

Effect Of Aqueous Extract of *Cola Nitida* (Kola Nut) On Haematological and Plasma Biochemical Parameters in Male Albino Rats

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Abstract: *Cola nitida* (Kola nut) is a caffeine-containing nut native to tropical North African and cultivated extensively in the new world tropics. Several studies have reported its effects on body weight and reduction in body fat without undesirable side effect as well as on gastric secretion, but there is a dearth of information on its effect on blood chemistry. This study was designed to investigate the effect of its aqueous extract on haematological and plasma biochemical parameters in male albino rats.

Aqueous extract of *Cola nitida* designated as AECON was prepared using cold maceration. The extract (25 mg/kg BW, 50 mg/kg BW, 100 mg/kg BW) was administered to the rats for 30 days for haematological and biochemical study. Distilled (0.5 ml) served as the control. Red Blood Cell (RBC) and Total White Blood Cell (TWBC) counts were determined using haemocytometer. Differential leucocyte count was done using the Schilling method. Activities of plasma Alanine Aminotransferase (ALT) and Aspartate Aminotransferase (AST) as well as total protein, creatinine and albumin levels were determined by spectrophotometry. Data were analysed using ANOVA at $p < 0.05$.

Treatment of rats with 25 mg/kg BW, 50 mg/kg BW and 100 mg/kg BW of AECON caused significant increase in monocyte values relative to the control, while 100 mg/kg BW produced significant increase in eosinophil value relative to the control. In addition, 50 mg/kg BW of AECON caused significant decrease in albumin and total protein levels relation to their controls.

These findings indicate that *Cola nitida* extract caused deleterious effect on the blood chemistry in male albino rats.

Key words: *Cola nitida*, Albino rats, Red blood cell, Total protein, Albumin.

I. Introduction

Cola nitida (Kola nut) belongs to the family of *Malvaceae*. It is commonly called kola nut in English language, “Guoro” in Hausa language, “Oji” in Igbo language and “Obi” in Yoruba language.

It is important in various social and religious customs and to counteracts hunger and thirst (Purgesleve, 1977). It is used as a gesture of peace, friendship and hospitality (Hatasaka and Goldstain, 2001). secretion

Cola nitida administration has been reported to stimulate gastric acid in cats (Osim et al., 1991). Its extract has been reported to induce behavioral changes in rodents such as increasing exploratory, general reactivity and locomotor activity in rats (Scotto et al., 1987). Its extract has also been reported to produce a dose-dependent biphasic effects on locomotion in mice (Ajarem, 1990). It has been reported to contain stimulants that prevent fatigue and dispel sleepiness (Oludemokun, 1982).

However, due to scarcity of information from literature on the effect of *Cola nitida* on haematological and reproductive parameters in male albino rats, this study therefore aims at investigating the effect of aqueous extract of *Cola nitida* on these aforementioned parameters.

II. Materials And Methods

Experimental Animals

Adult male albino rats weighing between 160 g and 180 g bred in the Animal House of Physiology Department, LAUTECH, Ogbomosho were used. They were housed under standard laboratory conditions with a 12 hours daylight cycle and had free access to feed and water; they were acclimatized to laboratory conditions for two weeks before the commencement of the experiments. All experiments were carried out in compliance with the recommendations of Helsinki’s declaration on guiding principles on care and use of animals.

Plant Materials

Fresh specimens of *Cola nitida* were bought from Ogbomosho market and authenticated in the taxonomy unit of the department of Pure and Applied Biology, LAUTECH, Ogbomosho.

Preparation of the Aqueous Extract of *Cola nitida* (AECON).

Large quantities (1.43 kg) of the fresh specimens of *Cola nitida* were washed and cut into smaller bits and air-dried for four weeks, the dried specimens were pulverized using laboratory mortar and pestle.

Weighed portion (480.20 g) of the pulverized specimens were macerated and extracted with distilled water (1:2 wt./vol.) for 72 hours at room temperature (26 – 28°C). The resulting solution was then filtered using a wire-gauze and a sieve with tiny pores (0.25 mm). The distilled water was later evaporated using steam bath to give a percentage yield of 12.4% of the stating material.

Ten grams of the aqueous extract of *Cola nitida* (AECON) was dissolved in 100 ml of distilled water to give a concentration of 0.1 g/ml.

Experimental Design

Twenty-four animals were randomly divided into four groups with each group consisting of six rats. The four groups of rats were subjected to the following oral treatments once a day for 30 days:

Group I rats received 25 mg/kg BW of AECON

Group II rats received 50 mg/kg BW of AECON

Group III rats receive 100 mg/kg BW of AECON

Group IV rats received 1.0 ml of distilled water as the control group.

Twenty-four hours (day 31) after the last dosing of the four groups, blood samples were collected.

Collection of Blood Sample

Blood samples were collected through the medial cantus into EDTA bottles for haematological and plasma biochemical studies. Before assays, the blood samples were centrifuge for 5 minutes using a bench-top centrifuge (Centromix) and the supernant plasma was then used for the determinations of the biochemical parameters.

Determination of Haematological Parameters

The red blood cells (RBC) and white blood cells (WBC) counts were determined by the improved Neubauer haemocytometer method. The haemoglobin (Hb) concentration was determined according to Jain (1986), using the cyanomethaemoglobin method. The packed cell volume (PCV) was determined by the micro-haematocrit method according to Dacie and Lewis (1991). Schilling method of differential leucocyte count was used to determine the distribution of the various white blood cells (Mitruka and Rawnsley, 1977). Mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were computed according to Jain (1986).

Determination of Plasma Biochemical Parameters

The total protein concentration was determined using the Bivret method (Reinhold, 1953) and the albumin concentration by the method of Doumas *et al.* (1971). The globulin concentration was calculated by subtracting the albumin concentration from the total protein concentration. Activities of plasma alanine transaminase (ALT) and aspartate transaminase (AST) were determined according to the method of Duncan (1994). The level of cratinine was determined using the method of Tietz *et al.* (1994). All the above biochemical parameter were determined in the plasma using the Randox kits.

Statistical Analysis

The mean and standard error of mean (S.E.M.) were calculated for all values. Comparison between the control and experimental groups was done using one-way analysis of variance (ANOVA) with Duncan's Multiple Range Test. Differences were considered statistically significant at $p < 0.05$.

III. Results

Treatment of rats with 25mg/kg BW, 50 mg/kg BW and 100 mg/kg BW of AECON for 30 days caused significant ($p < 0.05$) increase in monocyte values relative to the control, while 100 mg/kg BW of AECON produced significant ($P < 0.05$) increase in eosinophil value relative to the control; but 50 mg/kg BW and 100 mg/kg BW of AECON produced significant ($p < 0.05$) decreases in MCH and neutrophil values relative to their controls respectively. The administration of various doses of AECON to the rats caused non-significant ($p > 0.05$) changes in PCV, Hb, RBC, MCV, MCHC, TWBC, platelet and lymphocyte values relative to their controls.

Table 1: Effect of aqueous extract of *Cola nitida* (AECON) on haematological parameters after treatment of male albino rats for 30 days (n=6, *p<0.05).

Parameter	Control	25 mg/kg BW AECON	50 mg/kg BW AECON	100 mg/kg BW AECON
PCV (%)	50.20±1.88	50.40±1.12	46.60±2.21	46.20±1.36
Hb (g/dl)	15.58±0.52	15.56±0.51	14.58±0.67	14.72±0.51
RBC (× 10 ⁶ /μl)	7.91±0.32	8.06±0.38	7.68±0.47	7.12±0.42
MCV (FL)	63.51±1.01	62.87±1.87	60.87±1.17	65.38±2.19
MCHC (g/dl)	31.0±0.56	30.85±0.32	31.30±0.14	31.84±0.29
MCH (pg)	19.67±0.20	19.36±0.41	19.06±0.44*	20.83±0.80
TWBC (10 ³ /μl)	6.54±0.83	7.50±0.91	8.11±0.85	7.10±0.49
Platelets (×10 ⁵ /μl)	1.15±0.09	1.33±0.18	1.19±0.12	1.09±0.10
Neutrophils (%)	32.0±2.88	31.80±4.13	26.20±1.56	21.60±2.25*
Lymphocytes (%)	66.60±2.50	66.20±3.84	71.20±1.74	73.60±1.81
Eosinophils (%)	0.80±0.37	0.6±0.25	1.0±0.32	2.80±0.49*
Monocytes (%)	0.60±0.25	1.40±0.25*	1.60±0.25*	2.20±0.49*

Treatment of rats with 50 mg/kg BW of AECON caused significant (p<0.05) decrease in albumin and total protein levels relative to their controls, while 25 mg/kg BW of AECON caused significant (p<0.05) decrease in ALT level relative to the control. Treatment of rats with various doses of AECON caused non-significant (p>0.05) changes in globulin, creatinin and AST values relative to their controls.

Table 2: Effect of aqueous extract of cola nitida (AECON) on haematological parameters after treatment of male albino rat for 30 days (n = 6, *p<0.05)

Parameter	Control	25 mg/kg BW AECON	50 mg/kg BW AECON	100 mg/kg BW AECON
Total protein (gm%)	5.60±0.38	6.30±0.13	5.12±0.26*	5.88±0.19
Albumin (gm%)	2.14±0.21	2.58±0.17	1.84±0.14*	2.08±0.37
Globulin (gm%)	3.50±0.39	3.52±0.13	3.28±0.20	3.80±0.17
Creatinine	0.9±0.11	0.74±0.08	0.98±0.26	0.72±0.10
AST (μI)	22.00±3.08	20.40±1.50	21.80±1.02	17.00±1.92
ALT (μ/L)	18.60±2.72	12.00±1.38*	15.60±2.25	14.00±1.52

IV. Discussion

The haematological study has revealed that the extract caused increase in monocyte values which probably indicates an enhancement in the phagocytic function of the blood. Similar report was given by Biu *et al.* (2009) when they reported that aqueous extract of Neem caused significant increase in monocyte values. The extract also caused significant increase in eosinophil value which probably indicates the anti-allergic and anti-parasitic infectious responses of the blood have been boosted by the extract which is in consonance with the report of Biu *et al.* (2009). The extract caused significant decrease in MCH values which could indicate absence of induction of macrocytic anaemia, since increased MCH values are known to be indicative of macrocytic anaemia. This is in contrast with the report of Adedapo *et al.* (2007) which indicated that *P. amarus* and *C. acontifolius* extracts caused significant decrease in MCH values. The extract caused significant reduction in neutrophil value which could indicate a reduction in the ability of the body to attack and destroy invading bacteria, viruses and other injurious agents (phagocytosis). Contrary report was given by Yakubu *et al.* (2007) in rats treated with *Fadogia agrestis* extract. The administration of the extract has non-significant effect on the RBC count and indices relating to it (Hb, PCV, MCV and MCHC) which might indicate that there were no destruction of matured RBC and no change in rate of erythropoiesis. This also shows that the extract does not have the potential to stimulate erythropoietin release from the kidneys as well as being unable to effect changes in the oxygen-carrying capacity of the blood and the amount of oxygen delivered to the tissues since RBC and Hb are known to be very important in transferring respiratory gases. The extract caused non-significant change in the platelet value which probably indicates its inability to stimulate hemostasis.

The extract caused significant decrease in total protein level which probably indicates that the buffering capacity of the blood and the body fluid balance have been compromised. This is contrary to the report of Adewusi and Afolayan (2009) in rats treated with *Pelargonium reniforme* extract. The extract caused reduction in albumin level which might indicate a decrease in serum levels of metals, ions, fatty acids, amino acids,

bilirubin and enzymes. Contrary report was given by Adedapo *et al.* (2007) in rats treated with *P.amarus* and *C. acontifolius* extracts. The extract caused significant decrease in the activity of ALT which probably indicates an hepato-protective potential of the extract. The extract caused non-significant change in globulin level which could indicate that both the natural and acquired immunity of the body have not been compromised by the extract. The extract caused non-significant change in creatinine level which probably indicates that the integrity and functions of the nephrons have not been compromised by the extract. The extract also caused non-significant change in the activity of AST which could indicate an absence of tissue necrosis induction by the extract.

References

- [1] Adedapo AA, Abatan MO, Olorunsogo OO (2007): Effects of some plants of the spurge family on haematological and biochemical parameters in rats. *VETERINARSKI ARHIV* 77 (1), 29 – 38.
- [2] Adewusi EA, Afolayan AJ (2009): Safety evaluation of the extract from the roots of *Pelargonium reniforme* Curtis in male wistar rats. *Afr. J. Pharm and Pharmacology*, vol. 3(8): pp. 368 – 373.
- [3] Ajarem JS (1990): Effect of fresh kola-nut extract (*Cola nitida*) on the locomotor activities of male mice. *Acta Physiol. Pharmacol. Bulg.* (16) pp 10 – 15.
- [4] Biu AA, Yusufu SD, Rabo JS (2009): Studies on the effects of aqueous leaf extracts of Neem (*Azadirachta indica*) on haematological parameters in chicken. *African scientist* Vol. 10 No 4 pp 189 – 192.
- [5] Dacie JV, Lewis SM (1991): *Practical haematology*, 7th edition ELBS with Churchill Livingstone, England, pp 37-85.
- [6] Doumas BT, Watson W, Biggs HC (1971): Albumin standards and the measurement of serum albumin with bromocresol green. *Clinica Chimica Acta* 31, 87-96.
- [7] Duncan JR, Praise KW, Mahaffey EA (1994): *Veterinary Laboratory Medicine (Clinical Pathology)* 3rd ed. Iowa State University Press, U.S.A.
- [8] Hatasaka H, Goldstain M (2001): Kola nut as a symbol and its effect on the central nervous system. *J. sep.* 25: 257 (5): 2133 – 2136.
- [9] Jain NC (1986): *Schalm's Veterinary Haematology* 4th ed. Lea and Fabiger, Philadelphia.
- [10] Mitruka BM, Rawnsley H (1977): *Clinical, biochemical and haematological references values in normal experimental animals.* Masson Publishing USA Inc. Pp. 53-54.
- [11] Oludemokun AA (1982): Processing, storage, and utilization of kola nuts: *Cola nitida* and *Cola acuminata*. *Trop. Sci.* (24), pp 11 – 117
- [12] Osim EE, Arthur SK, Etti KM (1991): Influence of kola nuts (*Cola nitida*) on *in-vivo* secretion of gastric acid in cats. *Pharmaceutical Biology* vol. 29, No 3, pp 215 – 220.
- [13] Purgesleve JW (1977): *Tropical crops cotyledons.* Longmans London pp 564 – 612
- [14] Reinhold JG (1953): *Manual determination of serum total protein, albumin and globulin fractions by the Biuret method* *Standard Methods of Clinical Chemistry* (Academic Press, New York).
- [15] Scotto G, Maillard C, Vion-Dury J, Balansard G, Jadot G (1987): Behavioral effects resulting from subchronic treatment of rats with extract of fresh stabilized cola seeds. *Pharmacol. Biochem*, (26) pp 841 – 845.
- [16] Tietz NW, Prude EL, Sirgard – Anderson O (1994): *Textbook of clinical chemistry.* Ed. Burtis C.A. and Ashwood E.R. pp 1354 – 1374. W.B. Saunders Company, London.
- [17] Yakubu MT, Akanji MA, Oladiji AT (2007): Haematological evaluation in male albino rats following chronic administration of aqueous extract of *Fadogia agrestis* stem. *Pharmacog. Mag.* 3: 34 – 38.