Evaluation of Anti-anaemic potential of aqueous extract of *Alchornea laxiflora* (Benth) leaf in iron deficient Rats

*****Oladiji, A.T., Olatunde, A. and Oloyede, H.O.B.

Department of Biochemistry, Faculty of Life Sciences, University of Ilorin, P. M. B. 1515, Ilorin, Nigeria
E-mail: temidayooladiji@yahoo.com; Telephone: +234 8033736996

Abstract: The effects of administration of aqueous extract of *Alchornea laxiflora* leaf at doses of 100, 200 and 300 mg/kg body weight on haematological indices (PCV, Hb, RBC, MCV, MCH and MCHC) of iron deficient rats were investigated. Thirty six albino rats (51.17±1.02g) were used for the study. Eight rats were fed on iron-sufficient diets while the remaining twenty eight were made iron deficient by maintaining them on iron-deficient diets. After five weeks of feeding, haematological indices of the iron deficient rats were significantly ($p<0.05$) reduced compared with rats fed on iron-sufficient diets (Control). The iron deficient rats were then treated with the extract, reference iron drug ($FeSO_4$) and iron-sufficient diets for two weeks. Proximate analysis of the iron-sufficient and iron-deficient diets showed that they were similar except in the amount of iron while proximate analysis of *Alchornea laxiflora* leaf showed that the leaf contained 3.22% moisture, 11.73% crude fat, 26.06% crude fibre, 7.26% crude protein, 9.13% ash and 42.60 carbohydrate. Phytochemical screening of the extract revealed the presence of alkaloids, tannins, flavonoids, anthraquinones, phenolics, cardiac glycosides, steroids, phlobatannins and saponins. Compared with the control (Iron sufficient group), the extract administration significantly ($P<0.05$) increase the haematological indices at all doses of the extract. Furthermore, the highest dose of the extract increased haematological indices significantly than the reference iron drug ($FeSO_4$) and iron-sufficient diets. Hence, the extract compares well with the reference iron drug ($FeSO_4$) as well as the iron-sufficient diets in the treatment of iron-deficient anaemia. This is an indication that the aqueous extract of *Alchornea laxiflora* leaf has anti-anaemic potential and thus lends credence to its use in folklore medicine in the management of iron deficient-anaemia.
[Oladiji, A.T., Olatunde, A. and Oloyede, H.O.B. **Evaluation of Anti-anaemic potential of aqueous extract of *Alchornea laxiflora* (Benth) leaf in iron deficient rats.** *Academ Arena* 2014;6(7):22-29]. (ISSN 1553-992X). http://www.sciencepub.net/academia. 5

Keywords: Haematological indices; Iron deficiency; Anaemia; *Alchornea laxiflora;* Haematinic potential

1.0 Introduction

Iron plays a vital role in the maturation, growth, and division of cells (Vandewalle *et al.*, 1985). Iron deficiency is the most common nutritional deficiency in the world and the most common cause of anaemia worldwide (John, 1995). The vulnerable groups are infants, young children and women of child-bearing age (WHO, 1968). The deficiency is usually due not to the absolute lack of the element in the diet but rather to its poor bioavailability. Iron bioavailability is influenced by the degree of iron deficiency of the individual, the adequacy of intestinal secretions, and the various components of food that inhibit or enhance iron absorption. Most cases of iron deficiency are mild and do not results in symptoms that are recognized as requiring medical attention (Oladiji, 2003).

Over the years, medicinal plants have been recognized to be of great importance to the health of individuals and communities. In traditional medicine of Nigeria, *Alchornea laxiflora* leaf is one of the plants used in the management of anaemia without scientific credence to its efficacy. *Alchornea laxiflora* (Benth) (Euphorbiaceae) is a forest understorey tree of about 6m high growing in Nigeria. It is also found in other part of Africa. The leaves play important role in the preservation of kolanuts, stem and branchlets are also used in Nigeria as chewing sticks. Decoction of the leaves is used in the treatment and management of inflammatory and infectious diseases as well as an important component of herbal antimalarial (Adewole, 1993), antibacterial (Lamikanra *et al*, 1990), anti-inflammatory and antimicrobial (Ogundipe et al 1999) formulations.

The aim of this work is to study the potential of aqueous extract of *Alchornea laxiflora* leaf in the treatment of anaemia in iron deficient albino rats while comparing its effect with that of a standard iron drug and iron sufficient diet.

2.0 Materials and Methods

2.1 Laboratory animals

Albino rats (*Rattus norvegicus*) of both sexes weighing between 51.17±1.02g were obtained from the small animal holding unit of the Department of Biochemistry, University of Ilorin, Ilorin, Nigeria.

2.2 Feed Components

Maize (*Zea may*) and locust bean [*Parkia biglobosa* (A.) Jacq] seeds were obtained from Baboko Market, Ilorin, Nigeria while the soybean oil used was a product of Grand Cereal and Oil Mills

Limited, Bukuru, Jos, Nigeria. The vitamin mix was a product of BASG Aktiengesellschaft, Germany Pantex, Holland. Component chemicals of the mineral mix used were products of BDH Chemicals Limited, London.

2.3 Plant identification and preparation of extract

The leaves of *Alchornea laxiflora* was obtained from Faculty of Agriculture, University of Ilorin, Ilorin, Nigeria and was authenticated in the Department of Plant Biology, University of Ilorin, Ilorin, Nigeria, where a voucher specimen (UIH 739) was deposited at the Departmental Herbarium.

The leaves of *Alchornea laxiflora* were separated from the stem and oven dried at 40°C for 72 hours to a constant weight. The dried leaves were then pulverized using Beltone Luinohun Blender/Miller III (model MS-223, Taipei, Taiwan). The powdered material was stocked in a plastic container from which 1000 g was extracted in 1.5 Litre of cold distilled water for 48 hours at 37^0C. This was then filtered with Whatman No. 1 filter paper. The filtrate was concentrated on a steam bath to give 24.7 g of the extract. The extract was reconstituted in distilled water to give the required doses of 100, 200 and 300 mg/kg body weight as used in this study. (Value arrived at from information obtained during ethnobotanical survey). The reconstituted aqueous extract was administered orally using oropharyngeal cannula to all the animals in different groups (Yakubu *et al.,* 2005).

Table 1: Feed Components of Iron Sufficient and Iron Deficient Diets

Feed Components	Iron sufficient (g/kg)	Iron deficient (g/kg)
Locust beans	710	710
Corn starch	40	40
*Soybean oil	40	40
Sucrose	100	100
Methionine	20	20
Lysine	10	10
**Vitamin mix	10	10
***Mineral mix	30	30
Fibre	40	40

*Soybean oil: Polyunsaturated Fatty acids (58%), monounsaturated fatty acids (29%) saturated fatty acid is (13%).

**Vitamin mix (per kg of diet): vitamin A, 100,000 lU; vitamin D_3, 10,000 lU; vitamin E, 100 mg; vitamin B_1, 20 mg; vitamin B_2, 40 mg; d-calcium pantothenate, 100 mg; vitamin B_6, 15 mg; vitamin B_{12}, 10µg; vitamin C, 250 mg; vitamin K_3, 15 mg; folic acid, 5000 mcg; nicotinic acid, 200 mg; biotin, 150 mcg; choline chloride, 400µg; inositol, 80 mg, vitamin c, 250mg; folic acid, 5000mcg.

***Mineral mix (g/kg diet): $CoCl_2.6H_2O$ (0.001), $CuSO_4.5H_2O$ (0.078), $MnSO_4.2H_2O$ (0.178), KI (0.032), KH_2PO_4 (10.559), NaCl (3.573), $MgSO_4.7H_2O$ (1.292), Zn $(CO3)_2$ (1.6), $CaSO_4$ (11.61), $FeSO_4.7H_2O$ (1.078).

Iron deficient diet contains no additional $FeSO_4.7H_2O$.

2.4 Composition of diet

The composition of iron deficient and iron sufficient diets per kg diet is shown in Table 1. The components of the diets were thoroughly mixed and made into pellets to ensure good handling by the animals. The proximate analyses of the compounded feeds were also carried out as shown in table 4.

2.5 Animal grouping and Administration of Plant Extract and Iron Supplement

The animals were individually housed in metabolic cages of 33cm × 20.5cm × 19cm under standard condition (12 hours light: 12 hours dark cycle; 28^OC and 40-55% humidity). Rats were then fasted for 24 hours (without food but given water) prior to the commencement of the experiment. The animal grouping consisted of an initial two groups:

A: Rats maintained on iron sufficient diet designated as ISG (iron sufficient group)

B: Rats maintained on iron deficient diet designated as IDG (iron deficient group)

Animals in groups A and B were maintained on their respective diets for 5 weeks. At the end of the 5 weeks feeding period, 4 rats each from IS and ID groups were sacrificed and their haematological indices were determined. The remaining rats in groups B were further grouped into six with four rats in each group as follows:

B1- Iron deficient rats fed on iron deficient diet for 14 days (iron deficient diet all through) designed as IDG (iron deficient group)

B2- Iron deficient rats fed on iron sufficient diet for two weeks (change of diet) designed as CDG (change of diet group)

B3- Iron deficient rats orally administered on daily basis for 14 days with reference iron supplement tablet (ferrous sulphate) designated as RDG (reference drug group)

B4- Iron deficient rats orally administered *A. laxiflora leaf* extract (100 mg/kg/rat/day) orally for 14 days designed as IDA-100mg.

B5- Iron deficient rats orally administered *A. laxiflora leaf* extract (200 mg/kg/rat/day) orally for 14 days designed as IDA-200mg.

B6- Iron deficient rats orally administered *A. laxiflora leaf* extract (300 mg/kg/rat/day) orally for 14 days designed as IDA-300mg.

The rest of the rats in group A were still fed on iron sufficient feed for days (iron sufficient all through) designated as ISG (iron sufficient group).

The aqueous extracts of *Alchornea laxiflora* leaf at various doses were administered to the various groups using cannula.

2.7 Phytochemical screening (Qualitative)

The presence of alkaloids and phlobatannins were determined according to the method described by Harborne (1973) while the method described by Odebiyi and Sofowora (1978) was used for flavonoids and tannins. Steroids and phenolics were qualitatively determined by the method of Trease and Evans (1989) while glycosides, Anthraquinones and saponins were determined by the methods of Sofowora (1993) and Wall *et al*. (1954) respectively.

2.8 Phytochemical screening (Quantitative)

Alkaloid, Saponins, Flavonoids and Anthraquinones were quantitatively determined by adopting the procedure described by Harborne, (1980), Obadoni and Ochuko (2002), Allen *et al.* (1973) and El-Olemy *et al.* (1994) respectively.

2.9 Proximate analysis of formulated diets and the plant extract

Proximate analysis carried out on the formulated diet and the plant extract included ash and organic mineral Content according to the method described by Pearson (1981); fat (ether extraction), crude fibre and crude protein were estimated according to the method described by Heidrich (1990) while carbohydrate was estimated by difference according to the method described by Oyeleke (1984). The mineral elements were determined by the wet digestion method described by Heldrich (1990).

2.10 Total Minerals

Samples were acid-digested, using nitric acid and perchloric acid mixture (HNO: HClO, 5:1 w/v). The total amounts of Na, K, Ca, Mg, Fe, and Zn in the digested samples were determined by atomic absorption spectrophotometry (Thermo–Elmental, Model 300VA) (Onwuka, 2005).

2.11 Collection of Blood Sample

The rats were placed under diethyl ether anesthesia, the neck area was quickly shaved to expose the jugular veins. The veins after being slightly displaced (to avoid contamination with interstitial fluid) were then sharply cut with a sterile scalpel blade and about $3cm^3$ of blood was collected into EDTA sample bottle for the haematological assay (Yakubu *et al.,* 2005).

2.12 Estimation of Haematological Parameters

The haemoglobin concentration was determined using the method described by Jain (1986). PCV was estimated using a Hawksley microhaematocrit centrifuge at 40-2 x g for 5min while red and white blood cells counts were determined using the Naubeaur haemocytometer, mean corpuscular volume (MCV) mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were calculated as described by Dacie and Lewis (1991).

2.13 Statistical Analysis

The data were expressed as mean ± standard error of mean (SEM). Statistical analysis was performed using analysis of variance (ANOVA) and Duncan multiple range test at 5% level of confidence ($p<0.05$).

3.0 Results

Table 2 shows the phytochemical present in the aqueous extract of *Alchornea laxiflora* leaf. Alkaloids, tannins, flavonoids, anthraquinones, saponin, phenolics, cardiac glycosides, steroids and phlobatannins were detected. The presence of triterpenes, chalcones, resins and cardenolides were not detected as shown in Table 2.

Table 3 shows the quantitative phytochemical screening of aqueous extract of *Alchornea laxiflora* leaf. The quantitative phytochemical screening revealed that the plant contain 33.67% flavonoids, 5.77% saponin, 2.5% alkaloids and 0.15% anthraquinones (Table 3).

Table 4 revealed that aqueous extract of *Alchornea laxiflora* leaf contained sodium (25.00mg/100 g), potassium (55.67mg/100 g), calcium (17.33 mg/100 g), magnesium (26.00 mg/100 g), iron (8.73mg/100 g) and zinc (7.77 mg/100 g).

Table 5 shows the proximate composition of the plant extract and the two diets. Proximate analysis of aqueous extract of *Alchornea laxiflora* leaf revealed that it contains a very high percentage of carbohydrate (42.60%) and crude fibre (26.07%) while others like protein (7.26%); fat (11.73%); ash (9.13%) and moisture (3.22%) are present in relatively small quantities. Proximate analysis of the diets showed that the components in iron deficient formulated diet were essentially similar to those in the iron sufficient formulated diet. On the other hand, the diets contains high percentage of protein and carbohydrate while others like fat, crude fibre, ash and moisture are present in small amount (Table 5). The carbohydrate and crude fibre contents of the plant extract are higher than in the diets while the protein and moisture contents of the diets are higher than in the plant extract (Table 5).

Table 6 shows the haematological parameters of rats fed with iron sufficient diet and iron deficient diet for the period of five (5) weeks. At the end of the five (5) weeks of feeding with iron deficient and sufficient diet, the level of haematological parameters of rats placed on iron deficient feed (IDF) decreases significantly ($p<0.05$) when compared with rats fed on iron sufficient feed (ISF). This is an indication that the rats fed on iron deficient diet were anaemic.

Table 2: Qualitative phytochemical analysis of aqueous *Alchornea laxifora* leaf extract

Phytochemicals	Status
Alkaloids	+
Flavonoids	+
Anthraquinones	+
Saponin	+
Tannins	+
Phenolics	+
C. glycoside	+
Cardenolides and Dienolides	–
Triterpenes	–
Steroids	+
Phlobatannins	+
Chalcones	–
Resins	–

Key= + present; - absent.

Table 3: Quantitative phytochemical analysis of aqueous *Alchornea laxifora* leaf extract

Phytochemicals	Quantitative (%)
Alkaloids	2.50±0.10
Flavonoids	33.67±0.58
Anthraquinones	0.15±0.00
Saponins	5.77±0.15

Each value is presented as mean ± S.E.M (n = 3).

Table 4: Mineral composition of aqueous extract of *Alchornea laxiflora* leaf

Minerals	Composition (mg/100g)
Na	25.00±1.15
K	55.67±0.88
Ca	17.33±0.88
Mg	26.00±1.15
Fe	8.73±0.12
Zn	7.77±0.09

Values are expressed as Mean ± SEM (n = 3).

Table 7 shows the effect of aqueous extract of *Alchornea laxiflora* leaf the haematological parameters (PCV, Hb, RBC, MCV, MCH and MCHC) of iron deficient rats. Result shows that there was significant decrease ($p<0.05$) in PCV, Hb, RBC, MCV, MCH and MCHC level in iron deficient group (IDG). However, oral administration of aqueous extract of *Alchornea laxiflora* leaf at all doses to the iron deficient rats significantly ($p<0.05$) increase the haematological parameter and these shows to be dose dependent. Although reference iron drug and iron sufficient diet also showed a significant increase ($p<0.05$) in these indices but this reversal effect is lesser than that produced by the extract (Table 7). The Rats placed on iron-deficient diet (not treated) showed lowest values for these haematological parameters when compared with those placed on iron-sufficient diet, reference iron drug and aqueous extract of *Alchornea laxiflora* leaf (Table 7).

Table 5: Proximate Composition of Iron Sufficient, Iron Deficient Diets and Aqueous extract of *Alchornea laxiflora*

Components (%)	*Alchornea laxiflora*	Iron sufficient feed	Iron deficient feed
Moisture content	3.22 ± 0.02^{c}	5.25 ± 0.03^{b}	5.75 ± 0.14^{a}
Ash content	9.13 ± 0.07^{a}	6.35 ± 0.02^{b}	6.43 ± 0.01^{b}
Crude Protein	7.26 ± 0.02^{b}	38.35 ± 0.09^{a}	38.13 ± 0.07^{a}
Crude Fat	11.73 ± 0.01^{a}	10.61 ± 0.01^{b}	10.57 ± 0.01^{b}
Crude Fiber	26.07 ± 0.07^{a}	5.53 ± 0.07^{b}	5.59 ± 0.01^{b}
Carbohydrate	42.60 ± 0.10^{a}	33.91 ± 0.15^{b}	33.53 ± 0.24^{b}

Values are expressed as Mean ± SEM (n = 3). Values in each row with different superscript are significantly different ($P<0.05$)

Table 6: Haematological Parameters of rats fed with iron deficient and sufficient diets for five weeks

	Iron Sufficient Group	Iron Deficient Group
PCV (%)	34.95 ± 3.85^{a}	28.75 ± 6.15^{b}
Hb (g/dL)	9.15 ± 0.750^{a}	6.70 ± 2.60^{b}
RBC ($10^{6}/\mu L^{3}$)	5.41 ± 0.08^{a}	4.60 ± 1.08^{b}
MCV (fL)	64.15 ± 5.85^{a}	62.90 ± 1.30^{b}
MCH (pg)	16.5 ± 1.50^{a}	14.00 ± 2.40^{b}
MCHC (g/dL)	26.25 ± 1.06^{a}	22.35 ± 6.01^{b}

Values are expressed as Mean ± SEM (n = 4). Values in each column with different superscript are significantly different ($P<0.05$). PCV: Packed Cell Volume; Hb: Haemoglobin; RBC: Red Blood Cell; MCV: Mean Corpuscular Volume; MCH: Mean Corpuscular Haemoglobin; MCHC: Mean Corpuscular Haemoglobin Concentration.

Table 7: Haematological parameters of iron deficient rats administered with aqueous extract of *Alchornea laxiflora* leaf

Groups	PCV (%)	Hb (g/dl)	RBC(10^6/µl^3)	MCVfL(µm^3)	MCH (pg)	MCHC(g/dl)
ISG	42.13±1.07^a	11.30±0.49^{abc}	5.72±0.14^a	73.98±1.51^a	17.78±0.80^{ab}	25.68±0.50^{abc}
IDG	34.78±1.66^b	7.88±0.95^d	4.28±0.48^b	66.48±1.72^{bc}	11.58±0.88^c	20.03±1.54^d
CDG	42.01±2.61^a	12.53±0.40^{ab}	5.92±0.28^a	76.61±1.24^a	19.40±1.02^a	22.30±1.75^c
RDG	41.50±0.48^a	9.80±0.56^c	6.05±0.31^a	62.40±1.35^d	15.65±0.32^b	23.13±1.09^{bc}
IDA-100 mg	40.18±0.33^a	10.50±0.55^c	5.32±0.61^{ab}	63.10±1.20^{cd}	17.35±0.83^{ab}	26.05±0.91^{ab}
IDA-200 mg	40.90±0.37^a	11.03±0.31^{bc}	5.87±0.32^a	62.53±0.84^d	15.95±1.07^b	26.95±0.66^a
IDA-300 mg	44.23±0.41^a	12.80±0.34^a	6.24±0.27^a	67.38±0.28^b	17.53±0.78^{ab}	27.48±0.17^a

Values are expressed as Mean ± SEM (n = 4). Values in each column with different superscript are significantly different (P<0.05). IDG: Iron Deficient Group; ISG: Iron Sufficient Group; CDG: Change of Diet Group; RDG: Reference Drug Group; IDA-100mg: 100mg/kg b.wt of *Alchornea laxiflora* leaf group; IDA-200mg: 200mg/kg b.wt of *Alchornea laxiflora* leaf group; IDA-300mg: 300mg/kg b.wt of *Alchornea laxiflora* leaf extract group; PCV: Packed Cell Volume; Hb: Haemoglobin; RBC: Red Blood Cell; MCV: Mean Corpuscular Volume; MCH: Mean Corpuscular Haemoglobin; MCHC: Mean Corpuscular Haemoglobin Concentration.

4.0 Discussion

Phytochemical is a natural bioactive compound found in plants, such as vegetables, fruits, medicinal plants, flowers, leaves and roots that work with nutrients and fibers to act as an defense system against disease or more accurately, to protect against disease (Krishnaiah *et al.*, 2009). An example of such plant is *Alchornea laxiflora*. *Alchornea laxiflora* has been used in most tropical countries as herb due to its high nutritional content and medicinal value in the alleviation of many ailments due to unaffordability of the cost of modern health care and orthodox medicine. In our present study of *Alchornea laxiflora* leaves, the phytochemical investigation indicates the presence of alkaloids (2.5%), flavonoids (33.67%), anthraquinones (0.15%), saponin (5.77%), tannins, phenolics, cardiac glycosides, steroids and phlobatannins (Table 2). Saponins and alkaloids have been reported to possess anti-anaemic potentials (Falcone *et al.*, 1997). Alkaloid inhibits cyclic adenosine monophosphate (cAMP) phosphodiesterase thereby accumulating cAMP (Magnani *et al.*, 1986). The effect stimulates phosphorylation of proteins and synthesis of protein, thereby enhancing erythropoiesis (Magnani *et al.*, 1986). Saponins are also known to inhibit platelet aggregation and thrombosis. Saponin containing herbs have been successfully used in the management of liver inflammation, as tonic sedative formulas and to promote and vitalize blood circulation (Shi *et al.*, 2004; Wang *et al.*, 2004). Since saponins are membrane active agents that lyse red blood cells or other wall, it is possible that the red blood cells were initially lysed by this plant; the cells overcome this inhibition by producing glycosidic enzyme which cleaves some of the terminal sugars from the saponin, thereby detoxifying it (Pathirana *et al.*, 1990). This detoxification of saponins, thus enhanced the proper utilization of the iron contained in the aqueous extract of *Alchornea laxiflora* leaf to synthesize heme/haemoglobin for new red blood cells thus leading to an improved Hb, PCV and RBC. Interestingly, saponins especially terpene glycosides enhance natural resistance and recuperative powers of the body (Singh *et al.*, 1991). Also, flavonoid which possess anti anaemic potential have veinotonic properties and protect capillaries (Bruneton, 1999). The anti-anaemic potential and haemoglobin restoring effect of aqueous extract of *Alchornea laxiflora* leaf as suggested by the data in the present study could be attributed in part to its phytochemical constituents.

Minerals play a vital role in the maintenance of human health (Muahamad *et al.*, 2010). For instance, Iron is an essential trace element for haemoglobin formation, normal functioning of the central nervous system and in oxidation of carbohydrates, proteins and fats (Adeyeye and Otokiti, 1999). In this study, the mineral composition of *Alchornea laxiflora* indicates the presence of calcium (17.33mg/100g), magnesium (26.00mg/100g), potassium (55.67mg/100g), sodium (25.00mg/100g), iron (8.73mg/100g) and zinc (7.77mg/100g) (Table 4). Hence, the presence of these minerals implies *Alchornea laxiflora* leaves could be utilized as a nutritionally valuable and healthy ingredient for animals. These nutrients may not be strictly medicinal but could be valuable in preventing diseases that are related to malnutrition e.g. iron deficiency anaemia.

Proximate analysis among others can give useful information on the chemical constituents of a material and their estimate in relation to other constituents (Oladiji *et al.,* 2005). In this study, the proximate analysis of *Alchornea laxif*lora leaf revealed that the plant contain carbohydrate (42.60%), crude fiber (26.07%) crude fat (11.73%), ash (9.13%), crude protein (7.26%) and moisture (3.22%) (Table 4). Proteins being a fundamental component of blood are found in aqueous extract of *Alchornea laxiflora* leaf. The relatively high percentage of carbohydrate in the extract indicates that the extract could be described as a carbohydrate containing material. Also, crude fiber content in the extract will help to maintain the movement of food through the gut and may be broken down by enzymes and bacteria in the gut to provide energy. The ash content is a reflection of the mineral contents preserved in the leaves. Minerals are essential for the proper functioning of tissues and act as second messenger.

It has been demonstrated previously that feeding animals with iron deficient diet for a period of four weeks or more will lead to iron deficiency anaemia in the rats (Hoffbrand and Broitman, 1969; Sriratanaban and Thayer, 1971; Lanzykowsky *et al*., 1981; Vieira *et al*., 2000) as also observed in this study. Iron deficiency anemia continues to be the most common specific nutritional deficiency in the world. Despite the advances in infant feeding during the last decades, it failed to eliminate iron deficiency as a public health problem (John, 1995). If not treated, iron deficiency anemia may cause stunted growth, impaired mental development, poor school performance, reduced productivity, increased morbidity and mortality, and lower self-esteemed. Iron is essential for all eukaryotes and most prokaryotes, where it is used in the synthesis of heme, iron-sulfur (FeS), and other cofactors. Fe-S proteins are involved in catalysis, redox reactions, respiration, DNA replication, and transcription. Iron homeostasis is tightly regulated to avoid iron toxicity or iron deficiency in normal condition. In human systemic iron metabolism, iron uptake, trafficking, export and fortification are highly regulated. (Tolentino and Friedman, 2007; King *et al.,* 2008; Hattangadi and Lodish, 2004; Ye and Rouault, 2010). The most reliable indication of iron deficiency anaemia is haemoglobin. This is because it is the iron-containing protein found in red blood cells that allows the red blood cells to function as the oxygen transport system to the tissues of the body. Next to haemoglobin in this regard is the haematocrit (Ht) or packed cell volume (PCV) which is a measure of the portion of the blood volume made up by red blood cells (Oladiji *et al.,* 2005). Result from table 1 showed the establishment of iron deficiency anemia in the first five weeks of this study. The significant decrease in the haematological parameters of the iron deficient group when compared with iron sufficient group is sufficient to conclude that the feed induced intended condition of this study i.e. iron deficiency anemia. In the present study, the haematological indices significantly increased in iron deficient groups after treatment with iron sufficient diet, reference iron drug and aqueous extract of *Alchornea laxiflora* leaf at doses of 100, 200 and 300mg/kg body weight.

The increase in the haematological indices exhibited by *Alchornea laxiflora* extract may be due to the presence of vitamins and mineral contents of the leaves of *Alchornea laxiflora*. These constituents are well known haemopoietic factors that have direct influence on the production of blood in the bone marrow. Also, the significant increase in the haematological parameters of the iron deficient rats following the administration of *Alchornea laxiflora* leaf may indicate that the plant extract has the ability to stimulate the erythropoietin release into the kidney which is the humoral regulator of RBC production (Sanchez-Elsner *et al.,* 2004). Erythropoietin increases the number of erythropoietin-sensitive committed stem cells in the bone marrow that are converted to red blood cell and subsequently to mature erythrocytes (Ganong, 1997).

5.0 Conclusion

This study has therefore lend credence to the haematinic potential of aqueous extract of *Alchornea laxiflora* leaf as widely acclaimed in traditional medicine. The haematinic potential of the plant might have been brought about by the phytochemicals which increase the haematological indices. Also, this may be made possible by its ability to make readily available iron which is needed in the restoration of the deficiency state. This may be an advantage to the poor in the less developed and developing countries in Africa who may be unlikely to have access to the conventional iron supplements.

Corresponding Author:
Professor Oladiji Adenike Temidayo
Department of Biochemistry,
Faculty of Life Sciences,
University of Ilorin, P.M.B. 1515, Ilorin,
Kwara State, Nigeria.
Telephone: +234 8033736996
E-mail: temidayooladiji@yahoo.com

References

1. Adewole AA. Personal communication with local traditional medical practitioner in Ibadan, Nigeria. 1993.
2. Adeyeye EI, Otokiti MKO. Proximate composition and some nutritionally valuable minerals of two varieties of capsicum annum (Bell and cherry peppers). Discovery and innovation. 1999; 11: 75-81.
3. Allen SE, Grinshaw HM, Parkinson JA, Quarmbay C. Chemical Analysis of Ecological Materials. 1st edition, Blackwell Scientific Publication, London. 1973.
4. Bruneton J. Pharmacognosie, Phytochimie et lantes médicinales. TEC & DOC et médicales internationales, éd., 3e, Paris, France. 1999; 1120 p.
5. Dacie JV, Lewis SM. Practical Haematology. Churchhill Livingstone. Edinbury. Seventh edition. 1991 P 521-534.
6. El-olemy MM, Al-muhtada FJ, Afifi AA (1994). Experimental Phytochemistry. A Laboratory Manual. King Saud University Press Riyadh, Saudi Arabia. 1994; Pp. 6 – 65.
7. Falcone A, Musto P, Rasario P, Rosella M. Compounds and Methods for treatment of chemotherapy – induced Anemia. European Journal of Haematology. 1997; 58:34 –319
8. Fermandes MIM, Galvao LC, Bortolozzi MF, Oliveira WP, Zucoloto S, Bianchi MLP. Disaccharidase levels in normal epithelium of the small intestine of rats with iron-deficient anaemia. Brazilian Journal of Medical Biological Research. 1997; 30, 849–854.
9. Ganong WF. A review of Medical Physiology Appleton & Lange Pubiisher. 1997; pp. a96.
10. Harborne JB. Phytochemical Methods. Chapman and Hall Ltd., London pp. 1973; 49-188.
11. Harborne JB. Phytochemical Methods. 2nd edition, Chapman and Hall, London. 1980; PP 288-293.
12. Hattangadi SM, Lodish HF. Regulation of erythrocyte life span: do reactive species set the clock. J Clin Invest. 2004; 117: 2075-77.
13. Heldrich K. Official Methods of Analysis of the Association of Official Analytical Chemisis. Arlington, Virginia, U. S. A. 1990; pp, 75-82
14. Hoffbrand AV, broitman SA. Effect of chronic nutritional iron deficiency of the small intestinal disaccharidase activities of growing dogs. Proceedings of the society for Experimental Biology and Medicine. 1969; 130: 595-598.
15. Jain NC. Schalm's Veterinary Hematology 4th Edn. Lea and fabiger, Philadelphia. 1986; pp: 564-572.
16. John NL. Iron metabolism and iron deficiency. In: Miller DR, Baehner 1- RL, eds. Blood diseases of infancy and childhood,7th ed, New York : Mosby. 1995; 193-219.
17. King SM, Donangelo CM, Knutson MD, *et al.*, Daily supplementation with iron increases lipid peroxidation in young women with low iron stores. Exp Biol Med. 2008; **233**: 701-7.
18. Krishnaiah D, Devi T, Bono A, Sarbatly R. (2009). Studies on phytochemical constituents of six Malaysian medicinal plants. Journal of Medicinal Plants Research. 2009; 3(2), pp. 067-072 ISSN 1996-0875© 2009.
19. Lamikanra A, Ogundani AO, Ogungbamila FO. Antibacteria constituents of *Alchornea cordifolia* leaves Phytotherapy Res. 1990; 4: 98-100.
20. Lanzkowsky P, Karayalcin G, Miller F, Lane BP. Disaccharidase values in iron deficient infants. Journal of Pediatrics. 1981; 99: 605-608.
21. Magnani M, Stocchi V, Cucchiarini L, Chiarantini L, Foranini G. Cell Biochem. Funct. 1986; 4: 263 –269.
22. Muahamad N, Faqir MA, Rai MA, Moazzam RK, SH, Muhammad SJ. An overview of anti-nutritional factors in cereal grains with special reference to wheat-A review. Pakistan Journal of Food Sciences.2010; 20 (1-4) p 54-61.
23. Obadoni BO, Ochuko PO. Phytochemical Studies And Comparative Efficacy Of The Crude Extracts Of Some Haemostatic Plants In Edo And Delta States Of Nigeria. Global Journal of Pure and Applied Sciences. 2002; 8(2):203-208.
24. Odebiyi OO, Sofowora EA. Phytochemical screening of Nigerian medicinal plants. Lloydia. 1978; 41(3): 234-246.
25. Ogundipe OO, Moody JO, Odelola HA. Biological activities of *Alchornea laxiflora* extractives. In: proceedings of the 1st International Workshop on the Standardization and Utilization of herbal Medicines: Challenges of the 21sT Century, University of Ibadan.1999; pp 201-208.
26. Oladiji AT, Abdullahi SA, Yakubu MT. Evaluation of haematinic potentials of aqueous extract of *Waltheria indica* (L) root on rats reared on iron sufficient and iron deficient diets feeds. Nigerian Journal of Biochemistry and Moecular Biology. 2005; 20(2): 115 – 122.
27. Oladiji AT. Tissue levels of lron, Copper, Zinc and Magnessium in iron deficient rats. Biochemistri. 2003; 14: 75-81.
28. Onwuka GL. Food Analysis and Instrumentation; Theory and Practice. 1st

Edition, Naphthal Prints, Lagos Nigeria. 2005; Pp 115-153.
29. Oyeleke OA. Outlines of Food Analysis. Department of BiochemistryAhmadu Bello University, Zaria, Nigeria. 1984.
30. Pathirana C, Gibney MJ, Taylor TG. Effects of soy protein and saponins on serum and liver cholesterol in rats. Atherosclerosis. 1990; 36, 595–599.
31. Pearson P. The Chemical Anaiysis of Food. J. A. Churchill Production. London. 1981
32. Sanchez -Eisner T, Ramirez JR, Rodriquez-Sanz F, Varela E, Bernabeu C, Botelia LM. A Crosstalk between hypoxia and TGF- beta o rchestraies erythropoietin gene reguiation through SPI and Smads, J. Mol. Biol. 2004; 336 (1): 9 - 24.
33. Shi J, Arunasalam K, Yeung D, Kakuda Y, Mittal G, Jiang Y. Saponins from edible legumes: chemistry, processing, and health benefits. Journal of Medicinal Food. 2004; 7, 67–78.
34. Singh N, Verma P, Mishara N, Nath R. A complete evaluation of some antistress agents of paints origin. lndian J. Pharmacol. 1991; 21: 99.
35. Sofowora A. Medicinal Plants and Traditional Medicine in Africa (2nd Edition). Spectrum Books Ltd, Ibadan, Nigeria. 1993; p. 289
36. Sriratanaban A, Thayer WR. Small intestinal disaccharidase activities in experimental iron and protein deficiency. American Journal of Clinical Nutrition. 1971; 24: 411-415.
37. Trease GE, Evans WC. Pharmacognosy, 11th edn., Bailliere *Tindall*, London. 1989; pp. 45-50.
38. Tolentino K, Friedman JF. An update on anemia in less developed countries. Am J Trop Med Hyg. 2007; 77(1): 44-51.
39. Vandewalle B, Granier AM, Peyrat JP, Bonneterre J, Lefebvre J. Transferrin receptors in cultured breast cancer cells. J Cancer Res Clin Oncol. 1985; 110:71-6.
40. Vieira MR, Galzvao LC, Fernandes MIM. Relation of the disaccharidases in the small intestine of the rat to the degree of experimentally induced iron-deficiency anaemia. Braz J Med Biol Res. 2000; 33: 539-544.
41. Wall M8, Krider MM, Krewson CF, Eddy CR, Wiliaman JJ, Cordell DS, Gentry HS. Steroidal sapogeninins Xlll. Supplementary table of data for sterciJal sapogenins Vll. Agrbuftural Rsearcfi SeMce Circular. 1954; 363, 17.
42. Wang J, Xu J, Zhong JB. Effect of Radix notoginseng saponins on platelet activating molecule expression and aggregation in patients with blood hyperviscosity syndrome. Alternative Medicine Review. 2004; 24, 312–316.
43. World Health Organization (WHO). Nutritional Anaemias. WHO technical reoort series. 402 Geneva. World Health Organization. 1968
44. Yakubu MT, Akanji MA, Oladiji AT. Aphrodisiac potentials of aqueous extract of Fadogia agrestis (Schweinf. Ex Heirn) stem in male albino rats. Asian J Androl. 2005; 7:399–404.
45. Ye H, Rouault TA. Human iron-sulfur cluster assembly, cellular iron homeostasis and disease. Biochemistry 2010; 49(24): 4945-56.

7/19/2014