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EFFECT OF *DETARIUM MICROCARPUM* FRUIT PULP ON THE HAEMATOLOGICAL INDICES OF RATS IN MUBI ADAMAWA STATE, NIGERIA

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Abstract

The study was carried out to investigate the effect of Detarium microcarpum fruit pulp on haematological parameters of rats, in 28 days. The research was aimed to investigate the effect of D. microcarpum on some haematological parameters of rats and weekly weight gain by the rats. 24 healthy Albino rats were used for the experiment. Data collected was subjected to oneway analysis of variance and Least Significant Difference (LSD) was used to separate the mean differences at 5% probability level. The results obtained revealed that the fruit pulp of D. microcarpum has significant effects on the haematological parameter and weight gain by the rats. There was a significant increase in Red Blood cells (RBC) of the experimental rats in groups A and C (3.19±0.09 and 3.21±0.04 respectively). The White Blood Cells (WBC) of the rats also increased significantly, while the Packed Cells Volume (PCV) and Haemoglobin (Hb) concentrations showed no significant differences between the groups. It was also revealed that treatment C has more effect on the weekly body weight gain by the rats. Generally, it can be concluded that the D. microcarpum fruit pulp has effects on the haematological parameters, and the body weight gained by the rats.

Key Words: *Detarium microcarpum*, Haemoglobin, Packed Cells Volume, Rat.

Introduction

Through the ages, plants have been used by humans as a source of food, cosmetics and medicine. Plants have served as the basis of supplementing traditional medicine for thousands of years, in countries such as India and China (Cragg, 1999). Trees and shrubs not only serve as fuel but also as a shade, shelter, food or fodder and as a source of important product such as resin and medicine (Banard *et al*, 1989). The plant parts commonly used for treatment of ailments include roots, stems, leaves, seeds, flowers or bark or a combination of any of the parts (Tella, 1979). Fruits of plants bark and leaves are used not only for texture and flavour, but also for their chemical and nutritional properties (Abulude *et al.*, 2004).

Medicinal plants are sold in large quantities and varieties in local markets to people in search of cures for particular ailments or as usually claimed, all kinds of diseases (Sofowora, 1993). Hence, there is a need to analyze the plants for their potency and efficacy. The origin of herbal medicine and its practices have been described by some authors (Wambebe, 1998; Akunyili, 2003).

Detarium microcarpum belongs to the family *Fabaceae*. The plant *D. microcarpum* is hard dark-brown wood which provides very good quality timber which is very durable under water and is used in carpentry and construction. It is also used as good quality fuel wood and charcoal. The leaves, stems, roots, barks, as well as the fruits have found tremendous usage in treatment of various ailments e.g. tuberculosis, meningitis, itching and diarrhea (Obun *et al.*, 2010). The fruit is edible and rich in vitamin C and the leaves and seeds are also used in cooking. However, the foliage is avoided by most large mammals (Keay, 1989; Dalziel, 1995). The powdered seeds are applied to skin infections and inflammations, whereas the fruit is eaten to cure meningitis and malaria (Obun *et al.*, 2010). The fruit pulp is used for treating skin infection. The seed flour has good nutritional quality and the functional properties confirmed their suitability for use in various food preparations. Glycosides and alkaloids are present in the leaves of *D. microcarpum* (Iwu, 1986). The seeds have no endosperm but they contain 7.4% oil and 3.2% protein (Baerts and Lehman, 2002).

The use of herbal medicine including *D. microcarpum* by humans for curing several ailments undoubtedly posses several mild effects since doses of such local preparations could not be determined. It is necessary that the fruits be investigated to study its effect on rats, which invariably could affect humans as the flesh is eaten raw and cooked by many ethnic groups, especially in the northern part of Nigeria. Hence, this research is aimed at investigating the effect of *Detarium microcarpum* fruit pulp on rats.

Materials and Methods

Study area

The study area is Mubi, a town which comprises of Mubi North and Mubi South local Government areas of Adamawa State. Mubi is located in the North Eastern region of Nigeria between latitude 10^{o} 14^{1} N and 10^{0} 18^{1} N of the equator and longitude 13^{0} 14^{1} E and 13^{0} 19^{1} E of the Greenwich Meridian. It occupies a land of about 725.85 km^{2} with an estimated population of about 300,000 people. The area has tropical climate with an average temperature of 32^{0}C and lies

within the Sudan Savannah vegetation zone of Nigeria. The area has an average relative humidity from 28% - 45% and annual rainfall of about 1056 mm (Adebayo and Tukur, 1999).

Collection of sample

Fresh fruit of *Detarium microcarpum* were collected from 'Borong' Demsa local Government Area of Adamawa state, and were brought to the Laboratory of Department of Biological Sciences, Adamawa State University Mubi for the experimental work.

Preparation of the powdered pulp

After the collection of fresh fruits of *D. microcarpum,* the matured fresh fruits were well sun-dried for period of 2 weeks and grounded using pestle and mortar, to get the pulp which was kept in a sealed container until use.

Experimental animals

Twenty-four healthy and mature male white albino rats weighing (160 – 260g) were obtained from the Animal Unit, Department of Biological Sciences, Adamawa State University. The rats were individually housed in cages at 12 hours light cycle.

Animal Feeding and experimental design

Test diets were formulated by mixing separately 10ml of palm oil (PO) with 500g of rat mash; 500g of *D. microcarpum* pulp with 500g of rat mash, the control diet contained mash only. The rats (n = 24) were allowed to acclimatized for two weeks prior to diet treatment. The rats were divide into three groups (n = 8 rats/group). The diet groups included the control group "A" (CG); palm oil group "B" (POG), and *Detarium* pulp group "C" (DPG). Water and feeds were provided *ad libitum* for the duration of the study (28days). Weekly weight gained, total feeds consumed were estimated, and at the end of the study, blood samples were collected from the *orbital sinus* under anaesthesia for haematological study (Ezekiel and Onyeyili, 2007), and the method of (Brown, 1976) was adopted for the haematological parameters.

Determination of White Blood Cells (WBC)

A well mixed venous blood was drawn using a WBC pipette of a haemosytomer to 0.5 marks; WBC diluting fluid was drawn to the mark. The blood and fluid were mixed gently, to avoid bubbles. A cover slip was placed on the counting chamber; the fluid blood mixture was then shaken and transferred using a micro pipette onto the counting chamber for 2 minutes, ensuring that the fluid does not get dried. It was also ensured that the mixture does not overflow the charged chamber and was mounted on the stage of microscope. Using the x10 objective lens of the microscope, the WBC was counted uniformly in the four large corner squares and the cells present on the outermost lines on the side were also counted while those that appeared on the opposite line were not counted.

$$WBC\ Count = \frac{Number\ of\ cells\ counted\ x\ blood\ dilution\ x\ depth\ factor}{Area\ of\ chamber\ counted}$$

$$WBC\ Count = \frac{Number\ of\ cells\ counted\ x\ 20\ x\ 10\ (depth\ factor)}{4}$$

$$WBC\ Count = Number\ of\ cells\ counted\ x\ 50$$

Determination of Red Blood Cells (RBC)

Blood was drawn to the 0.5 mark using the RBC pipette. The top was cleaned and diluting fluid was drawn to the 10 mark, it was then shaken for three minutes and the chamber was charged. The RBC was counted using x 40 objective lens of the micro scope in the 80 smallest square.

$$RBC\ Count = \frac{Number\ of\ cells\ counted\ x\ dilution\ \ factor\ x\ depth\ \ factor}{Area\ counted}$$

$$Dilution\ \ factor\ is\ 1in\ 200\ depths = \frac{1}{10}m$$

$$Area\ counted\ \frac{80}{400} = \frac{1}{5}mm2$$

$$RBC\ Count = \frac{Number\ counted\ x\ 200\ x\ 10}{\frac{1}{5}}$$

$$RBC\ Count = Number\ counted\ x\ 10{,}000$$

Determination of Packed Cell Volume (PCV)

Blood sample was collected through the jugular vein and it was brought to the laboratory for test. One end of the capillary tube was applied to the blood, which rose into the capillary tube by capillary attraction and surface tension. The capillary tube was 2/3 filled and was removed and the back was cleaned with a dry cotton wool; it was then sealed with a sealant. Both the capillary tubes were placed into groves of the centrifuge so that the sealed ends are away from the center. The cover of the centrifuge was fastened and was then spinned for five minutes at 10,000 rpm. The reading was taken with the help of a microhaematocrit reader.

Determination of Haemoglobin (Hb)

Haemoglobin was determined using haemoglobin meter. The graduated measuring tube of the haemoglobin meter was filled to the graduation line (mark 2) with Hcl and 20cl blood was sucked into the capillary pipette up to the mark, the pipette point was wiped and the blood was blown into the measuring tube. The good mixture of the liquid was achieved by repeating suction and blowing. Water was added by means of dropping pipette and it was mixed with glass stirrer until its color matches the test rods. The Hb was read directly from the tube (%).

Statistical Analysis

Data collected was subjected to oneway analysis of variance (ANOVA), Least Significant Difference (LSD) was used to determine the level of significance of the three treatment groups at 5% confidence level.

Results

Effect of Detarium microcarpum on mean Red Blood Cells (RBC) count of rats.

Table 1 shows the effect of *Detarium microcarpum*, significantly increased the Red Blood Cells (RBC) of rats in group A and C, and significantly decreases in group B at 5% probability level under 2 and 21 degree of freedom.

TABLE 1: Effect of *Detarium microcarpum* on RBC Count (10^3/mm^3), WBC Count (10^3/mm^3), PCV (%) and Hb(g/dl) Concentration on Rats.

PARAMETER	GROUP (Mean ± SD)			LSD 5%
	A	B	C	
RBC	3.19±0.09^a	3.29±0.08^b	3.21±0.04^a	0.072
WBC	2.28±0.31^a	2.33±0.16^a	2.68±0.09^b	0.22
PCV	39.39±0.09^a	37.65±4.16^a	39.64±3.92^a	3.61
Hb	12.136±0.51^a	11.76±1.19^a	12.44±1.09^a	21.0

Mean ± SD: Based on three observations and mean values carrying different superscript across the row are significantly different at 5% probability level.
Where RBC= Red Blood Cells, WBC= White Blood Cells, PCV= Packed Cells Volume, Hb= Haemoglobin.

Effect of Detarium microcarpum on White Blood Cells (WBC) count of rats.

The result revealed that there was a high significant increased in the WBC (10^3/mm^3) between the treatments, at 5% probability (Table 1).

Effect of *Detarium microcarpum* on Packed Cells Volume (PCV) of rats.

The result obtained on the *D. microcarpum* on PCV of the rats, revealed that there was no significant difference in the PCV (%) at 5% probability level (Table 1).

Effect of *Detarium microcarpum* on Haemoglobin (Hb) Concentration on Rats

There was no significant difference in the Hb (g/dl) concentration of rats between the treatments, at 5% probability level (Table 1).

Table 2 shows the weekly weight gained by the rats in A, B and C. in week 1, the rats in A, B, and C significantly gained weight.

TABLE 2: Effect of *Detarium microcarpum* on the weekly weight gained (g) by the rats

WEEK	GROUP (Mean±SD)			LSD 5%
	A	B	C	
1st	279.63±9.75^a	237.38±8.43^b	208±16.32^c	12.56
2nd	277.50±50.57^a	271.88±15.53^a	195.88±38.29^b	25.8
3rd	303.75±14.22^a	257±15.80^b	175.88±24.63^c	1.95

Mean ± SD: Based on three observations
Mean values carrying different superscript across the row are significantly different at 5% probability level.

There was no significant difference between the rats in group A and B in the 2nd week of the treatment, in weight gained. The weight gained by the rats in A and B are significantly higher than those in C at 5% and probability level (Table 2). In the 3rd week of treatment, the weight gained by the rats in A was significantly higher than B and C in the following order: A>B>C (303.75±14.22>257±15.80>175.88±24.63 respectively).

Effect of Detarium microcarpum on the feed intakes of rats

Table 3 shows the effect of *D. microcarpum* on the feed intake of the experimental rats.

TABLE 3: Effect of *Detarium microcarpum* on the feed intake of rats

PARAMETER	GROUP (Mean ± SD)			LSD 5%
	A	B	C	
Feed intake	1250±288.67[a]	1000±408.25[b]	1250±288.67[a]	1.04

Mean ± SD: Based on three observations

Mean values carrying different superscript across the row are significantly different at 5% probability level.

The result in Table 3 shows that group A and C significantly consumed equal feed (1250±288.67) at 5% level of significant, more than group B.

Discussion

The effect of *Detarium microcarpum* fruit pulp on the haematological indices of rats in Mubi Adamawa State was evaluated. From the results in Table 1, the RBC counts in treatment B (3.29±0.08) was significantly higher than treatments A and C (3.19±0.09 and 3.21±0.04 respectively). However, there was a slight increased observed in the WBC counts at treatment B (2.33±0.16) and a highly significant increase observed in the WBC counts at treatment C (2.68±0.09). This could be as a result of possible stimulation of immune defense system in the rats. This coincides with findings of Ezekiel and Onyeyile, (2007), who reported that persistent antigen load in the body, would result in an increase WBC counts. The long-term effect of the fruit pulp of *D. microcarpum* showed a slight increase in PCV at treatment C (39.64±3.92), and a very low decrease in treatment B (37.65±4.16). The decrease in PCV value (which is an indication of anemia) might have resulted from lesions in the intestine, which could have affected the digestion and absorption of nutrients (Ezekiel and Onyeyile, 2007). Also from Table1, the haemoglobin (Hb) of the experimental rats decreases in treatment B (11.76±1.19), and slightly increases in treatment C (12.44±1.09). This could be as a result of haemolysis, which might have allowed free haemogbolin in the plasma of the rats, or as a result of haemo-concentration due to dehydration observed in tubular urine (Pieme *et al.*, 2006).

In Table 2, the effect of *D. microcarpum* on the weekly weight gained by the rats revealed that there is no significant difference between the treatments A, B, and C, during the 1st week of treatment. In the 2nd week, treatment C recorded a lower body weight (195.88±38.29) than treatment A and B (277.5±50.57 and 271.88±15.53 respectively). And in the 3rd week all the treatments significantly differ from each other with treatment A recording the highest weight gain (303.75±14.22) followed by treatment B (257±15.80) and then treatment C (175.88±24.63), in the following order A>B>C. This revealed that treatment C has more effect on the weekly body weight gain by the rats.

Analysis also showed that treatments A and C significantly consumed equal feeds (1250±288.67), while treatment B consumed less feed (1000±408.25) (Table, 3). This could be as a result of incorporation of the *D. microcarpum*, which reduced significantly the total feed intake of the test rats. This supported the findings of Church and Pond (1988) who reported that when the energy contents of diets were increased, feed consumption decreased.

Conclusion

From the results, it can be concluded that the *Detarium microcarpum* fruit pulp has great effects on haematological parameters, and the body weight gained by the rats. This implies that the prolonged consumption of the fruit pulp of *D. microcarpum* by humans to cure certain ailments could affect the haematological parameters and body weight gain at excess medicinal doses.

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