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ANTI-ANAEMIC AND HEPATO-RENAL ACTIVITIES OF ETHANOL LEAF EXTRACT OF *Alchornea cordifolia* IN PHENYL HYDRAZINE INDUCED-ANAEMIC WISTAR RATS

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ABSTRACT

The effect of ethanol leaf extract of Alchorne acordifolia on some biochemical parameters in phenyl hydrazine-induced anaemic Wistar rats was studied. A total of thirty-six (36) Wistar rats weighing (95-200) g were selected for this study and randomly divided into six groups of six animals per group. Group A (normal control), group B (negative or anaemic) control, group C (standard), group D, E and F (treated groups). Animals in groups B, C, D, E and F were induced with anaemia via intraperitoneal (I.P.) injection of 10mg/kg body weight phenyl hydrazine (PHZ) for 3 days and group A received distilled water in place of the PHZ for the same duration. After the induction, group A and B rats received oral administration 0.9w/v normal saline solution while groups C. D. E and F received oral administration of enzoron (10mg/kg.bwt), 100, 200 and 400 mg/kg.bwt of extract respectively for 14 days. The percentage yield of the extract was determined to be 18%. Phytochemical screening revealed the presence of saponins, carbohydrates, protein, alkaloids, phytosterols, phynols, flavonoids and glycosides at varying concentrations. A dose of 5000mg/kg. bwt was found to be safe in the LD₅₀ studyof the extract The oral administration of the extract showed a significantly (p<0.05) level of total protein (TP) ,packed cell volume (PCV), haemoglobin (Hb) and red blood cells (RBCs) of the animals in treated groups compared to those of the animals in anaemic groups. The aspartate transaminase (AST), alanine transaminase (ALT) and total bilirubin (TB) were significantly (p<0.05) lowered in the extract treated groups than in the anaemic non-treated groups. There was a no-significant decrease (p>0.05) in the serum urea, creatinine, Na⁺, K⁺ and CI of animals in the extract treated groups when compared to both the normal and standard control. The histology of the spleen revealed the regeneration of damaged cells in extract treated groups unlike that of the anaemic non-treated groups which showed distorted architecture. The study suggests that treatment with Achorne acordifolia leaf extract in phenyl hydrazine induced anaemia enhances anti- anaemic and hepatoprotective effect possibly due to both its anti-inflammatory and antioxidant properties.

KEYWORDS: Anti-anaemic, Anti-diabetic, Anti-inflammatory, Hepato-protective, Phytochemical

INTRODUCTION

Anaemia is a condition that develops when blood lacks enough healthy red blood cells or haemoglobin. Anaemia affects the lives of more than 2 billion people globally, accounting for over 30% of the world's population particularly in developing countries occurring at all stages of the life cycle (Ayensu *et al.*, 2020).It is characterized by a decrease in haemoglobin concentration (Hb), red blood cells (RBCs) count and packed cell volume (PCV).

It causes hypoxia due to failure to meet tissue oxygen demand (Bruner etal., 1996). The anaemia prevalence remains high in Africa, with an overall incidence of 64.6% in children, 55.8% among pregnant women and 44.4% among young girls (Diallo et al., 2008). Anaemia can be categorised into either morphological or causal categories (Bruner et al.,1996). The morphological category depends on the size of haemocytes and Hb concentration, whereas the causal category consist of hypochromic anaemia, haemolytic anaemia, aplastic anaemia, and nutritional deficiency of red blood cells

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(iron, vitamin B₁₂ and folic acid) (Seo,1996).The exposure to many chemicals has been associated with red blood cells destruction and haemolytic anaemia (Beutler., 2001). The haemolytic activity of any aryl hydrazines such as phenyl hydrazine, hydroxylamine and divicine may lead to acute haemolytic anaemia in vertebrates (Prasuna et al., 2015). Presently, more than half of the world experience some form of anaemia in their life time (Duff, 2008). Several studies have shown that haemolytic anaemia is associated with oxidative stress within the erythrocytes . This concept is supported by the fact that haemolytic damage is accompanied by the generation of reactive oxygen species, glutathione depletion, haemoglobin oxidation and Heinz body formation in red blood cells (Prasuna et al., 2015). Haemolytic agents have also been reported to cause membrane lipid peroxidation and denaturation of cytoskeletal proteins (Prasuna et al., 2015).

Medicinal plants have long played important role(s) in the management of diseases all over the world (Dasofunjo et al., 2018). Medicinal plants naturally synthesize bioactive metabolites like alkaloids, terpenes, saponins, sterols. flavonoids and phytosterols (Dasofunjo et al., 2018). Developing therapeutic agents from natural products has renewed the worldwide attention and stimulates new wave of research on the benefits of the herbal medicine as an effective alternative therapeutic tool for various illnesses (Li et al., 2020). The increasing discovery of more medicinal plants demanded for increased scientific scrutiny of their bioactivity so as to provide data that will help physicians and patients make wise decision before using them. A typical example of this medicinal plant is Alchorne acordifolia.

Alchornea cordifolia is a plant that belongs to the family of Euphorbiaceae. It is locally known as "ububo" (Igbo), "ipaesinyin" (Yoruba), "banbani" (Hausa), "upia" (Igede), "uwonwen" (Benin), "mbom" (Efik) and "ukpaoromi" (Ijaw) among others in Nigeria (Osei et al., 2019).

The leaves and stems are mostly used therapeutically in African countries as remedies for veneral diseases, treatment of inflammatory disorders, cancer, and ulcers (Osei et al., 2019). The stem bark is tinctured with local gin for its aphrodisiac effects ,remedy for cold, rheumatism, arthritis and muscle pains (Adounkpe et al., 2022). Similarly, it is used as an antidote for poison, respiratory problems such as sore throat, cough and bronchitis; genital-urinary conditions such as venereal diseases, menstrual problems, impotency and female sterility; intestinal problems such as gastric ulcers, diarrhoea, amoebic dysentery and worms (Adounkpe et al., 2022).

Anaemia is lives threatening global disorder affecting millions of people either as haemolytic or iron deficiency anaemia due to toxicants or oxidants (Duff, 2008). Although, there are many drugs used in the management of anaemia but they are not affordable to many poor people especially in developing and under

incidence of anaemia. Hence, the need for this present research works. Therefore, this work was designed todetermine the effect of ethanol leaf extract of *Alchorne acordifolia* on some biochemical parameters in phenyl hydrazine-induced anaemia in Wistar rats.

MATERIALS AND METHODS

Materials

Sample collection and identification

The leaves of *Alchorne acordifoilia* were harvested from Mkar in Gboko Local Government Area of Benue State. The leaves were air dried at room temperature (25±4°c). The dry samples were macerated into coarse powdered, weighed and stored in air- tight containers.

Chemicals and reagents:

All chemicals and reagents (Phenyl hydrazine, ethanol, sulphuric acid, ferric chloride, chloroform, hydrochloric acid, Mayer's reagent and Wagner's reagent) used were of analytical grade. Fresh distilled water was used throughout the experimental period. Assay kits used in the analysis in this study were products of Randox Laboratories (England).

Experimental animals:

Thirty- six (36) Wistar rats weighing between 95-200 g were used for this study. The animals were housed in metal cages and acclimatized for two weeks in the animal house of Department of Biochemistry University of Mkar, Mkar. They were maintained under standard conditions (room temperature $25 \pm 3^{\circ}$ C, humidity 60%, light and dark period 12/12 hours). All animals had regular supply of clean drinking water and food. The animals were handled in line with ethical guidelines as approved by the Faculty of Sciences Ethical Committee, University of Mkar, Mkar

METHODS

Preparation of extract

The coarse powder (400 g) was dissolved in 1200 mls absolute ethanol in a tightly closed container for 72 hours. The mixture was shaken on daily basis until it was filtered using sterile cotton wool and later with Whatman No. 1 filter paper. The residue was pressed with a spatula to ensure complete filtration. The ethanolic filtrates were transferred into separate clean beaker and concentrated by evaporation to dryness in a rotary evaporator under vacuum at 40°c for 24 hours. The concentrates were then collected into pre-weighed small glass bottles and stored in the refrigerator prior to use and after daily administration to the experimental animals.

Determination of extract yield

The percentage yield of the ethanol extract of *A.cordifolia* leaf obtained was determined by weighing the dried leaf powder and the concentrated extract and calculated by the formula shown below

Weight of concentrated extract

Weight of concentrated extract
Weight of dried leaf powder

Phytochemical screening:

Phytochemical analysis was carried out on the extract using a standard procedure for identification of phytochemical constituents as described by Harborne, (1973); Trease and Evans (1989) and Sofowora (2008).

LD

The acute toxicity was carried out by the method described by (Lorke, 1983). The mice were divided into five groups of four mice each and were treated with the plant extracts dose of 500, 1000, 2000, 3000 and 5000 mg/kg body weights orally and observed for 24hours for signs of toxicity including death.

Induction of haemolytic anaemia

Haemolytic anaemia was induced by intraperitoneal (I.P.) injection of phenyl hydrazine (PHZ) at 10 mg/kg body weight for 3 days. Anaemia was considered to be induced by comparing the PCV of the PHZ-induced animals with that of the normal control (non-induced) animals after 24hours of the last induction. The rat's with a PCV lower than 50% were considered anaemic and suitable for the study. The PCV was carried out by the capillary tube method whose procedures are as follows; blood was collected from the tail into the capillary tube and one end of the tube was sealed with plastasine. The capillary tube was placed inside a haematocrite centrifuge and spun for five minutes at 2000 rpm.

Experimental design

The experimental rats were randomly divided into six (6) groups of six animals per group and treated for a period of fourteen (14) days.

Group A: Normal control (non-anaemic control)

Group B: Anaemic rats without treatment (negative control)

Group C: Anaemic rats treated with 10mg/kg body weight of enzoron orally (standard).

Group D: Anaemic rats treated with 100mg/kg body weight of ethanol extract of *A.cordifolia* leaf orally.

Group E: Anaemic rats treated with 200mg/kg body weight of ethanol extract of *A.cordifolia* leaf orally.

Group F: Anaemic rats treated with 400mg/kg body weight of ethanol leaf extract of *A.cordifolia* orally.

All administrations were done orally using oropharyngeal cannula once per day for 14days (2 weeks).

Blood sampling and analysis

At the end of the 14 days, the rats were sacrificed under diethyl-ether anaesthesia. Blood samples were immediately withdrawn by cardiac puncture from each rat. The first tube contain sodium salt of ethylene diamine tetra acetic acid (EDTA) anticoagulant and used for the assessment of haematological indices, while the second blood sample was collected into a plain centrifuge tube without any anticoagulant and after centrifugation process, the serum obtained was used for measurement of biochemical parameters.

Haematological analysis

An auto heamatologicalanalyzer (XE-2100 by Sysmex Corporation) was used to determine the heamoglobin concentration (Hb), red blood cell count (RBCs), total white blood cell count (WBC), lymphocytes, and packed cell volume (PCV).

Biochemical analysis:

Aspartate amino transferase, Alanine amino transferase and alkaline phosphatase activity was assayed as described by Reitman and Frankel (1957) using assay kits (Agape Diagnostics, Switzerland). Serum bilirubin was determined colorimetrically according to the method described by Jendrassic and Grof (1938) using assay kits (Agape Diagnostics, Switzerland). Serum proteins was assayed as described by Lowry et al. (1951). The method of Natelson et al. (1951) was used for the determination of serum urea and uric acid. Serum creatinine was determined as described by Brod and Sirota (1984). Serum creatinine concentration and serum urea concentration were respectively estimated in addition to serum electrolytes (sodium, potassium and chloride) based on the methods of Kaplan (1965) and Tietz et al. (1994) as modified by Stephen et al. (2007) using Randox assay kit.

Histological examination

Each animal spleen were dissected out and washed on ice cold saline immediately. A portion of each spleen was fixed in 10% neutral formal saline fixation solution for histological studies. After fixation, tissues were embedded in paraffin; solid sections were cut at 5mmX, 2mmX and 1mmX. Various sections were stained with haematoxylin and eosins .The slides were viewed at magnification of X 400 and photomicrographs taken.

Statistical analysis

Data obtained was expressed as mean \pm SD and statistically analyzed using one-way analysis of variance (SPSS) with Turkey's multiple comparison post hoc tests to compare the level of significance between the test groups. The values of p<0.05 were considered as significant.

RESULTS

The extract percentage yield was found to be 18.00%, it was dried and dark green in colour. Preliminary phytochemical screening of Alchorne acordifolia leaf extract from ethanol carried out reported the presence of various phytochemicals such as alkaloids, saponins, flavonoids, glycosides, phynols, carbohydrate, proteins, phytosterols, reducing sugar and tannins (Table 1). The LD₅₀ result of the ethanol extract of Alchorne acordifolia leaf on adult mice showed that the extract was safe at a dose of 5000mg/kg body weight since there was no record of death during the study (Table 2). The effect of ethanol leaf extract of Alchorne acordifolia on some biochemical parameters of PHZ induced anaemic rats reveals that ALT, ALP and AST recorded a significant decrease (p<0.05) in the anaemic control and anaemic treated groups when compared with the normal control group following the administration of the extract. Also,

the extract produced a significant increase (p<0.05) in serum total protein in the anaemic control group when compared with the normal control group .The total protein level also decreased significantly (p<0.05) in all the anaemic untreated groups compared with normal and standard control following the administration of the extract. The total bilirubin level was significantly (p<0.05) decreased in the anaemic groups treated with standard drug, 200mg/kg and 400mg/kg of the extract compared with untreated group (Table 3).

In table 4, urea concentration was significantly increased (p<0.05) in anaemic control group (6.56±1.03) compared with normal control (3.23±0.52). There was no-significant increase (p>0.05) in all the treated groups (4.29±0.52, 4.59±0.56, 5.54±1.33, 5.67±0.67) compared with normal control (3.23±0.52). It was also notsignificantly different (p>0.05) in the group treated with standard drug (4.29±0.52) compared with anaemic control (untreated) group (6.56±1.03). There was a significant increase (p<0.05) in the concentration of creatinine in the anaemic control group (73.31±8.19), anaemic group treated with 400mg/kg (54.55±4.97) compared with the normal control group (39.83±8.02). Creatinine concentration was significantly decreased (p<0.05) following the administration of the extract in the anaemic groups treated with standard drugs, 100mg/kg and 400mg/kg of extract (49.82±4.89, 51.61±7.25, 54.55±4.97) compared with untreated (73.31±8.19). There was a significant decrease (p<0.05)

with 200mg/kg of extract (155.48±.65) compared with untreated group (180.97±7.36) and a significant (p<0.05) increase in (155.48±.65) compared normal control group (174.29±9.23).

Table 5 revealed theeffect of ethanol leaf extract of Alchorne acordifolia on haematological profile in phenyl hydrazine induced anaemic rats. The result of some haematological parameters of anaemic control group, normal control group and anaemic group administered with 100mg/kg, 200mg/kg, and 400mg/kg body weight of EE of Alchorne acordifolia leaves. In this, there was a significant increase (p<0.05) in the values of PCV, RBC, Hb in all the anaemic treated groups compared with the untreated compared with normal control group .There was no significant increase (p>0.05) in WBC value of groups treated with standard drug, 100mg/kg and 200 mg/kg. (3.82±0.92, 3.72±0.44.3.66±0.91) and a no significant (p>0.05) decrease in group treated with 400mg/kg (3.42±0.52) compared with normal control (3.44±0.90). There was a significant (p<0.05) increase in lymphocytes value of untreated group (66.80±2.78), groups treated with 100, 200 and 400mg/kg of the extract (59.00±9.82^a, 58.80±10.23^a, 55.60±11.28) compared with normal group (40.80±17.28). There was also a significant (p<0.05) decrease in anaemic group treated with standard drug (49.80±11.21) compared with untreated group (66.80±2.78) and there was a no significant (p<0.05) increase in anaemic group treated with standard drug (49.80±11.21) compared with normal control (40.80±17.28).

Table 1: Preliminary analysis of phytochemicals in ethanol extract of Alchorne acordifoilai

Phytochemmicals	Bioavailability
Alkaloids	++
Carbohydrate	++
Flavonoids	++
Glycosides	++
Phytosterols	++
Phynols	++
Proteins	+++
Reducing sugar	++
Saponins	+++
Tannins	++

Key:- = Absent, += Present in small quantity, ++moderately present, +++abundantly present

Table 2: LD₅₀ Lethal dose (LD₅₀) of ethanol extract of Alchorne acordifolia leaf

Groups	Number of mice	Doses (mg/kg weight)	body Number recorded	of	death
Α	4	500	Nil		
В	4	1000	Nil		
С	4	2000	Nil		
D	4	3000	Nil		
E	4	5000	Nil		

Table 3:Effect of ethanol leaf extract of *Alchorne acordifolia* on liver function indices in phenyl hydrazine induced anaemic rats.

Parameters	Normal (+ve) controls	PHZ group	PHZ+ Standard	PHZ+ 100mg/kg bwt extract	PHZ+ 200mg/kg bwt extract	PHZ+ 400mg/kg bwt extract
AST (IU/L)	44.96±4.19 ^b	74.85±5.11 ^a	46.83±5.67 ^{ab}	29.92±3.77 ^{ab}	24. ±5.3	22.05±3.84 ^{ab}
ALT (IU/L)	28.40±7.16 ^b	38.40±2.70 ^a	27.40±3.36 ^b	25.20±3.12	25.85±5.	24.60±6.23
TB (IU/L)	2.85±0.30 ^b	5.54±0.75 ^a	3.15±0.67 ^b	3.53±0.67	3.11±0.94 ^b	3.53±0.4 ^b
TP (g/dl)	17.83±2.16 ^b	16.52±6.45 ^a	18.36±0.99 ^b	38.85±4.38 ^b	38.14±0.92 ^b	39.52±0.76 ^b

The values are means ± SD and n=5 for each group ^ap<0.05 versus Normal (+ve) control group and ^bp<0.05 versus PHZ untreated group. AST: Aspartate transaminase, ALT: Alanine transaminase, TB: Total bilirubin and TP: Total protein.

Table 4: Effect of ethanol leaf extract of *Alchorne acordifolia* on some renal function indices in phenyl hydrazine induced anaemic rats.

Parameters	Normal (+ve)	PHZ group	PHZ+	PHZ+	PHZ+	PHZ+
	controls		Standard	100mg/kg extract	200mg/kg extract	400mg/kg extract
Urea(mmol/L)	3.23±0.52	6.56±1.03 ^a	4.29±0.52	4.59±0.56	5.54±1.33°	5.67±0.67 ^a
Cr(mmol/L)	39.83±8.02	73.31±8.19 ^a	49.82±4.89 ^b	51.61±7.25 ^b	52.69±7.71	54.55±4.97 ^{ab}
Na (mmol/L)	174.29±9.23	180.97±7.36	173.59±7.79	175.95±6.65	155.48±.65 ^{ab}	166.46±11.75
K ⁺⁽ mmol/L)	2.68±0.79	3.50±0.58	3.65±0.71	2.61±0.68	3.02±0.47	3.27±1.05
Cl (mmol/L)	63.99±8.73	75.36±10.56	74.72±5.22	66.67±11.10	69.61±7.78	64.48±10.02

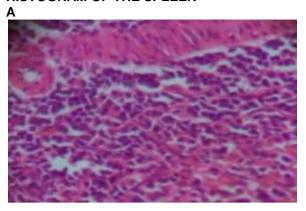
Values are presented as mean ± SD, n=5, ^ap<0.05 significant different compared to normal control, ^bp<0.05 significant different compared to untreated group,

Table 5:Effect of ethanol leaf extract of *Alchorne acordifolia* on haematological profile in phenyl hydrazine induced anaemic rats.

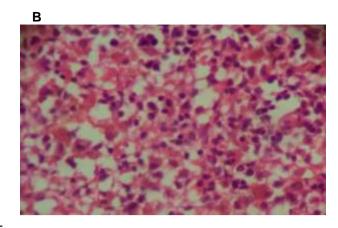
Parameters	Normal (+ve) controls	PHZ group	PHZ+ Standard	PHZ+ 100mg/kg extract	PHZ+ 200mg/kg extract	PHZ+ 400mg/kg extract
PCV (%)	46.20±9.91	34.40±3.85	51.60±5.86 ^b	53.20±8.04 ^b	54.40±5.94 ^b	56.40±8.02 ^b
WBC(10 ⁹ cells/L)	3.44±0.90	4.58±1.65	3.82±0.92	3.72±0.44	3.66±0.91	3.42±0.52
RBC(10 ¹² cells/L)	2.30±0.61	3.07±0.71 ^a	3.90±0.55	4.00±1.02	4.20±0.75	4.28±1.00
Hb (g/dl)	10.12±0.65	8.94±1.17	13.68±2.06 ^b	13.08±2.33 ^b	13.68±2.21 ^b	14.00±2.48 ^b
Lymphocytes(%)	40.80±17.28	66.80±2.78 ^a	49.80±11.21 ^b	59.00±9.82 ^a	58.80±10.23 ^a	55.60±11.28 ^a

Values are presented as mean ± SD, n=5, a=p<0.05 significant difference compared to normal group, b=p<0.05 significant difference compared to untreated group

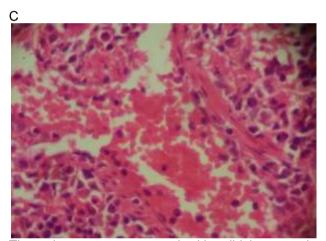
HISTOGRAM OF THE SPLEEN



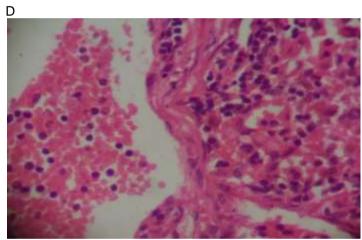
The spleen architecture appears normal. H and E $\rm X400$



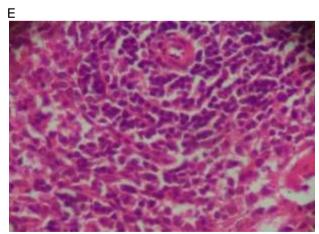
There is generalised atrophy as seen in the increase in interstitial paces and tissue cellular degeneration evident by the presence of naked nuclei. H and E ${\bf B}$: X400



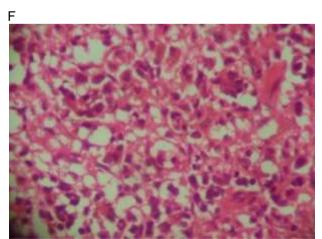
The spleen appears normal with mild increase in connective tissue fibre H and E X400



Spleen showing mild perivascular inflammation and increase in connective tissue H and E X400



The spleen appears normal. H and E X40



There is genralised atrophy evident by the increase in interstitial spaces H and E X400

DISCUSSION

Preliminary phytochemical screening of ethanol leaf extract Alchornia cordifolia revealed the presence of flavonoids, tannins, reducing sugars, phenols, protein, saponins, alkaloids, carbohydrate, glycosides, and phytosterols. It has been reported that phenyl hydrazine causes oxidative damage to red cells by increasing the formation of reactive oxygen species (Ebenyi and Uraku, 2017). These phytochemicals such s flavonoids protect cells as powerful antioxidants which prevent or repair damage done to red blood cells by free radicals or highly reactive oxygen species. Adewoye et al. (2012) stated that some of the biological functions of flavonoids include protection against allergies, free radicals, microorganisms, platelet aggregation hepatotoxins and tumours.

Alkaloids are said to be pharmacologically active and their actions are felt in the autonomic nervous system. blood vessels, respiratory system and gastrointestinal tract. In addition, alkaloids are anti plasmodic, antianaemic, analgesic and also have bactericidal effects (Dasofunjo et al., 2018). Tannins are well known for their anti-oxidant and anti-microbial properties as well as for soothing relief, skin regeneration, as anti-inflammatory and diuretic properties. Saponins lower cholesterol level, have anti-diabetic and anti-carcinogenic properties. In addition, saponins are expectorants and suppressant (Dasofunjo et al., 2013). The presence of these phytochemicals might have contributed to the haematinic and hepato-protective activity of Alchornea cordifolia observed in this present study.

In haemolytic anaemia, there is a destruction of circulating red blood cells before the normal life span (120 days), thereby leading to a decreased level of circulating haemoglobin, less than 13 g/dl in male and 12 g/dl in females (Okochi et al., 2004). PHZ decreases haemoglobin level, red blood cell concentration, and packed cell volume, and impairs ervthrocvte deformability. It induces extra medullar haematopoiesis in the spleen and liver (Berger, 2007). The determination haematological indices provides physiological information on a proper blood assessment. According to al., (2004), accurate Okonkwo et laboratory determination of blood parameters remains the only sensitive and reliable foundation for ethical and rational research, diagnosis, treatment and prevention of haemolytic anaemia. Researchers have shown that ingestion of medicinal compounds or drugs can alter the normal range of haematological parameters (Asuk et al., 2015). The major concern of the scientific communities with regard to medicinal plants and haematological studies focuses on the measures that can maintain a normal haematological state of being and reverse any negative haematological status associated with various anaemic conditions .This study revealed that the increased haemoglobin, red cell count and packed cell volume which suggests its anti anaemic and or haematopoietic effect. These results are also consistent with the findings of some researchers on the effects of the plant extracts on red cell indices of experimental animals (Dasofunjo et al., 2020).

Hepatic cells contain higher concentrations of AST and ALT in the cytoplasm and AST in particular exists in the

mitochondria. Damage or assault to hepatic cells induces leakage of plasma leading to an increased level of hepato-specific enzymes in serum (Asuk et al., 2018). The measurement of serum AST and ALT levels serve as means for indirect assessment of liver function. Alchornia cordifolia leaf extract reduced the serum levels of AST and ALT also preserved the functional ability of the liver, since it did not trigger any assault or alter the functional integrity of the liver in phenyl hydrazine induced haemolytic anaemia. A similar observation was made by (Okwari et al., 2018). The mechanism of the hepatoprotective activity of Alchornea cordifolia leaf extract in PHZ induced anaemic Wistar rats may be derived from some anti-inflammatory and antioxidant properties.

Urea and creatinine are non electrolytes found in the body. Urea is produced from metabolism of amino acid and creatinine is formed from the metabolism of muscle creatinine and creatine phosphate. They are both excreted by the kidneys (Guyton and Hall, 2006). The excretion of urea and creatinine is used to ascertain renal function. This process is actively carried out by the kidneys. Creatinine is usually produced at a fairly constant rate by the body and filtered out of the blood by the kidneys. If the filtering capacity of the kidney is deficient, creatinine blood levels rise (Guyton and Hall, 2001). In general, increased urea levels are associated with nephritis, renal ischemia and urinary tract obstruction (Oluwole et al., 2012). Urea is the major end product of protein catabolism in mammals and is the primary vehicle for removal of toxic ammonia from the body. It is primarily produced in the liver and secreted by the kidneys (Oluwole et al., 2012). From this present work, the damage caused by PHZ administration was reversed in groups treated with standard drugs, 200mg/kg and 400mg/kg BW while the spleen of the negative group showed inflammation and damage. It appears that the presence of these antioxidants in the plant extract was not so much to reverse the damaging affect of phenylhydrazine as seen in the spleen of groups administered with 100mg/kg of the extract.

CONCLUSION

The result of this study suggest that the ethanol leaf extract of *Alchorne acordifolia* has anti –anaemic and hepato-protective properties in phenyl hydrazine induced anaemia.

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