

SJMLS-9(3)-018

Haematological Effect of *Tridax Procumbens* Methanolic Extract in Wistar Rats

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<https://dx.doi.org/10.4314/sokjmls.v9i3.18>**Abstract**

This study seeks to assess the haematological effect of *Tridax procumbens* methanolic extract (TPME) in Wistar rats. *Tridax procumbens* is a common procumbent plant with several therapeutic potentials. Forty (40) Wistar rats divided into two (2) sets of twenty-one (21) and eighteen (18) were used. The set of twenty-one (21) rats was used to carry out the acute oral toxicity of the TPME to obtain the LD₅₀ of the extract using the Lorke's method. The set of eighteen (18) rats was divided into six (6) groups of three (3) rats each. Rats in the first group served as the normal control and received 0.5 mL of distilled water. Rats in the remaining five (5) groups received 50 mg/kg, 100 mg/kg, 200 mg/kg, 400 mg/kg and 800 mg/kg of TPME per body weight for twenty-eight (28) consecutive days respectively. Haematological indices of blood such as PCV, HCT, HGB, MCH, MCHC, MCV, WBC, LYM, NEUT, MXD and PLT were measured in the blood sample to evaluate the effect of the extract in the exposed animals. Statistical analysis using ANOVA and Tukey post hoc test was done for all parameters measured. Change in weight of the exposed animals reduced, albeit non-significant ($p > 0.05$). There was a significant increase in neutrophils count, with the highest effects produced by groups that received 400 mg/kg and 800 mg/kg of TPME. There was a non-significant increase of PCV, HCT, HGB, MCH, MCHC and MCV across the group as well as a non-significant decrease in RBC, WBC, MXD and lymphocytes counts across the groups ($p > 0.05$). In summary, the result from this study indicates that the administration of TPME may possess some haematinic effect and as such can be

investigated further for potential use as a haematinic agent.

Keywords: *Tridax procumbens*, haematinic, trypanosomiasis.

Introduction

Plant derived products have been used for medicinal purposes for centuries. The medicinal plants are rich sources of natural remedies to treat the pathogens and other diseases (Bhagat and Kondawar, 2019). Herbal treatment or folklore medicines are widely used for the treatment of many diseases in both developed and developing countries. According to WHO, almost 65% of the world's population has incorporated herbal medicines into their primary modality of healthcare (Dattaray, 2022). One of these medicinal plants is *Tridax procumbens* (Linn.), recognized for its numerous therapeutic potentials (Ingole *et al.*, 2022). *T. procumbens*, a wild herb distributed through India, Nepal, and Nigeria. It is used to treat bronchial catarrh, dysentery, diarrhoea, and inflammation (Ingole *et al.*, 2022). It commonly referred to as *Coat buttons* or *Tridax daisy* in English, *Ghamra* in Hindi is a species of flowering plant of the daisy family, native to tropical America and naturalized in tropical Africa, Asia, and Australia (Bhagat and Kondawar, 2019). Beck *et al.* (2018) said Igbo people of southeastern Nigeria call it *Mbuli*, *Harantama* in Hausa and *Igbalode* in Yoruba. *T. procumbens* was introduced in Nigeria as an ornamental plant but later spread to other tropical countries (Dattaray, 2022). *Tridax procumbens* is widely used as feed in rabbits in Nigeria (Akintunde *et al.*, 2017).

Medicinal plants have active phytochemical components responsible for the various pharmacological properties exhibited by the plants (Akintunde *et al.*, 2017). Bhagat and Kondawar (2019) revealed that several active chemical constituents were isolated and reported from the plant *Tridax procumbens*. The phytochemical screening of *Tridax procumbens* reported by Ahmed *et al.* (2019), unconcealed the presence of saponins, alkaloids, tannins, flavonoids (catechins and flavones) and carotenoids. Variety of chemical constituents viz: n-hexane, β -sitosterol, fumaric acid, luteolin, oxoester, quercetin, lauric acid, palmitic, myristic, arachidic and linoleic acid. Linolenic acid was also reported in the aerial parts. Mineral composition of *T. procumbens* reported from leaves is calcium, magnesium, potassium, sodium, and selenium. It has been observed that *T. procumbens* can serve as a good source of plant protein and potassium supplement, as well as being potential source of provitamin A (carotenoids) to the population (Bhagat and Kondawar, 2019).

Babayi *et al.* (2018) revealed that the plant is used traditionally for the treatment of bronchial catarrh, dysentery, malaria, stomach-ache, diarrhoea, high blood pressure and to check haemorrhage from cuts, bruises, and wounds and to prevent falling of hair. They stated that it also possesses antiseptic, insecticidal and hepatoprotective properties, has marked depressant actions on respiration, used as an antidote to arrow poison and the powdered leaves are applied to the wounds. Pharmacological studies have shown that *T. procumbens* possesses properties like - anti-inflammatory, hepatoprotective, wound healing, immunomodulatory, antimicrobial, antiseptic, and hypotensive, bradycardiac effects (Pareek *et al.*, 2009).

Aim of the Study

The aim of this study is to assess the haematological effect of TPME in Wistar rats.

Study Design

This study is based on an experimental study design.

Methodology

Materials

Reagents and Chemicals

All the reagents used for the study were of the highest grade and purity. Absolute methanol was purchased from Alli Shuaibu store, Sokoto. The animal feed used was obtained from Grand Cereal Soil Mills Limited, Jos, Nigeria.

Methods

Collection and Preparation of Plant

Fresh plant samples of *Tridax procumbens* were collected from and around Usmanu Danfodiyo University Teaching Hospital (UDUTH) premises, Sokoto, Sokoto state, Nigeria. The specimen was identified and authenticated at the Herbarium unit of the Pharmacognosy and Ethnopharmacy Department, Faculty of Pharmaceutical Sciences, Usmanu Danfodiyo University, Sokoto, by comparing with established Herbarium specimen with voucher number: PCG/UDUS/Aste/0007 reference number periodically which was kept at the Herbarium. The plant sample was rinsed in running tap water and air-dried under a shed at room temperature to avoid decomposition of the active components by the sunlight for 4 weeks. The dried plant was pulverized to fine powder with mortar and pestle.

Plant Extraction Procedure

The pulverized powder (200 g) was then extracted using maceration method of extraction for 72 hours with 2 L of absolute methanol in Erlenmeyer flask followed by periodic stirring to obtain the hydro-alcoholic crude extract. The collected solution was separated from the marc by using filter paper (Whatman No.1). The extract was evaporated to dryness with mild heating in water bath to dry at 40 °C to obtain the concentrated extract and stored in sterile plastic bottles at 4°C until used for further analysis.

Phytochemical Screening

The phytochemical tests were conducted employing standard chemical test procedures described by Trease and Evans (1989).

Drug Formulation

The residual filtrate of TPME obtained was reconstituted taking into consideration the average

weight of the albino rats, duration of extract administration and the required volume of doses. The crude extract was diluted with distilled water to obtain varying concentration of the extract per kg body weight (Ahmad *et al.*, 2013).

Management of Experimental Animal

Forty (40) albino rats (either sex) of wistar strain weighing 110-160 g were obtained from and kept at animal house of Faculty of Pharmaceutical Sciences, Usmanu Danfodiyo University, Sokoto for the duration of the study. The rats were housed in polypropylene cage floor area under controlled conditions at 25 ± 3 °C and kept under 12/12 light/dark cycle. The animals were fed with standard pellet feed obtained from Grand Cereal Soil Mills Limited, Jos, Nigeria, and water ad libitum throughout the period of acclimatization and administration of the test compounds.

Animal Grouping and Experimental Design

The TPME was screened for acute oral toxicity using Lorke's method. Eighteen (18) Wistar rats were divided into six (6) groups of three (3) rats each., which were treated with varying dosage of TPME for 28 consecutive days.

Group I: Normal control group feed with normal diet and 0.5 mL of distilled water.

Group II: Served as the test group that received 50 mg/kg of TPME.

Group III: Served as the test group that received 100 mg/kg of TPME.

Group IV: Served as the test group that received 200 mg/kg of TPME.

Group V: Served as the test group that received 400 mg/kg of TPME.

Group VI: Served as the test group that received 800 mg/kg of TPME.

Sample Collection and Processing

After 28 days period of TPME administration, the animals were fasted for 12 hours and was anaesthetized in a glass jar containing wool soaked with chloroform. About five millilitres (5 ml) of blood samples was collected from the animals through cardiac puncture, into labelled EDTA anticoagulant non-vacutainer containers. The sample was mixed by repeated inversion to prevent blood clot formation. The samples were immediately analysed using PEC MEDICAL

Hb7021 Hema analyzer, a three-part haematological analyzer.

The analyzer was switched on and allowed to run a self-check so as to undergo calibration. After the calibration, the sample probe was ejected automatically. The sample was homogenized and a volume of the unclotted blood sample was placed below the probe and removed immediately after the required volume had been aspirated. The indices of the full blood count were analyzed by the analyzer within 3 minutes and the result was displayed on the digital screen.

Ethical Consideration

The animals were handled humanly in conformity with the university of Usmanu Danfodiyo university ethical guideline for care and use of laboratory animals, following approval.

Statistical Analysis

All statistical analyses were conducted Statistical Package for Social Sciences (SPSS) version 27.0 and Microsoft Excel 2021. The results obtained from the control and experimental groups were expressed as mean \pm standard error of mean (S.E.M) (n=3). One way ANOVA was used to determine the differences among the various groups. Tukey *post* multiple comparisons was done for comparisons between the control group and exposure groups. Differences were considered significant at p 0.05 level of significance.

Results

Physical Properties of TPME

The extract obtained after the extraction of *T. procumbens* plant sample with methanol was a green, thick, oily, waxy paste with herbaceous odour. The methanolic extract obtained solidified at room temperature, forming a glossy cake. A yield of 18.8 g crude extract representing 9.5% was obtained upon concentration of the extract.

Qualitative Phytochemical Screening of TPME

The presence of the following phytochemicals as shown in Table 1 was confirmed in the TPME obtained after extraction.

Table 1: Phytochemicals constituents of TPME

Phytochemicals	Result
Alkaloid (Dragendroff test)	+
Steroids (Liebermann -buchard's test)	+
Saponin (Froth test)	+
Tannins (Ferric chloride test)	+
Flavonoid (Alkaline test)	+

Keys: + Positive

- Negative

Acute Oral Toxicological Evaluation of TPME

In the first phase of the acute oral toxicity study, no remarkable signs of toxicity were observed at 10 mg/kg and 100 mg/kg dosages. However, there were rubbing at the site of application, nose, and mouth on the floor of the cage and restlessness at 1000 mg/kg dosage. The signs of toxicity observed in the second phase of the

study, were like those observed in the phase one. Despite the behavioural changes exhibited by the animals in both phases, zero mortality was recorded. The result of the acute oral toxicity (LD_{50}) in Wistar rats is shown in Table 2. The result showed that no death was recorded in the rats after 24 hours and up to 14 days post oral treatment. This indicates that the LD_{50} is greater than 5000 mg/kg.

Table 2: Acute oral toxicity (LD_{50}) study of TPME in Wistar rats.

Groups	Dose (mg/kg body weight)	Observation Period	Behavioural Changes	Mortality
Control	0.5 mL Distilled water	Up to 48 hours	Absent	0/3
I	10	Up to 72 hours	Absent	0/3
II	100	Up to 72 hours	Absent	0/3
II	1000	Up to 72 hours	Absent	0/3
IV	1600	Up to 72 hours	Present	0/3
V	2900	Up to 72 hours	Present	0/3
VI	5000	Up to 72 hours	Present	0/3

Change in Body Weight

The animals were weighed pre-treatment and weekly throughout the 28 days duration. Figure 1 is a graphical representation of change in the mean body weight (g) of the rats in each group.

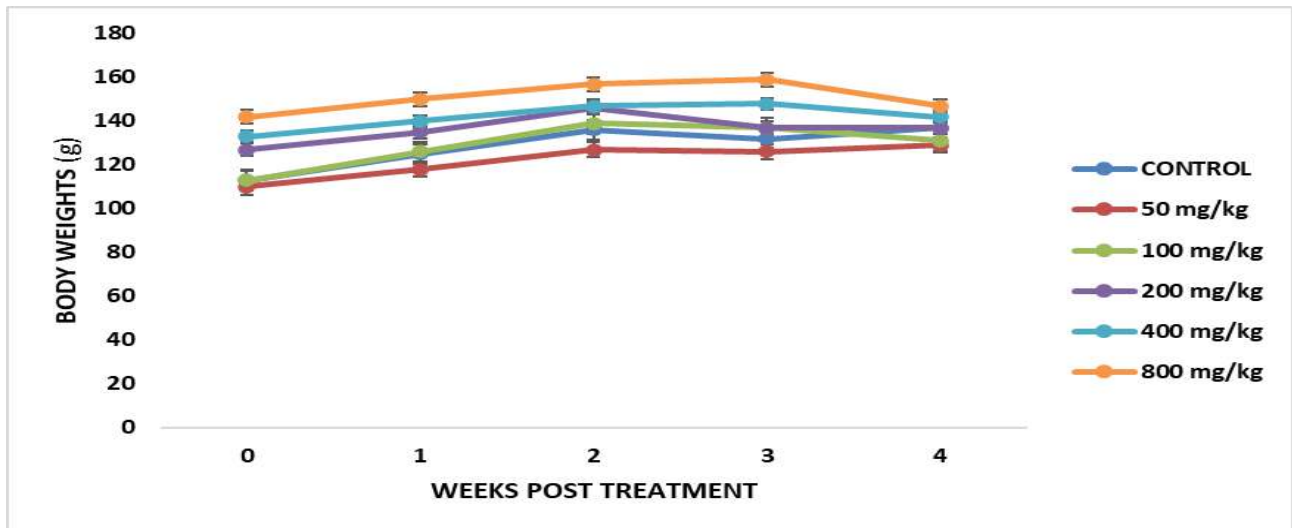


Figure 1: Body weight of rats pre- and post-exposure to TPME over the duration of the experiment.

Haematological Analysis

A nonsignificant increase in the mean of haematocrit or PCV, RBC, HGB, MCV, MCH, MCHC, RDW of the treated groups when compared to the control ($p > 0.05$) was observed. Although nonsignificant, decreased mean values of RBC, WBC and lymphocytes compared to the control was observed ($p > 0.05$). A significant increase in neutrophils count was observed with

doses 400 and 800 mg/kg producing the highest values ($p < 0.05$). There was no statistical significance between the values of PLT, MPV and PCT of the treated animals and control group ($p > 0.05$). Tables 3a, 3b and 3c show the effect of TPME on RBC indices, WBC indices and platelet indices respectively. Figure 2a, 2b and 2c are bar chart representations of RBC indices, WBC indices and platelet indices respectively.

Table 3a: RBC indices of both control and test groups treated with TPME

Parameters	Contro l	50 mg/kg	100 mg/kg	200 mg/kg	400 mg/kg	800 mg/kg	F- value	P- value
RBC ($\times 10^{12}/L$)	6.86 \pm 0.24 ^a	6.13 \pm 0.90 ^a	4.95 \pm 2.19 ^a	5.26 \pm 1.69 ^a	6.44 \pm 0.84 ^a	7.58 \pm 0.23 ^a	0.679	0.648
HGB (g/L)	117 \pm 3.46 ^a	108.00 \pm 14.01 ^a	123.00 \pm 10.21 ^a	123.33 \pm 6.00 ^a	118.00 \pm 1.00 ^a	135.67 \pm 3.76 ^a	1.380	0.299
HCT (%)	34.86 \pm 0.65 ^a	32.46 \pm 3.51 ^a	38.07 \pm 2.71 ^a	36.80 \pm 2.04 ^a	36.61 \pm 0.23 ^a	41.56 \pm 1.20 ^a	2.364	0.103
MCV (fL)	48.47 \pm 0.78 ^a	49.43 \pm 1.34 ^a	50.37 \pm 0.42 ^a	50.27 \pm 1.17 ^a	51.27 \pm 1.01 ^a	52.17 \pm 0.33 ^a	2.019	0.148
MCH (Pg)	17.07 \pm 0.88 ^a	17.74 \pm 0.53 ^a	17.63 \pm 0.37 ^a	18.34 \pm 0.26 ^a	18.19 \pm 0.24 ^a	17.92 \pm 0.91 ^a	2.900	0.061
MCHC(g/L)	351.67 \pm 3.76 ^a	358.33 \pm 1.76 ^a	349.67 \pm 3.38 ^a	364.67 \pm 10.49 ^a	351.33 \pm 4.10 ^a	343.00 \pm 0.58 ^a	2.157	0.128
RDW (%)	28.00 \pm 0.58 ^a	26.33 \pm 1.76 ^a	25.00 \pm 1.16 ^a	24.67 \pm 1.20 ^a	24.67 \pm 1.45 ^a	23.33 \pm 0.33 ^a	1.863	0.175

Keys: Values are mean of three determinations (\pm SEM), RBC: Red Blood Cell, HGB: Haemoglobin, HCT: haematocrit, MCH: Mean Corpuscular Haemoglobin, MCHC: Mean Corpuscular Haemoglobin Concentration, MCV: Mean Cell Volume, RDW: Red blood cell Weight, ^a indicates values which has show no significance, ^{b,c} indicate values which show significance.

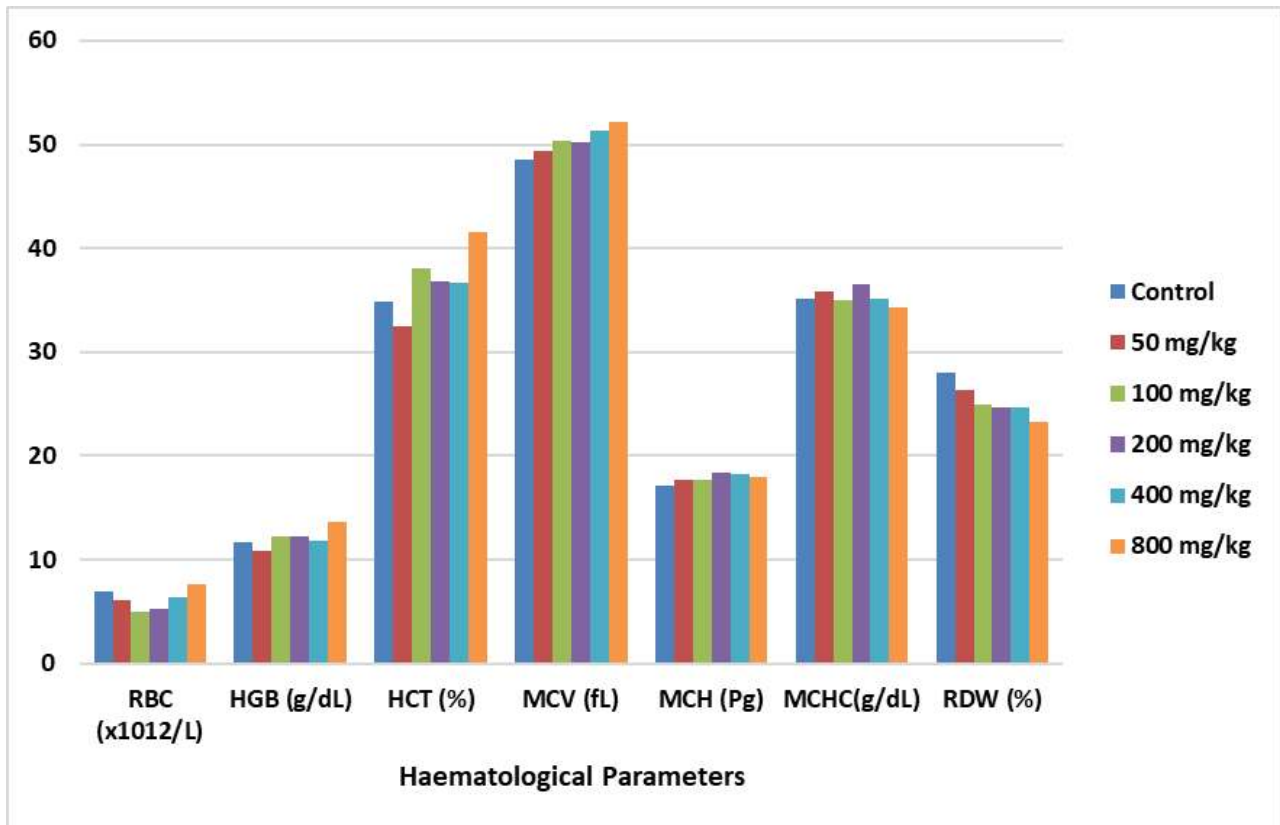


Figure 2a: RBC indices of both control and test groups treated with TPME.

Table 3b: WBC indices of both control and test groups treated with TPME

Parameters	Control	50 mg/kg	100 mg/kg	200 mg/kg	400 mg/kg	800 mg/kg	F-value	P-value
WBC (x10⁹/L)	...	4.40± 1.27 ^a	4.23± 1.65 ^a	8.77± 4.19 ^a	6.50± 2.25 ^a	8.60± 1.63 ^a	1.034	0.441
LYM (x10⁹/L)	7.41± 0.83 ^a	3.36± 0.99 ^a	3.20± 1.41 ^a	7.07± 3.69 ^a	4.42± 1.64 ^a	5.80± 1.03 ^a	0.958	0.480
NEUT (x10⁹/L)	0.58± 0.07 ^a	0.40± 0.08 ^a	0.58± 0.15 ^a	0.48± 0.12 ^a	1.14± 0.22 ^a	1.19± 0.22 ^b	4.902	0.011
MXD (x10⁹/L)	1.37± 0.04 ^a	0.64± 0.22 ^a	0.46± 0.16 ^a	1.20± 0.51 ^a	0.95± 0.39 ^a	1.61± 0.47 ^a	1.652	0.221
LYM (%)	78.63± 2.82 ^a	76.34± 1.00 ^a	69.49± 13.96 ^a	76.65± 4.77 ^a	65.19± 2.70 ^a	67.97± 2.82 ^a	0.756	0.596
NEUT (%)	6.33± 1.25 ^a	9.58± 1.18 ^a	21.75± 11.85 ^a	8.68± 3.44 ^a	21.72± 4.15 ^a	14.16± 1.20 ^a	1.559	0.244
MXD (%)	15.05± 1.57 ^a	14.08± 1.13 ^a	11.43± 0.74 ^a	14.40± 2.75 ^a	13.09± 1.45 ^a	18.03± 2.37 ^a	1.483	0.266

Keys: Values are mean of three determinations (± SEM), WBC: White Blood Cell, LYM: Lymphocyte, NEUT: Neutrophil, MXD: Mixed Differentials, ^a indicates values which has show no significance, ^{b,c} indicate values which show significance.

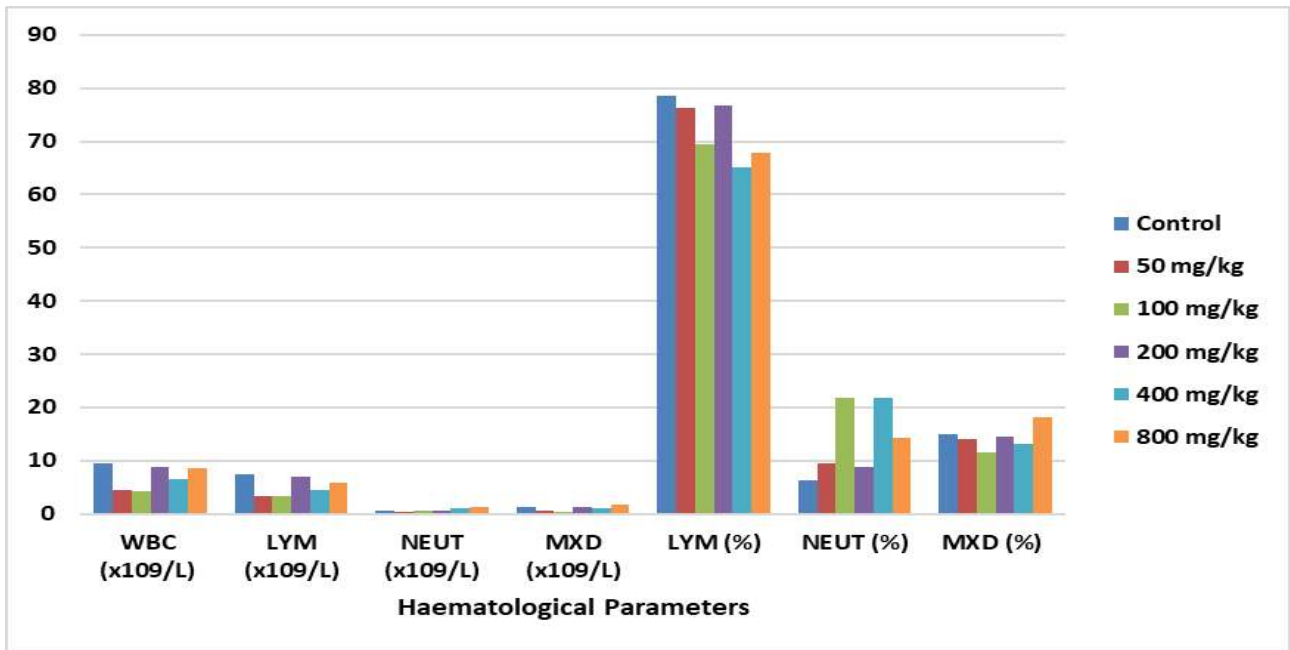


Figure 2b: WBC indices of both control and test groups treated with TPME.

Table 3c: Platelet indices of both control and test groups treated with TPME.

Parameters	Control	50 mg/kg	100 mg/kg	200 mg/kg	400 mg/kg	800 mg/kg	F-value	P-value
PLT (x10 ⁹ /L)	394.67±	263.33±	156.00±	330.67±	317.67±	344.00±	0.479	0.785
MPV (fL)	65.53 ^a	137.41 ^a	38.44 ^a	146.86 ^a	110.85 ^a	164.85 ^a	1.445	0.278
PCT (%)	3.24±	6.22±	4.44±	5.94±	6.67±	6.48±	0.981	0.706
	1.67 ^a	0.11 ^a	2.20 ^a	0.16 ^a	0.12 ^a	0.00 ^a		
	0.25±	2.21±	0.10±	0.17±	0.18±	0.22±		
	0.43 ^a	2.04 ^a	0.47 ^a	0.10 ^a	0.90 ^a	0.11 ^a		

Keys: Values are mean of three determinations (± SEM), PLT: Platelet, MPV: Mean Platelet Volum, PCT: Platorcrit, ^a indicates values which has show no significance, ^{b,c} indicate values which show significance.

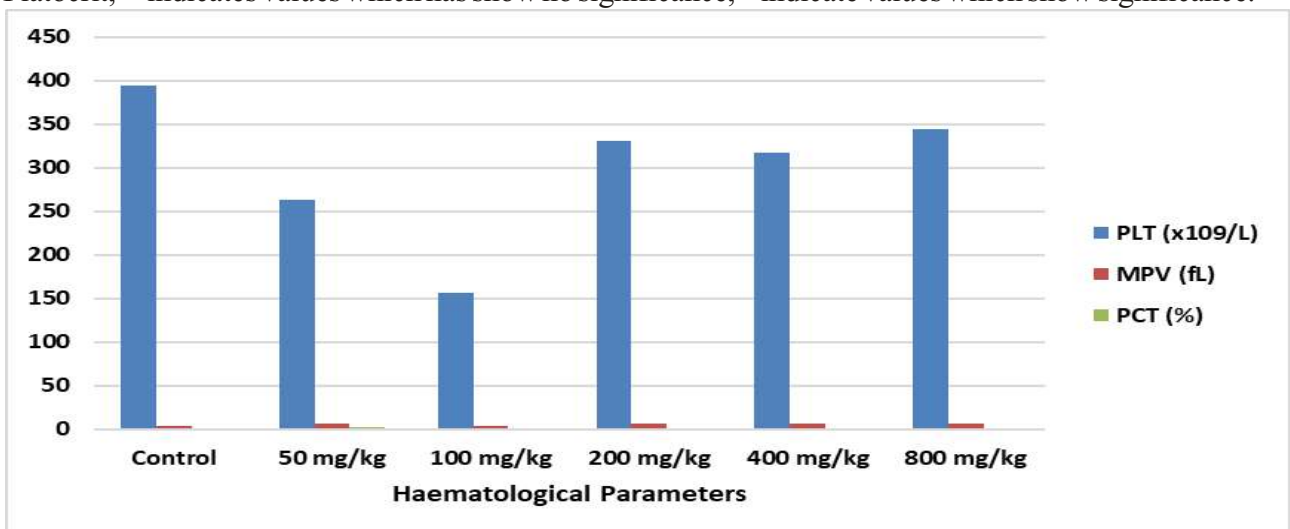


Figure 2c: Platelet indices of both control and test groups treated with TPME.

Discussion

This study was designed to assess the haematological effect of TPME in Wistar rats. Photochemical components, though naturally occurring plant chemicals providing it with colour, odour, and flavour, have been shown by research to influence the chemical processes inside the human bodies, mostly in helpful ways. Significant physiological effects, be it as antioxidants, stimulation of enzymes, hormones mimicking, interference with DNA replication, destruction of bacteria, or curbing of the onset of diseases such as cancer and heart disease, are several biological effects exerted by phytochemicals in the body (Adeluwoye *et al.*, 2017).

In this study, the preliminary phytochemical study shows that TPME is abundantly composed of a variety of phytochemical components (Table 1). This finding is as confirmed by previous studies by Dhanobalan (2008), which established the presence of alkaloids, tannin, steroid, flavonoids, amino acids, phenols, cardiac glycosides etc. in the methanol extract of *Tridax procumbens* leaves (Dattaray, 2022).

Acute toxicity is the first step of toxicological investigation of an unknown substance and its index is LD₅₀ (Abubakar *et al.*, 2012). The LD₅₀ obtained from the acute oral toxicological evaluation of TPME (Table 2) carried out was greater than 5000 mg/kg body weight, hence it is relatively safe. This claim is supported by the report of Babayi *et al.* (2018), which showed that the LD₅₀ estimate for this extract is 50 times greater than the 100 mg/kg body weight minimum effective dose in rats. This finding is also in agreement with the work of Clarke and Clarke (1967), who considered any compound or drug with oral LD₅₀ estimates greater than 1000 mg/kg body weight to be of low toxicity and safe. According to Lorkes (1983), substances more toxic than 1 mg/kg are so highly toxic that it is not so important to calculate the LD₅₀ while LD₅₀ values greater than 5000 mg/kg are of no practical interest (Abubakar *et al.*, 2012).

The high LD₅₀ estimate may be due to the oral route of administration, by which the compounds in the extract are transported through the

gastrointestinal tract, undergo metabolism to form a new product, which could be less or not toxic to the target organ.

Reports have shown that if the median lethal dose of a test substance is three times greater than minimum effective dose, the substance can be considered a good candidate for future studies (Babayi *et al.*, 2018). This suggests that oral application of TPME may not produce toxic effect at dose lower than 5000mg/kg body weight. In summary, this finding shows that TPME can be regarded as relatively non-toxic acutely. However, variables such as animal species, strain, age, gender, diet, bedding, ambient temperature, caging conditions, and time of the day were suggested to be of influence on the LD₅₀ values obtained and that such are considerable uncertainties in extrapolating the LD₅₀ obtained for species to other species (Zbinden and Roversi, 1981).

It has been recommended that body weight be measured at least once a week during toxicity studies because body weight is one of the most sensitive indicators of the condition of an animal if it is monitored frequently and carefully during the study. The body weight status of the animals can be used to evaluate the effect of the extract at all the dose levels (Abubakar *et al.*, 2012). In this study, the test animals did not show any significant variations in the body weight compared to the control group ($p > 0.05$) (Figure 1). The loss of weight may be due to disturbances in the metabolism of carbohydrates, protein, or fats. Crude extract may metabolize to a toxic end-product at higher doses, which could interfere with gastric function and decreased food conversion efficiency (Ping *et al.*, 2013). Rapid and/or marked body weight loss is usually a harbinger of ill health or death as rapid body weight loss can be due to either decreased feed or water consumption, disease, or specific toxic effect (Abubakar *et al.*, 2012).

Blood cells are the mobile units of the body protecting system (Sampathkumar *et al.*, 2018). In this study, a non-significant decrease of RBC count across the group was observed ($p > 0.05$) (Table 3a, Figure 2a). Only the group that received 800 mg/kg of the methanolic extract produced an

increased RBC count when compared to that of the control. This shows that the extract has little or no effect on the RBC count.

The measurement of PCV and RBC count can be used as a simple screening test for anaemia. Anaemia is an absolute decrease in the total number of RBC per millilitre (RBC/mL) of blood and/or decrease in PCV due to fewer RBC (Abubakar *et al.*, 2012). Apart from the group treated with lower tested dosage, where there was a decrease in value, although not significant ($p>0.05$), PCV increased at all other dose level (Table 3a, Figure 2a). This can be indicative of a boost in total blood volume. This supports the findings of Enwa *et al.* (2020) on the haematological effect of *Tridax procumbens* methanolic extract in Wistar rats. This result also corroborated the findings of Ikewuchi *et al.* (2013) on the effect of aqueous leaf extract of *Tridax Procumbens* Linn (Asteraceae) in subchronic salt-loaded rats and the findings of Sampathkumar *et al.* (2018) on the impact of *Tridax procumbens* on haematological parameters in aflatoxin induced liver toxicity in albino rats. This shows that the extract may have a positive effect on the haematopoietic system of the rats. Raised haematocrit indicates haemoconcentration, often due to increased red cell mass (Ikewuchi *et al.*, 2013). This indicated the protecting effects of the plant and its extract on the haematopoietic system (Sampathkumar *et al.*, 2018).

Apart from the group treated with 50 mg/kg of the extract, where there was a decrease in value, although not significant ($p>0.05$), the haemoglobin level of the animals increased at all other dose levels (Table 3a, Figure 2a). The raised haemoglobin is also an indication of the boosting effect of the extract. This is also reflected by the haematocrit level and negates the finding of Bhagwat *et al.* (2008), of a slight, but not significant lowered haemoglobin values in animals treated with the plant. However, this finding supports the work of (Abubakar *et al.*, 2012). This means that administration of *Tridax procumbens* methanolic extract may be beneficial in anaemia related disorder particularly trypanosomiasis.

An increase in MCH, MCHC and MCV values was observed (Table 3a, Figure 2a). The MCH, MCHC

and MCV values depend on an increase or decrease in the synthesis of haemoglobin and so these values increase due to presence of increased amounts of haemoglobin being synthesized. Therefore, this helps to determine the presence or the occurrence of anaemia in an individual and can be used to prevent haemorrhage (Enwa *et al.*, 2020). A non-significant reduction in platelet count was also observed (Table 3c, Figure 2c) ($p>0.05$). Platelet aggregation together with fibrin stabilizes the platelet plug formation which stops bleeding from minor injuries (Gubbiveeranna *et al.*, 2019).

There was non-significant general reduction in the in total white blood cell (leucocytes) and lymphocyte counts ($p>0.05$), with extreme reduction produced at doses 50 mg/kg and 100 mg/kg and moderate reduction at dose 400 mg/kg (Table 3b, Figure 2b). This finding is in contrary to the work of Abubakar *et al.* (2012). However, Enwa *et al.* (2020) reported a reduction in leucocytes of the test group when compared to the control. Poisoning from chemicals, drugs, stress, etc. are among the main causes of leukocytosis. Takeda *et al.* (2003) posited that an elevated white blood cell count in peripheral blood is a known risk factor of coronary artery disease. Thus, the observation of WBC counts lowering effect of the extract in the test animals, can be an indication of reduced risk of coronary artery disease. Hence, the methanolic extract of *Tridax procumbens* may be effective in the management of some cardiac related conditions.

The statistical analysis of the hematological data revealed only neutrophils as the parameter with statistical significance (Table 3b, Figure 2b). A significant increase in neutrophils was observed in the test groups with the highest increase produced in the group that received 400 mg/kg and 800 mg/kg ($p<0.05$). The finding of this study is in line with the work of Abubakar *et al.* (2012). However, neutrophils count at dose 50 mg/kg and 200 mg/kg were low when compared to that of the control. Neutrophils is most frequently caused by systemic or severe local bacterial infection where their primary role is in immunity against bacterial and fungal infection by phagocytosis. Neutrophils, also known as granulocytes or segmented neutrophils, are the main defence of the body against infections and

antigens. High levels may indicate an active infection; a low count may indicate a compromised immune system or depressed bone marrow (low neutrophils production). Therefore, administration of this extract may be of help in the presence of a clinical antagonistic like trypanosomes (Abubakar *et al.*, 2012).

Conclusion

The LD₅₀ estimate for TPME obtained from this study was above 5000 mg/kg. There was reduction in body weight of the animals treated with the extract compared to the control animals. This study revealed that TPME has the capability of increasing the levels of MCH, MCHC, MCV and neutrophils count. It also showed its ability to boost total blood volume by increasing PCV. This indicates that the extract may have haematinic effect and can potentially be used as a haematinic agent in patients having low blood volume. Its effect on neutrophil count shows that it can be used to boost immunity. In conclusion, TPME seems a good drug, which is relatively safe. However, this study can be regarded as a preliminary project, necessitating further studies on the extract.

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Citation: Ngaski, A.A., Abubakar, M. and Bunza, F.U. Haematological Effect of *Tridax Procumbens* Methanolic Extract in Wistar Rats. *Sokoto Journal of Medical Laboratory Science*; 9(3): 173 – 183. <https://dx.doi.org/10.4314/sokjmls.v9i3.18>

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