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Haematopoietic effect of ethanolic leaf extract of *Piliostigma thonningii* on male albino rat

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ABSTRACT

This research was an attempt to provide an ameliorative measure for patients with haematological disorders by considering the effect of ethanol leaf extract of *P. thonningii* on some haematological parameters of male albino wistar rats. Twenty (20) male albino Wistar rats were randomly assigned on the basis of average body weight into four (4) groups of five (5) rats each. While group A served as the control and received distilled water of treatment equivalence, group B, C and D were administered with 50,100 and 200mg/kg body weight of the extract respectively via oral gavages and treatment lasted for twenty-one (21) days. The extract produced a significant ($p < 0.05$) increase on white blood cells (WBC), and Monocytes at various dosages administered. However, a significant ($p < 0.05$) decrease was recorded for neutrophils. The red blood cells (RBC), packed cell volume (PCV), haemoglobin (HB), lymphocytes and platelets levels all recorded a significant ($p < 0.05$) increase at 100, and 200mg/kg body weight. Sequel to results, the ethanolic extract of *P. thonningii* appears to possess haematopoietic potentials.

Keywords: Haematopoietic, *Piliostigma thonningii*

INTRODUCTION

Herbal medicine (phytomedicine) refers to using plant's seeds, berries, roots, leaves, bark, or flowers for medicinal purposes. Herbalism has a long tradition of use outside of conventional medicine. It is becoming more main stream as improvements in analysis and quality control along with advances in clinical research show the value of herbal medicine in the treating and preventing disease (Atschuler, 2007). All cultures from ancient time to the present days have used plants as a source of medicine (Elujoba, 2005). Today according to the World Health Organization (WHO), estimates that 4 billion people, 80 per cent of the world population, presently use herbal medicine for some aspect of primary health care using plant extract (Kenneth, 2003).Some of the plant extract have been found nourishing while others were harmful due to toxic nature of the constituents of such plant (Sofowora, 1986).

P.thonningii is a plant with innumerable medicinal roles ranging from aphrodisaic, antioxidant,anticancer, anti malarial, anti inflammatory and anti cough (Igoli *et al.*, 2005; Jimoh and Oladiji, 2005 ;Kwaji *et al.*, 2010;Dasofunjo *et al.*,2012).In Nigeria, the plant is found growing abundantly as a wild uncultivated tree in many locations such as Ogun, Bauchi, Kwara, Lagos (Jimoh and Oladiji, 2005) and some parts of Kogi , Benue and Nasarawa States. Its local names include abefe in Yoruba, Kalgo in Hausa, okpoatu in Igbo (Rabo and Sanusi, 2001; Odukoya, 2002; Igoli *et al.*, 2003; Oguche *et al.*, 2004; Edeoga *et al.*, 2005; Jimoh and Oladiji, 2005; Aderogba *et al.*, 2006; Brummitt *et al.*, 2007; Sofowora, 2008; Ayisire *et al.*, 2009; Ozolua *et al.*, 2009); nyihar in Tiv ,omepa in Igede and ejei-jei in Igala (*Dasofunjo et al.*,2012) languages of Nigeria. In the absence information on it effects on

the haematological indices, this research aimed at accessing the effect of ethanolic leaf extract of *P.thonningii* on some haematological parameters of male albino rats.

MATERIALS AND METHODS

Collection and Preparation of Plant Materials

Fresh *P. thonningii* leaves were obtained from Mkar hills, Gboko, Benue State, North Central Nigeria. Identification and authentication was done at the Federal College of Forestry Jos, Plateau State, Nigeria, with the voucher number #25. The leaves were collected and air dried for 14 days until constant weight was obtained. The dried leaves were then pulverised after which 300g was soaked in 1000mls of Ethanol and agitated, then allowed to stay in refrigerator for 48 hours at 4°C. The mixtures were first filtered with cheese cloth, then with WhatMan No 1 filter paper (24cm). The filtrates were separately concentrated using water bath (Model RE52A, China) to 10% of its original volume at 37° C - 40° C.

Laboratory Animals

Twenty (20) male albino rats of wistar strain were obtained from the animal holding unit, Department of Chemical Sciences, University of Mkar, Mkar, Nigeria and were allowed acclimatization period of seven (7) days in well ventilated room with a temperature and relative humidity of 29±2°C and 70% respectively. They were maintained with rat chow (Vital Feeds LMT) and water *ad libitum*. The animals were housed in a cage and were exposed to 12 hour light-dark cycle and handled according to standard protocol. At the end of the acclimatization period, they were divided into four groups of five (5) each. Group A serve as the control, while B, C and D were the test groups. The control group were treated with distil water using orogastric tube. Test groups B, C and D were treated with 50,100 and 200 mg/kg body weight of the extract respectively. The administration of the extract lasted for 21 days period after which the animals sacrificed after 24hrs after the last administration in accordance with the guidelines of the European Convention for the Protection of Vertebrate animals and other scientific purposes –ETS-123 (European Treaty Series, 2005).

Determination of Haematological Parameters

At the end of the treatment period, the animals were anaesthetized in chloroform vapour and the blood collected via cardiac puncture into a plane tube. Heparinised test tubes were used to collect blood samples for haematological indices assay, i.e. White blood cells (WBCs), Red blood cells (RBCs), packed cell volume (PCV), Neutrophil, Lymphocytes, Monocytes, platelets, Haemoglobin (Hb), were assayed by automated techniques using the Elexes 2010 and Sysmex Automated machine respectively.

Statistical Analysis

The data were analyzed using a one-way ANOVA (in SPSS package) and the results expressed as Mean±standard deviation. All p-values <0.05 were considered significant (Mahaja, 1997).

RESULTS

The results of assessment of the effect of ethanolic leaf extract of *P. thonningii* on the haematological parameters of male wistar rats is presented in table 1 base on 50mg, 100mg and 200mg/kg body weight.

Statistical evaluation reveals that the extract recorded a significant ($p<0.05$) increase on the levels of red blood cells (RBC), white blood cells (WBC), packed cell volume (PCV), platelet and Hb only at 100 and 200mg/kg body weight. However, a significant ($p<0.05$) decreased on was observed at 50mg/kg body weight in a consistent similar trends. More so, statistical analysis also shows that the extract of *Piliostigma thonningii* also produced a significant ($p<0.05$) decrease on neutrophyll level while a significant ($p<0.05$) increase was observed for monocytes at 50, 100 and 200 mg/kg body weight.

Table 1 Effect of ethanolic leaves extract of *Piliostigma thonningii* on haematological parameters

	WBC (x10 ⁵ /L)	RBC (x10/L)	PCV (%)	PLATELET (x10 ⁵ /L)	Hb (g/dl)	NEU (%)	LYMP (%)	MONO (%)
A (CONTROL)	4.00±1.1	4.32±0.2	25.70±0.2	270±1.2	8.90±0.2	31.38±1.0	50.80±0.2	17.84±1.2
B(50mg/kg)	4.64±1.2*	3.84±0.1	20.94±0.1	234±1.0	7.96±0.3	29.94±0.5	47.80±1.1	22.26±1.0
C(100mg/kg)	4.98±1.0*	4.74±0.1*	29.76±0.1*	277.8±0.3*	9.70±0.2*	27.62±0.2	52.96±1.2	19.44±0.2
D (200mg/kg)	4.57±1.1*	4.64±0.2*	27.32±0.2*	278.2±0.1*	9.46±0.1	22.8±1.2*	59.20±1.3*	18.20±0.1

Results are expressed in mean ± SD (n=5). *significant at P<0.05 compared with the control.

WBC: White Blood Cells, RBC: Red Blood Cells, PVC: Packed Cell Volume,

Hb: Haemoglobin, NEU: Neutrophils, LYM: Lymphocytes, MONO: Monocytes

DISCUSSION

The effect of ethanolic leaves extract of *piliostigma thonningii* on haematological parameters was evaluated, analysed and interpreted. The assessment of haematological parameters is a biomarker for evaluating the haematotoxic potential of the extract in area of pharmacognosy (Aboyade, *et al.*, 2009). Therefore, the significant (p< 0.05) increase recorded for WBC following the administration of ethanolic leaf extract of *p. thonningii* suggest that the extract contain some bioactive constituents or phytoconstituents which should have imposed or boosted the production of WBCs and hence the immune system. It has also been reported that granulocytes, macrophage colony stimulating factor interleukons IL-2, IL-4 and IL-5 regulate the proliferation, differentiation and maturation of committed stem cells responsible for the production of WBC (Guyton and Hall, 2000, Ganoy, 2001). Therefore, it may be inferred that the bioactive constituents of the extract most especially the flavonoids increased the production of regulatory factors or interfered with the sensitivity of the committed stem cell at 100 and 200mg/kg body weights as also observed by Johnson *et al.*, (2012). Hence, it might have an effect on foreign materials and pathogenic organisms. Similar trend (increase) observed for both Hb and PCV at 100 and 200mg/kg body weight is an indication that the extract might have an haematopoietic potential or that the extract aids the incorporation of haemoglobin in the RBC. Conclusively, *piliostigma thonningii* leaf appears to be aid full in the management anaemic conditions since it possess erythropoietic potential.

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