**International Journal of Chemical Science**
**Online ISSN: 2523-2843, Print ISSN: 2523-6075**
**Impact Factor: RJIF 5.22**
**www.chemicaljournals.com**
**Volume 1; Issue 2; November 2017; Page No. 09-12**

# Comparative anti-anaemic potentials of *Telfairia occidentalis* milk, raw albumin and sugar combination, *Spondias mombin*, *Ribes nigrum* and ferrous gluconate in wistar rats

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**Abstract**
Investigation of the anti-anaemic activities of aqueous leaves extracts of *Telfairia occidentalis, Spondias mombin,* Ribes nigrum, Raw albumin and sugar combination and ferrous gluconate on experimental rats. The aqueous leaves extracts of *T. occidentalis, S. mombin, R. nigrum* were obtained by cold maceration and processed for 48 h, while albumin and sugar combination was mixed well by a mixer. Chloramphenicol model for experimental induction of anaemia in rats was utilized and the anti-anaemic activities was determined by measuring some haematological parameters of packed cell volume (PCV), haemeglobin count (Hb) and white blood cell count (WBC). The PCV and Hb levels of extracts treated rats were significantly ($p < 0.05$) increased compared to control group. The aqueous extract of *T. occidentalis* produced higher anti - anaemic activity compared to, *R. nigrum, S. mombin*, Raw albumin and sugar combination and ferrous gluconate but there was a small significant rise in the WBC level in all the extracts. The leaves extracts of *T. occidentalis, R. nigrum, S. mombin* and Raw Albumin and sugar combination has anti-anaemic activities, and this has revealed their uses as haematinics by many cultures.

**Keywords:** *telfairia occidentalis*, albumin and sugar, *ribes nigrum*, *spondias mombin,* ferrous gluconate, anaemia, rats

## 1. Introduction
Anemia is a health state where there is a lower than normal number of healthy red blood cells in the body or with lower than normal amount of hemoglobin in the red blood cells [1].
Red blood cells transport oxygen from the lungs to the rest of body, this action is performed by the haemoglobin. Red blood cells also transport waste products from the cells to the urinary and respiratory systems to be removed [1]. When either the number of red blood cells or the amount of hemoglobin is low, the body's cells receive less oxygen than normal. A low oxygen level can cause fatigue and other symptoms such as weakness, difficulty exercising, and light-headedness [1].
Many plants have been listed as remedies for anemia in the Nigerian traditional medicine including *Spondias mombin*, *Khaya grandifoliola*, *Telfairia occidentalis* [2, 3]. These plants extracts are sometimes prepared as polyherbal mixtures [2, 3]. Though most of the remedies have shown positive pharmacological activities with European scientific method, yet a lot of them lack scientific proof and are used superstitiously [2, 3, 4]. For example, despite being listed as remedy for anaemia [2, 3, 4], *Khaya grandifoliola* has been scientifically proven to lack anti-anaemic effect [2, 3, 4]. Regarding the economic importance of this disease in developing countries due to the lack of good diet has prompted the investigation of the listed used in different cultures to boost the blood levels in man and therefore the treatment and prevention of anaemia.
*Spondias mombin* belongs to the family Apacardiaceae, and it is a tropical tree up to 20 m tall [2, 3, 4], bearing blunt spines or knobs, growing easily from stakes for making fence and enclosure [1, 2]. It has a common name of Yellow mombin [2, 3, 5], others also call it "caja" in Brazil, Jobo-blanco (Colombia), Baja (Costa Rica), Jobito (panama), Ubo (Peru); and in the Northern, Western, and Eastern Nigeria it is called Isada, Iyeye and Ijinkaraka or Uvuru [2, 3, 5, 6]. The leaves of this plant have been reported by [2, 3, 4, 7] to be an excellent anthelminthic, anxiolytic [2, 3, 8], antipsychotic, sedative and antiepileptic [2, 3, 9] properties. Also, have been reported to exhibit antiviral and antibacterial [2, 3, 10]; and also used during and after delivery in pregnant women [2, 3, 11]. The leaves and bark powder are applied on wounds and inflammations in Peru [2, 3, 12]. It has also been reported by some researchers to contain saponins, terpenoids, iron, magnesium, potassium and some vitamins which has caused the various pharmacological activities [2, 3, 13].
*Telfairia occidentalis* is a vegetable crop that belongs to the family Curcubitaceae [2, 3]. It has local names of fluted pumpkin, and "ugu" in South eastern Nigeria where it is extensively cultivated [2, 3, 14] and used to prepare soup. Its leaves and stem are rich in Iron, Magnesiun, Potassium, Carotene and Vitamin C and used by pregnant women and to curb anaemia [2, 3, 14, 15].
*Ribes nigrum* is a deciduous and branched shrub which needs a fertile, damp, and well-drained soil to grow well [16, 17]. The plant grows up to the height of 1-2 m (3-6.5 ft) [16, 17, 18] and has an alternate, simple leaves with toothed edges that measures about 2.5-10.5 cm long and 2.5-12 cm broad [16, 17, 18]. The buds are yellowish, brown and oblong to ovoid to ellipsoid of about 4-7 mm in size. *R. nigrum* has bisexual flowers with colors of yellow or green of 10-11 mm long [16, 17, 18]. Flowers of this plant turns into a globe shaped with sparsely glandular fruit measuring 0.8-1.4 cm and 1 cm in

diameter [16, 17, 18], with an aromatic fruitings which are black in color with smooth peel [16, 17, 18]. The fruits of *R. nigrum* has a high content of anthocyanins which possess antioxidant activities and help in many health enhancing activities [16, 17, 18]. It has high content of antioxidants in comparison to other fruits and vegetables like blueberries [16, 17, 18]. Aside its taste, it has high nutritional and Vitamin C values that help the body to absorb iron and also gives immunity to the body to fight infections and diseases including flu, cold, vision and cardiovascular system enhancer [16, 17, 18].

Egg white or albumin are rich source of proteins and contain a lot of antioxidants that will help in stocking up vitamins in the body when you are suffering from anemia. A large egg is said to contain 1 mg of iron and hence consumption of an egg white everyday will help in fighting anemia [19]. The albumin of the egg helped to increase the Hb in the blood to carry the oxygen needed by the body, it also help to increase the WBC to help fight diseases including bacteria and viruses.

## 2. Materials and Methods

### 2.1 Plant Materials

The fresh leaves of the *T. occidentalis* and *S. mombin* and fruits of *R. nigrum* were collected from a farm in Ohuhu Umuahia and fresh chicken eggs were bought from Watt market Calabar and authenticated by a Taxonomist from Botany Department of the University of Calabar. The leaves and fruits were washed very well with clean tap water then with distilled water, they were later chopped into pieces by knife separately, and shade dried the chopped leaves and fruits separately. The small pieces of leaves and fruit were pulverized for further study. The aqueous extracts were prepared by macerating powders of *T. occidentalis*, *S. mombin* and *R. nigrum* fruits in cold water for 48 hours. After filtration, the extracts were concentrated separately using rotary evaporator.

### 2.2 Animals

Adult Wistar rats of different sexes weighing between 180-250g were kept in cages at the animal house of the Department of Pharmacology of the University of Calabar, and were used for this study. The animals were maintained under standard laboratory situations and had free access to standard pellets (Vital Feeds Plc, Nigeria) and clean water.

### 2.3 Induction of anaemia

Prior to experimental uses, the animals were allowed for two weeks to acclimatize. Haematological parameters were initially determined for untreated rats according to established method used earlier by [2, 3] although with slight modifications. Anaemia was induced by oral administration of chloramphenicol (50 mg/kg) for 2 weeks in six groups each containing five rats each with anaemia confirmed by the markedly low PCV compared to untreated rats.

### 2.4 Anti-anaemic screening

Anaemic groups of rats received oral administration of *T. occidentalis*, *S. mombin*, *R. nigrum*, Albumin and sugar combination and ferrous gluconate respectively. The positive control (reference drug), ferrous gluconate (900 mg/kg) and negative control, distilled water were similarly given to the two control groups respectively, All the treated animals were fed *ad libitum* with water and pellets all through the study period. Blood was collected via gentle pin prick then massage of the tail and was used for the parameters been studied - PCV, Hb, WBC before and after administration of the extracts and reference drug. PCV was determined using Wintrobes haematocrit tube by microhaematocrit centrifuge method, Haemoglobin was determined by Colorimentric method using Sahli haemometer kit and WBC was determined using improved Neubauer counting chamber via Hemocytometer.

### 2.5 Statistical analysis

Data were expressed as mean ± standard error of mean (SEM). Statistical comparisons were performed by one-way ANOVA, followed by Tukey-Kramer multiple comparisons test and student-Newman-Keuls multiple comparisons test and the values were considered statistically significant when p-value is less than 0.05 ($p<0.05$).

## 3. Results

**Table 1:** PCV (%) values on days 1,14 and 21.

| Groups | Day 1 | Day 14 | Day 21 |
|---|---|---|---|
| Normal control | 39.1 ± 0.3 | 39.1 ± 0.2 | 39.2 ± 0.2 |
| Anaemic rats | 23.3 ± 0.2 | 20.1 ± 0.3 | 16.2 ± 0.2 |
| Anaemic rats + *T.occidentalis* | 25.3 ± 0.3 | 46.3 ± 0.3 | 51.6 ± 0.2 |
| Anaemic rats + *S.mombin* | 26.2 ± 0.3 | 45.2 ± 0.2 | 49.6 ± 0.2 |
| Anaemic rats + *R. nigrum* | 25.3 ± 0.3 | 47.1 ± 0.3 | 49.5 ± 0.2 |
| Anaemic rats + Albumin+Sugar | 27.6 ± 0.3 | 48.3 ± 0.2 | 49.4 ± 0.4 |
| Anaemic rats + Ferrous gluconate | 25.2 ± 0.3 | 47.1 ± 0.3 | 47.6 ± 0.2 |

**Table 2:** Hb (g/dL) values on days 1, 14 and 21.

| Groups | Day 1 | Day 14 | Day 21 |
|---|---|---|---|
| Normal control | 13.2 ± 0.3 | 13.3 ± 0.2 | 13.3 ± 0.3 |
| Anaemic rats | 7.7 ± 0.3 | 7.5 ± 0.4 | 7.6 ± 0.3 |
| Anaemic rats + *T.occidentalis* | 8.4 ± 0.3 | 14.2 ± 0.3 | 16.2 ± 0.3 |
| Anaemic rats + *S.mombin* | 8.5 ± 0.2 | 13.2 ± 0.3 | 15.1 ± 0.3 |
| Anaemic rats + *R. nigrum* | 7.6 ± 0.2 | 13.0 ± 0.3 | 14.9 ± 0.3 |
| Anaemic rats + Albumin+Sugar | 8.1 ± 0.3 | 13.6 ± 0.3 | 14.8 ± 0.2 |
| Anaemic rats + Ferrous gluconate | 8.4 ± 0.2 | 14.3 ± 0.3 | 14.6 ± 0.3 |

**Table 3:** WBC ($10^3$/µL) values on days 1, 14 and 21.

| GROUPS | DAY 1 | DAY 14 | DAY 21 |
|---|---|---|---|
| Normal control | 3.96 ± 0.3 | 3.96 ± 0.3 | 3.98 ± 0.4 |
| Anaemic rats | 0.65 ± 0.2 | 0.66 ± 0.3 | 0.65 ± 0.2 |
| Anaemic rats + *T.occidentalis* | 0.62 ± 0.3 | 4.1 ± 0.3 | 5.3 ± 0.4 |
| Anaemic rats + *S.mombin* | 0.66 ± 0.3 | 3.9 ± 0.2 | 4.8 ± 0.3 |
| Anaemic rats + *R. nigrum* | 0.62 ± 0.2 | 3.4 ± 0.2 | 4.6 ± 0.3 |
| Anaemic rats + Albumin+Sugar | 0.65 ± 0.3 | 3.8 ± 0.3 | 4.5 ± 0.2 |
| Anaemic rats + Ferrous gluconate | 0.63 ± 0.2 | 3.5 ± 0.3 | 4.3 ± 0.3 |

## 4. Discussion

The developing nations rely more on medicinal plants, although its use is still poorly understood by the general public, although they are of great importance to health of individuals and the general community [20]. Hematological parameters are useful indices that can be employed to assess the toxic potentials of plant extracts in living system [21, 22].

They can also be used to explain blood relating functions of chemical compound and plant extract, such laboratory investigation have been reported to be highly sensitive, accurate, reliable and remains the bedrock of ethical and rational research, disease diagnosis, prevention and treatment [23]. Increase in PCV was observed in wistar rats administered with *T.occidentalis*, *S.mombin*, *R. nigrum*, Albumin + Sugar combination, and Ferrous gluconate when compared to the control, this implies that the extracts caused a good flow in the osmoregulatory system of the blood cells and helped in the decrease of the oxidative injury to the cell membrane, this plants extracts and Al + S combination helped to raise the haemopoietic system and has prevented lysis of blood cells. This work has shown that this plant extracts has aided erythropoiesis in the rats, and therefore helped the oxygen carrying capacity of the blood which has caused a surge in the oxygen delivered to the tissues.

There was also an increase in the Hb levels on the day 14 and 21 when compared with the control, but more on day 21. Hemoglobin in the blood help to carry oxygen from the respiratory organs (lungs or gills) to the rest of the body (i.e. the tissues). There it releases the oxygen to permit aerobic respiration to provide energy to power the functions of the organism in the process called metabolism. There was a steady rise in Hb levels in the anaemic rats treated with the plant extracts, Albumin + Sugar combination and ferrous gluconate, but more appreciable rise in the anaemic rats treated with *T.occidentalis.*

Likewise, there was an increase in WBC. WBC usually help the body against infections and tissue damage and therefore has immune boosting effects on the rats, showing that there was a raised vascular permeability. Immune boosters are usually giving to harmonize and strengthen degenerative body system and then assist the immune system to fight invading agents like bacteria and viruses [21, 24, 25].

## 5. Conclusion

This work has shown that this plant extracts has aided erythropoiesis in the rats, and therefore helped the oxygen carrying capacity of the blood which has caused a surge in the oxygen delivered to the tissues. The isolations and characterization of the active ingredients in this researched plants followed by further pharmacological and clinical studies would aid in the production of novel herbal drugs for the treatment and prevention of Anaemia a world health issue that is rampant in developing nations, which will be affordable and readily available as an alternative therapies to the conventional medications.

## 6. Consent

Not applicable

## 7. Ethical Approval

All the authors's hereby declare that all experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 declaration of Helsinki.


## 8. Acknowledgements

The authors are grateful to all the staff of the Department of Pharmacology for their technical assistance.


## 9. Competing Interests

All authors have declared that no competing interests exist.

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