the cashew, presented an antimutagenic effect on the *Salmonella thyphimurium* TA98 lineage. The bark and leaves of cashew are a rich source of tannins, a group of plant chemicals with documented biological activity.¹² These tannins, in a 2004 rat study, demonstrated anti-inflammatory and astringent effects, which may be why cashew is effective in treating diarrhea¹⁵. The aim of this study is to

determine the effect of Cashew apple juice *on* spleen and hematological parameter of gentamicin induced toxicity on albino rats.

Materials and Method

Plants collection

Anacardium occidentale fruits were collected from its natural habitat from nearby Oye village, Ekiti State, Nigeria. The plant was authenticated from Department of Botany, Federal University, Ekiti State Nigeria.

Extraction

The plant material cashew apple, orange reddish, was harvested from the Oye Local government area of Ekiti State, Nigeria. Cashew apple fruits were cut into small pieces and crushed under laboratory conditions. The resulting mash was pressed using a press to extract the juice

Animals

Adult albino rats of both sexes were obtained from Bingham university animal house. They were maintained on standard animal pellets and water *ad libitum*. Permission and approval for animal studies were obtained from the College of Health Sciences Animal Ethics committee, Federal University, Oye Ekiti, Ekiti State, Nigeria..

Animal treatment

A total of thirty rats were used for this work. Group one served as the positive control receiving normal saline. Group two received normal saline, while group 3, 4 and 5 received 10, 20 and 40 ml/kg of the extract respectively. Gentamicin 100mg/kg bw were administered daily to groups 2-6 concomitantly with the above treatment for 8 days. Twenty hours after the last administration, all animals were weighed again and sacrificed under light diethylether vapour.

Hematological study

Blood samples were collected from each rat by cardiac puncture immediately after the animals were sacrificed under diethylether anesthesia, using 21-gauge (21 G) needles mounted on a 5ml syringe into ethylene diamine tetra-acetic acid (EDTA) – coated sample bottles for analyzed. Hematological parameters such as full blood count (FBC), hemoglobin, (Hb), packed cell volume (PCV), platelet concentration (PLC) and Total and differential white blood cell count (WBC). These parameters were analyzed using automatic hematological system.

Histopathological examinations

Spleens were processed and stained with haematotoxylin and eosin (H&E). Prepared slides of the organs were mounted on high-defination microscope. The result was interpreted by a Pathologist in the Department of Chemical Pathology, Ekiti State University, Ekiti State. Morphological changes in the excised organs of the sacrificed animals were observed and recorded. Histologic micrographs were taken.

Results

Effect of *A. occidentale* fruit juice on haematological parameters of Gentamicin induced toxicity

Anacardium occidentale fruit juice extract caused significant increase ($P<0.05$) in the level of RBC, Hb, PCV, platelet and eosinophils when compared to the organotoxic group. Also, there was no significant difference in level of WBC caused by gentamicin (Table 1).

Effect of *A. occidentale* on platelet count on Gentamicin induced toxicity

Gentamicin caused decrease in platelet count in group 2, while there was significant ($P<0.05$) increase in groups administered *Anacardium occidentale* fruit juice across all doses administered when compared to group 2 (Table 2).

Effect of *Anacardium occidentale* on body weight ratio of spleen of rats in Gentamicin induced injury

A. occidentale cause significant ($P<0.05$) improvement in spleen of rats that receive concomitantly with gentamicin compared to the group that received gentamicin only (Table 3).

Effect of *Anacardium occidentale* of histopathology of rat spleen

The spleen showed lymphocyte degeneration in gentamicin group. Fruit juice caused histological improvement at all doses administered (Figure 3).

Table 1:Effect of *A. occidentale* fruit juice on haematological parameters of gentamicin induced toxicity

Group	RBC (X 10 ¹² /l)	PCV (%)	Hb (g/dl)	WBC (X 10 ⁹ /l)	Monoc ytes (%)
Normal	7.4±1.11*	44.00±1.87*	13.46±0.87*	6.4±1.43*	4.13±1.81*

saline					
GENT 100 mg/Kg	3.66±0.98	16.92±2.17	9.57±0.34	3.10±1.00	2.90±1.62
GENT + 1ML OA	6.11±0.43*	43.77±1.20*	10.44±0.38	3.08±1.11	6.11±1.61*
GENT+ 2ML AO	7.25±0.36*	44.4±1.34*	12.19*±0.37	3.06±1.43	7.56±1.33*
GENT+ 4ML AO	7.43±0.43*	44.22±1.8*	13.23*±0.34	3.17±1.10	8.27±1.35*

Table 2: Effect of *A. occidentale* on platelet count on gentamicin induced toxicity

Group	Platelet Count
Normal saline	350±3.42*
Gent 100 mg/Kg	199±4.12
Gent + AO 10 ml/kg	312±2.22*
Gent+ AO 20ml/kg	473±3.15*
Gent+ AO 40 ml/kg	482±3.81*

Table 3: Effect of *Anacardium occidentale* on body weight ratio of spleen of rats in weight induced injury

Group	SPLEEN BW RATIO
Normal saline	0.47±0.76*
PCM + 2000 mg/kg	0.19±0.15
A.O 1ML+PCM	0.38±0.11*

A.O 2ML+PCM	0.40±0.41*
A.O 4ML+PCM	0.45±0.16*

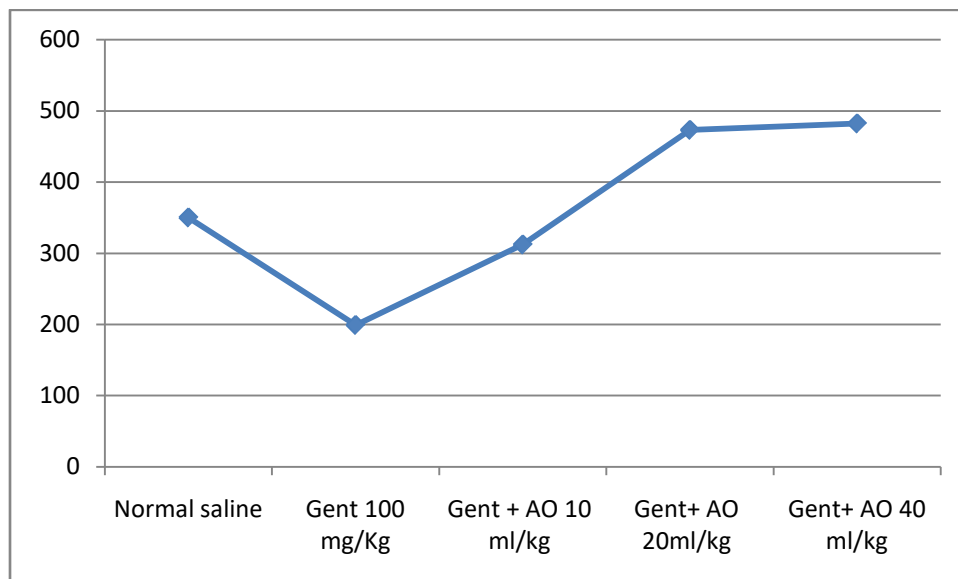


Figure 1: effect of *Anacardium occidentale* platelet count of gentamicin induced injury in rat.

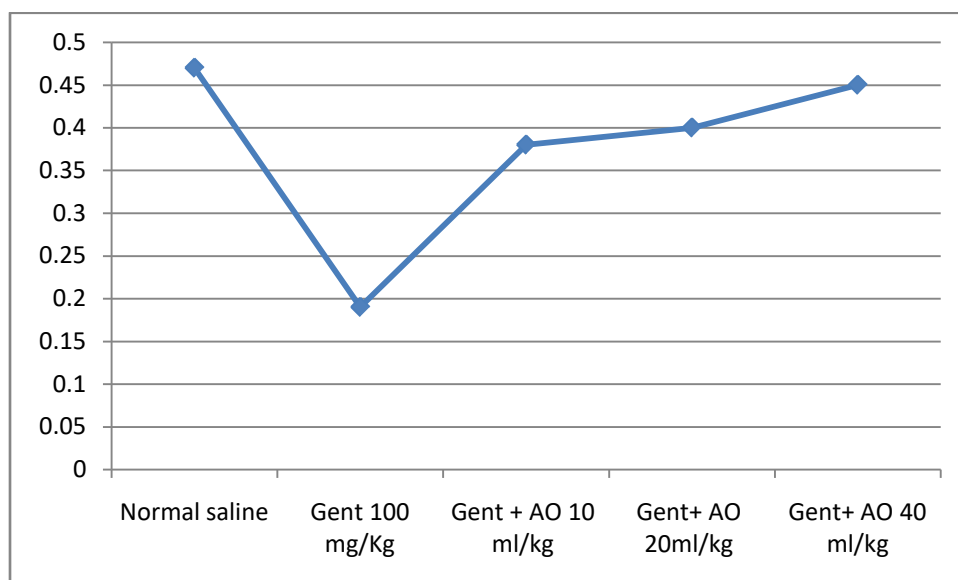


Figure 2: Effect of *Anacardium occidentale* body weight ratio of spleen in Gentamicin induced injury in rat.

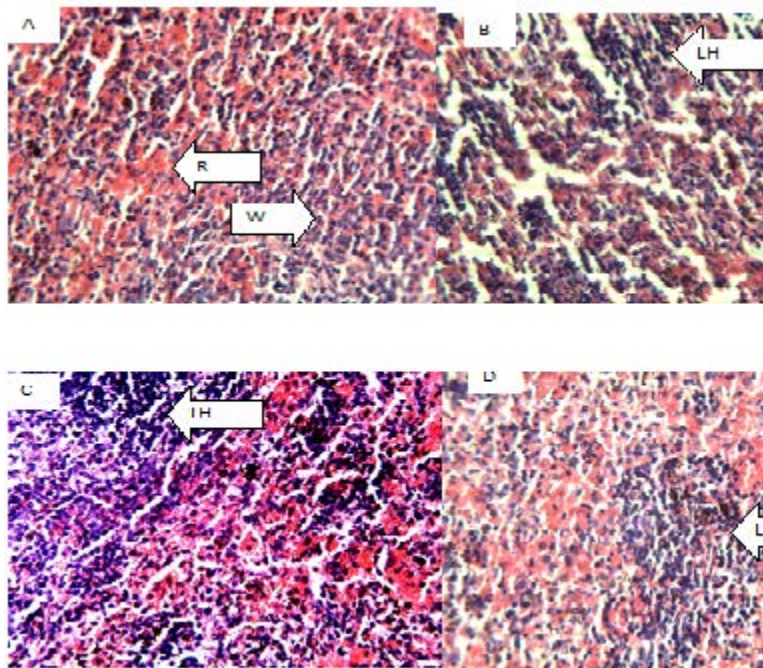


Figure 3: Histological section of the Spleen (H and E $\times 100$). (a) control group (b) Gent 100 mg/kg (c) Gent 100 mg/kg and A.O 10ml/kg (d) Gent 100mg/kg and A.O 20 ml/kg,

Discussion

Cashew apple juice is rich in sugars¹⁶, antioxidants¹³ and vitamin C (Azevedo and Rodrigues, 2000) and is widely consumed in Brazil, India, Nigeria etc¹⁶. Cashew apple juice has the potential to be a natural source of vitamin C and sugar in processed foods (De Carvalho et al., 2007)/ cashew apple juice has been consumed for years for several recreation and medicinal purpose, but not much has been done on it system protecting potentials.

Hematological parameters are important indices of the physiological and pathological status for both animals and humans^{17,18}. *Anacardium occidentale* caused significant increase in the value of RBC, PCV, Hb and monocytes while there was no changes observed in the of WBC value. Red blood cell and factors relating to it are major indices for evaluating circulatory erythrocytes and are significant in the diagnosis of anaemia and also serve as useful indices of the bone marrow capacity to produce RBC in mammals¹⁹. The significant increase in RBC and PCV following administration of *A. occidentale* may be an indication of erythropoiesis stimulation by the juice extract. The extract may have caused increase or prevented decrease in the rate of erythropoietin release in the

kidney, which is the humoral regulator of RBC production. A platelet count may be used to screen for diagnose of various diseases and conditions that can cause problems with blood clot formation^{20,21}. It may be used as part of the workup of a bleeding disorder, bone marrow disease, or excessive clotting disorder²². A high platelet count can happen when something causes the bone marrow to make too many platelets^{23,24}. When the reason is unknown, it is called primary, or essential, thrombocytosis. When excess platelets are due to an infection or other condition, it is called secondary thrombocytosis²⁵. A low platelet count can make it difficult for the blood to clot, putting a person at risk of excessive bleeding. The cause may be due to an inherited tendency to not produce enough platelets, but the cause may also be unknown. In other cases, it is due to an underlying medical condition²⁶. In this study, cashew fruit juice prevented thrombocytopenia induced by gentamicin across all groups.

Hemoglobin, the substance that gives color to red blood cells, is the substance that allows for the transport of oxygen throughout the body. If the level of RBC is too low the cells in the body will not get enough oxygen^{27,28}. In this work, it was observed that *A. occidentalis* fruit does not improve oxygen carrying capacity of the rat after been exposed to a toxin like gentamicin.

The spleen plays multiple supporting roles in the body. It acts as a filter for blood as part of the immune system^{29,30,31}. Old red blood cells are recycled in the spleen, and platelets and white blood cells are stored there. The spleen also helps fight certain kinds of bacteria that cause pneumonia and meningitis. Spleen is a centre of activity of the Mononuclear Phagocyte System and can be considered analogous to a large lymph node, as its absence causes a predisposition to certain infections^{32,33}. The spleen is an immunologic filter of the blood and it is made up of B cells, T cells, macrophages, dendritic cells, natural killer cells and red blood cells^{34,35}. In this study, aqueous fruit juice extract of *A. occidentalis* stimulated lymphoid follicular activation and sinus histiocytosis in the spleen which is capable of stimulating the production of more lymphocytes and histiocytes and eventually produced more antibodies, consequently improving the body immune/defence system. There was evidence of such activation in the experimental animals. Increase to near normal in the values of monocytes is also reflected in improvement of the body weight ratio, and histology of spleen of rats that received *A. occidentalis*.

Previous Phytochemical analysis has shown the presence of alkaloids, flavonoids, tannins, glycosides, saponins, terpenoids, vitamin C etc. in *A. occidentalis* leaf, fruit and nut. Some of these compounds have been demonstrated to posse's anti-inflammatory, antioxidant and

immunostimulatory potentials. It is possible that some these compounds may also be responsible for improved haematopoietic systems and tissue protective potentials observed in this study. Although the specific mechanism(s) through which the plant facilitated the increase in these hematological indices was not ascertained, this action is assumed to be a direct effect of the extract on the haematopoietic systems. It is possible that the extract contains such constituent(s) that can interact and stimulate the formation and secretion of erythropoietin, hematopoietic growth factors/committed stem cells as well as boost the immune system. Specifically, stimulations of hematopoietic growth factors and erythropoietin systems have been reported to enhance rapid synthesis of blood cells^{16,35}.

Conclusion

The results of this study also indicated that Cashew apple juice (*Anacardium occidentale* L.) may possibly serve as an acceptable blood booster in an anemic condition or prophylactic purpose. The Study also suggested that Cashew apple juice may be useful as an immune boaster.

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