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### Research Article

# Potential haematopoietic properties of crude methanolic seed extract of *Pentaclethra macrophylla* in Wistar rats.

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ABSTRACT: Numerous medicinal properties have been attributed to the seeds of Pentaclethra macrophylla (P. macrophylla) whereas its haematological properties have received little attention. This study was designed to evaluate the haematopoietic effects of crude methanolic seed extract of P. macrophylla in wistar rats. Thirty Wistar rats in five groups, labeled A to E were acclimatized for two weeks and used for this study. Groups A to D were administered orally with graded-doses of crude methanolic extract of P. macrophylla seeds (100, 200, 300 and 400) mg/kg body weight respectively for 28 days whereas Group E served as control. Blood sample was collected and haematological parameters were determined. Extracts of P. macrophylla significantly (p<0.05) increased the Hb, Hct, RBC and TWBC in a dose-dependent pattern in the treated groups compared to the control. The platelet count was however decreased in the treated groups compared with the control, although not in a dose-dependent pattern, with Group D having the least count (p<0.05). Thin blood films of the treated groups revealed normocytic and normochromic red blood cells and relative lymphocytosis. Administration of crude methanolic seed extract of Pentaclethra macrophylla demonstrated dosage-dependent significant increase in some haematological variables, while decreasing platelet count. This study suggests that the seed extract may possess haematopoietic, antimicrobial and immunity boosting potentials, and could be developed for future clinical use in managing blood-related conditions.

KEYWORDS: Graded-doses; Oral administration; Pentaclethra macrophylla.

#### INTRODUCTION

Despite the availability of orthodox medical treatments, significant parts of the population in developing countries currently depend on herbal preparations for the treatment of various diseases (Nwabuise, 2002). In Nigeria, traditional and herbal healing systems play an important role in healthcare delivery, and about 70-80% of the population depends on traditional healers for most of their ailments (Akah *et al.*, 1998).

Pentaclethra macrophylla (P. macrophylla) is a perennial legume tree commonly planted in Nigeria as shade trees and in some communities as cash crops. The fruit is a black, hard and woody pod measuring about 35–36 cm long and 5–10 cm broad. The glossy brown seed is about 5–7 cm in diameter and weigh between 15–20 g (Odunfa, 1986). P. macrophylla (Ugba in Igbo language) is consumed by an estimated 15 million people in Eastern Nigeria, majority of who are Igbos (Odunfa and Oyeyiola, 1985). It is an important and cheap source of protein (Obeta, 1983).

The extract of *P. macrophylla* seed have been known to posses' anti-inflammatory and anti-helmintic activity, and are used to treat gonorrheal infections and convulsion, and also used as analgesic (Cousins and Huffman, 2002). The seed extract is used to treat arthritis, pulmonary infections, dysentery, venereal diseases, leprosy, and as an antidote (Oliver-Bever, 1983). It has been reported that the administration of the fermented seeds as a food supplement has greatly reduced the risk of cancer and some tobacco related diseases.

Phytochemical analyses have revealed that *P. macrophylla* seed contains carbohydrates, oleic acid, linoleic acid, lignoceric acid, palmitic acid, saponins and crude protein that contains the nutritionally essential amino acids (Onwuliri *et al.*, 2004; Odoemelam, 2005). However, the raw seeds were found to contain some anti-nutritional factors such as cyanide, phytate, tannin, alkaloid and oxalate. Full fermentation of the seeds caused a reduction in the anti-nutritional factors (Onwuliri *et al.*, 2004).

Despite the availability of extensive information on the potential medicinal and nutritional value of *P. macrophylla*, there is little known about the haematological effects of the plant.

This study is aimed at determining the haematopoietic effects of crude methanolic seed extract of *Pentaclethra macrophylla* in Wistar rats. The hematological variables examined include hemoglobin (Hb), haematocrit (Hct), red blood cell count (RBC), total white blood cell count (TWBC), platelet count and thin blood film report.

Our findings suggest that the use of this crude extract as a herbal remedy may lead to either bone marrow stimulation for haematopoiesis or suppression leading to anaemia.

#### **MATERIALS AND METHODS**

The seeds of *Pentaclethra macrophylla* were obtained and authenticated by a taxonomist in the Department of Botany, University of Nigeria, Nsukka, and a voucher specimen was kept in the herbarium for future reference.

Thirty Wistar rats, weighing 100–200 g and aged 2–3 months were used for the study. The animals were purchased and housed in the Animal House of the College of Medicine, University of Nigeria, Enugu Campus. They were allowed to acclimatize for two weeks, during which they were fed with commercially available rat feed and allowed access to the feed and water ad libitum.

Two hundred grams of ground, shade-dried powder of *Pentaclethra macrophylla* seeds were subjected to extraction with methanol and the resultant mixture was sieved. The remaining methanol in the extract was evaporated to get the concentrated crude extract, after which it was reconstituted with 3% Dimethyl sulphoxide (DMSO). The extract was kept refrigerated at 4 °C until required for use.

Wistar rats (n=30) were divided into 5 groups of 6 rats per group. Groups A to D were orally administered with graded-doses of the crude seed extract (100 mg, 200 mg, 300 mg, and 400 mg) /kg bodyweight for 28 days. Group E served as control and was orally administered with 3% DMSO only, which was used to dissolve the crude seed extract. On Day 29, blood samples (3.0 ml) were collected from each rat through the median canthus of the eyes into  $K_3$ -EDTA anticoagulant containers for the haematological analysis using standard operative procedures as described by Dacie and Lewis (2006).

The Statistical Package for Social Science (SPSS) computer software version 15 was used for data analysis. The results of the tests were analyzed using student's t-test at 95% confidence interval with p-value of  $\leq 0.05$  being considered as significant. The results were expressed as mean  $\pm$  standard deviation ( $\pm$  SD).

## **RESULTS AND DISCUSSION**

This study is aimed at evaluating the haematopoietic effects of crude methanolic seed extract of *Pentaclethra macrophylla* in Wistar rats. The results revealed dosage-dependent significant increases (p<0.05) in the values of Hb, Hct, RBC and TWBC of the treated groups compared to animals in the control group. Blood films of the treated groups revealed normocytic and normochromic red blood cells and relative lymphocytosis.

Figures 1–4 show the mean  $\pm$  SD of the haematological parameters assayed in the treated and control rats in this study. The results revealed dosage-dependent significant increase (p<0.05) in hemoglobin (Hb) (Figure 1), haematocrit (Hct) (Figure 2), red blood cell count (RBC) (Figure 3), and total white blood cell count (TWBC) (Figure 4) in the treated groups compared to control.

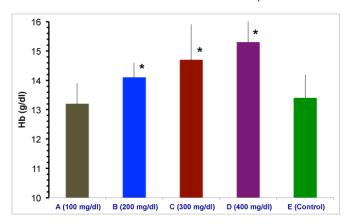


Figure 1: Effects of crude methanolic seed extract of *Pentaclethra macrophylla* on haemoglobin (Hb) concentration in Wistar rats. Each bar represents the mean  $\pm$  standard deviation of values from 5 rats per treatment group. \* = p<0.05; statistically significant compared to control.

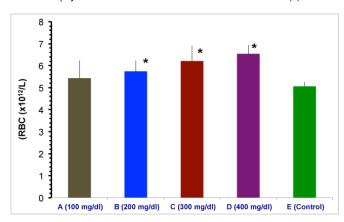


Figure 3: Effects of crude methanolic seed extract of *Pentaclethra macrophylla* on red blood cell count (RBC) in Wistar rats. Each bar represents the mean  $\pm$  standard deviation of values from 5 rats per treatment group. \* = p<0.05; statistically significant compared to control.

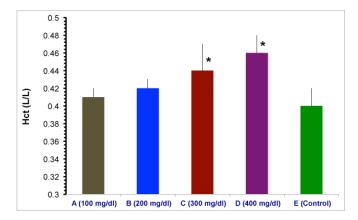


Figure 2: Effects of crude methanolic seed extract of *Pentaclethra macrophylla* on haematocrit (Hct) in Wistar rats. Each bar represents the mean  $\pm$  standard deviation of values from 5 rats per treatment group. \* = p<0.05; statistically significant compared to control.

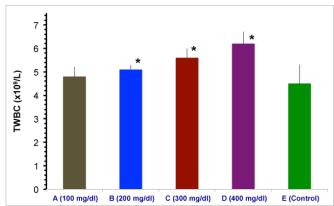


Figure 4: Effects of crude methanolic seed extract of *Pentaclethra macrophylla* on total white blood cell count (TWBC) in Wistar rats. Each bar represents the mean  $\pm$  standard deviation of values from 5 rats per treatment group. \* = p<0.05; statistically significant compared to control.

In contrast to the pattern observed with the other haematological parameters, there was a decrease in platelet count in the treated groups compared with the control (Figure 5), although not in a dose-dependent pattern, with group D having the least count (p<0.05). Thin blood films of the treated groups revealed norrmocytic and normochromic red blood cells and relative lymphocytosis.

The observed increase in Hb, Hct and RBC suggests that this crude seed extract may possess anti-anaemic potentials that could be developed for clinical use. The observed leucocytosis (lymphocytosis) suggests potential antimicrobial properties (Cousins and Huffman, 2002; Okwu, 2001) and immunity boosting potentials since lymphocytes are responsible for antibody production. These observed changes have demonstrated that crude methanolic seed extract of *Pentaclethra macrophylla* may probably possess

haematopoietic properties which manifest as the dosage of the seed extract increases. The observed haematopoietic effects could be attributed to some of the phytochemical constituents of Pentaclethra macrophylla seed extract which include carbohydrates, oleic acid, linoleic acid, lignoceric acid, palmitic acid, saponins and crude protein which contain all 20 amino acids required for normal growth and repair process (Onwuliri et al., 2004; Odoemelam, 2005). These nutrients may in themselves account for the boost in the haematological properties observed in the treatment groups. The observed normocytic and normochromic red blood cells suggests that this seed extract may possess haematopoietic properties. The decreased platelet count in the treated animals in the present study compared with the control should also be a source of concern to users of the extract, since it may actually affect coagulation in subjects being treated with the plant.

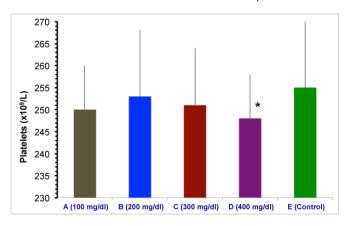


Figure 5: Effects of crude methanolic seed extract of *Pentaclethra macrophylla* on platelet count in Wistar rats. Each bar represents the mean  $\pm$  standard deviation of values from 5 rats per treatment group. \* = p<0.05; statistically significant compared to control.

Further studies are needed to fractionate the crude seed of Pentaclethra macrophylla using column extract chromatography and gas chromatography-mass spectrometry (GC-MS) in other to characterize the active constituent that mediates the observed effects on the haematological variables in the wistar rats. The crude methanolic seed extract of Pentaclethra macrophylla have demonstrated dosage-dependent significant increase in some haematological variables, while decreasing platelet count. The study suggests that the seed extract may possess haematopoietic, antimicrobial and immunity boosting potentials, and could be developed for future clinical use in managing blood-related conditions.

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