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Full Length Research Paper

Effects of *Cucumis metuliferus* (*Cucurbitaceae*) fruits on enzymes and haematological parameters in albino rats

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The effects of the powdered fruits of *Cucumis metuliferus* on enzymes and haematological indices were evaluated in adult albino rats. The study revealed a significant (P<0.05) dose-dependent decrease in white blood cells (WBC) count. 500 mg/kg body weight of the powdered fruit produced a significant (P<0.05) decrease in red blood cells (RBC), and an increase in platelet and haemoglobin (Hb), while there was an insignificant (P>0.05) decrease in clotting and bleeding time. 1000 mg/kg produced significant (P<0.05) increase in RBC, platelets, Hb and packed cell volume (PCV) values, and an insignificant (P>0.05) decrease in clotting and bleeding time. The biochemical parameters evaluation showed that 500 - 1000 mg/kg of the powdered fruit of the plant produced a dose-dependent significant (P<0.05) increase in the levels of serum alkaline phosphatase (ALP), aspartate aminotransferase (AST), alanine aminotransferase (ALT), Blood urea nitrogen (BUN) and Total protein. This result showed that *Cucumis metuliferus* produced alterations in the haematological and biochemical indices evaluated.

Keywords: Cucumis metuliferus, serum, enzymes, haematological.

INTRODUCTION

Cucumis metuliferus belongs to the family *Cucurbitaceae*, and is a monoecious, climbing, annual herb that can be grown practically anywhere, provided the season is warm (Benzioni et al., 1993). The plant is endemic to the semiarid regions of Southern and Central Africa (Morton, 1987), the fruits are ovoid berries of 8-10 cm long and 4 - 5 cm in diameter, reddish orange at maturity, hanging, covered with strong spiny outgrowths; and the seeds are embedded in the mesocarp which is emerald green and consists of juicy, bland – tasting tissues. It is commonly known as African horned cucumber, melano, Jelly melon, and more recently, kiwano. The fruits occur in two forms-the bitter and non-bitter forms, which occur mostly in the wild state. The bitter form contains cucurbitacins (triter-penoids), which is a highly toxic compound (Teuscher and Lindequist, 1994). The non-bitter form has been found to be less toxic and has also been widely cultivated (Enslin et al., 1954; Andeweg and De Bruyn, 1959).

The fruits of the non-bitter form have been claimed to cure HIV/AIDS positive patients in and around Jos of Plateau State, Nigeria (personal communication, 2006). It has been reported that the fruits and seeds of *Cucumis metuliferus* are eaten raw as supplements by local populations of Africa (Bruecher, 1977; Keith and Renew, 1975). Reports have also shown that the seeds can be ground into fine flour, made into an emulsion with water, and then eaten to expel parasites from the body (Chiej, 1984).

Available literature is deficient on documented evidence of the safety and efficacy of this plant species. Notwithstanding, the non-bitter fruit of the plant is consumed extensively on the basis of therapeutic claims by traditional herbalist. The aim of this study, therefore, is to investigate the effect (if any) of the fruit on some haematological indices and hepatic tissues in laboratory animals.

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Treatment (mg/kg)	RBC(cells/m) (x10 ⁶)	WBC (cell/mm ³) (10 ⁴)	Platelets (cells/mm ³) (10 ⁵)	Hb (g/dl)	PCV (%)	Clotting time (min)	Bleeding time (min)
Control	6.45±31.32	2.40±45.47	1.81±11.11	9.11±0.18	49.00±0.75	2.34±3.88	2.88±8.01
500	5.83±31.75*	1.71±16.01*	1.90±8.77*	10.36±0.06*	49.00±2.46	1.67±8.47	2.86±8.34
1000	6.67±54.28*	1.62±82.55*	2.21±3.85*	9.94±0.06*	57.00±0.75*	1.41±4.83	2.40±5.55

 Table 1. Effect of C. metuliferus fruits on some haematological parameter in rats.

P<0.05 compared to control. RBC = Red blood cells, WBC = white blood cells, Hb = haemoglobin and PVC = packed cell volume.

MATERIALS AND METHODS

Plant collection and authentication

The ripe fruits of the non-bitter form of *C. metuliferus* were collected from Rukuba Barrack road and Babale area of Jos, Plateau State, Nigeria in September, 2006. The whole plant and fruits were identified and authenticated by D. L. Wonang of the Department of Botany, University of Jos, Nigeria.

Preparation of the powder

The mesocarp of the fruits which is greenish and consists of juicy bland-tasting tissues and seeds were carefully scooped out of the pericarp using a spatula, and well stirred to separate the yellowish fiber portions which were then sun-dried. To separate the seeds from the greenish fluidy portion, sieves of different sizes that only allowed the fluidy solution to pass, thus retaining the seeds, were used. The resultant mixture was then spread in trays and placed in an oven set at 68°C until it was dried. It was then mixed with the yellowish sun-dried fibers portion and then pounded to powder using pestle and mortar. The powder was stored at room temperature in an airtight container prior to use.

Test animals

Male albino rats (wistar strain) weighing between 150 and 260 g bred in the Animal House of the University of Jos, Nigeria were used in this study. Food and water were supplied *ad libitum*.

Administration of Cucumis metuliferus

The rats were randomly allocated into three groups of five animals each. Animals in group 1 (control group) which administered equivolume (per kg body weight). Animals in groups 2 and 3 were respectively administered 500 and 1000 mg/kg orally of the powder dissolved in distilled water, using orogastric tube daily for a period of four weeks.

Haematological studies

At the end of four weeks, bleeding and clothing time of all the rats in the each group were determined using the method of Dacie and Lewis (1984). The rats were then anaesthetized with phenobarbitone 60 mg/kg intraperitoneally and their respective blood samples were collected from femoral vein in heparinised tubes. The following haematological indices were determined: red blood cells (RBC) count, white blood cells (WBC) count, platelets, haemoglobin (HB) and packed cell volume (PCV) by methods as described by Barbara and Brown (1980).

Enzymes analysis

The bloods collected as described above were centrifuged for 10 min at 1000 rpm to obtain the sera. Biochemical studies for blood urea nitrogen (BUN), aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), and total protein were determined using Randox test Kits.

Statistical analysis

All results were expressed in mean \pm SEM, and tests of significant differences between the means were carried out by students' *t*-test and a probability value of P <0.05 was considered to be statistically significant.

RESULTS AND DISCUSSION

From the result (Table 1), the oral dose of 500 mg/kg of *C. metuliferus* fruit powder produced a significant (*P* <0.05) decrease in RBC and WBC count, an increase in platelet count and Hb, while the decrease in clotting and bleeding time was not statistically significant compared to the control. However, at dose point of 1000 mg/kg, there was a significant (*P* <0.05) increase in RBC, platelets counts, Hb, and PCV values compared to the respective controls, while there was a significant dose-dependent decrease in WBC count. The decrease in clotting and bleeding times was not statistically significant (*P* >0.05).

The results of enzymes analysis revealed that there was a significant dose-dependent increase in the levels of BUN, AST, ALT, ALP, and total protein.

Results on the effect of C. metuliferus fruit on some liver enzymes are as shown in Table 2. The result observation showed dose-dependent increase (P < 0.05) in the levels of BUN, AST, ALT, ALP, and total protein compared to the controls. It has been demonstrated that changes in serum enzymes concentration compared to the control is a signal of an underlying pathological process (Hayes et al., 2002). It has also been reported that increase in enzymes concentration in the serum directly indicates major pathologic changes in cell membrane permeability or hepatic cell rupture (Benjamin, 1978). Both ALT and AST are located in the cytoplasm and mitochondria of liver cells, and also in cells of the heart, skeletal muscles, kidney and brain (Benjamin, 1978; Ringer and Dabieh, 1979). According to these authors, the activities of ALT outside the liver are low and

Treatment	BUN	AST	ALT	ALP	Total protein
(mg/kg)	(mmoL/L)	(IU/L)	(IU/L)	(IU/L)	(g/L)
Control	3.77±0.12	134.00± 4.52	48.33±1.70	384.57±3.23	63.83±1.39
500	4.87±0.16*	309.00±3.34*	57.67±0.59*	513.67±27.00*	64.43±1.15
1000	5.67±0.13*	427.33±4.87*	76.67±4.12*	553.70±13.51*	67.83±1.10*

Table 2. Effect of C. metuliferus fruits on some serum enzyme levels in rats

*P<0.05 compared to control. BUN = Blood urea nitrogen, AST = aspartate aminotransferase, ALT = alanine aminotransferase and ALP = alkaline phosphatase.

therefore this enzyme is considered more specific for hepatocellular damage. Alkaline phosphatase are widely distributed in the body with significant activities in the liver, gastrointestinal tract, bone and placenta (Klaassen and Watkins, 1999). The observed increase in the serum levels of these enzymes indicates the extent of cellular damage on the liver, more specifically the cell membrane, permitting the leakage of the said enzymes from the liver. Also, the dose-dependent increased levels of BUN signals pathological effects on the kidney cells, since it is mainly excreted through this organ.

The hematological result (Table 1) showed that *C*. *metuliferus* fruit, significantly decreased (P<0.05) the RBC and WBC counts in the group treated with 500 mg/kg, and an increase (P<0.05) in RBC was recorded in the 1000 mg/kg treated group and a decrease in WBC compared to the control. The result showed significant (P<0.05) increase in Platelets, Hb, and PCV. The decrease in clotting and bleeding time was not significant.

Increase in Hb, PCV and RBC is an indicator that the rats were not anaemic, while decrease level is a sign of anaemia. PCV measures the percentage by volume of packed RBC in a whole blood sample after centrifugation (Wynne and Edwards, 2003), while HB test measures the amount of HB in grams in 1 dl of whole blood and provides an estimate of oxygen carrying capacity of the RBCs. From the result (Table 1), it can be deduced that *C. metuliferus* fruit produced significant increase (*P* <0.05) in the production of HB, PCV and hence RBC by the bone marrow. This is useful in increasing the oxygen carrying capacity hence, increasing tissue oxygenation.

WBC count is the number of WBC in a cubic millimeter of whole blood; and is usually important in fighting against infections (Schalm et al., 1975). The significant decrease in WBC (Table 1) indicates low level of infection in the experimental rats, or may be related to suppression of the production of the WBC resulting from toxic reactions to substances.

Platelets are involved in blood clotting and vital to the formation of a haemostatic plug after vascular injury. The result in Table 1 shows a statistically significant (P < 0.05) increase in the platelets count in the treated groups compared to the control groups. This may be related to the corresponding insignificant decrease in the clotting time observed in the rats. Normally, increase in platelets should lead to increase in bleeding and decrease in clott-

ing time. The insignificant decrease in bleeding time (P > 0.05) might be due to the presence of some defective platelets granule, giving rise to storage pool disorders or it might be as a result of the ability of the plant to inhibit platelet cyclo-oxygenase, preventing the conversion of arachidonic acid to thromboxane B₂ (Craig et al., 2002). This mechanism may be useful in the treatment of inflammation (We are currently investigating this in our laboratory).

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