



## Evaluation of Haemopoietic Parameters of Selected Ethnomedicinal Remedies

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### Abstract

**Introduction:** According to the World Health Organization (WHO), approximately 2 billion people, or 30% of the world's population, are affected by anaemia. In low- and middle-income countries, the prevalence of anaemia is highest among women of reproductive age and young children, with estimates suggesting that up to 60% of pregnant women and nearly 50% of preschool children suffer from anaemia.

**Aim:** The aim of this study is to evaluate the haemopoietic parameters of four traditionally used blood building plants; *Justicia carnea*, *Jathropha tanjorensis*, *Sorghum bicolor* and *Tetracera alnifolia*; whose anti-anaemic parameters have not been compared.

**Method:** The crude extracts were obtained by macerating 100g of each plant sample in methanol and dichloromethane, followed by concentrating the extracts using a rotary evaporator and drying in a desiccator. Phytochemical screening, elemental analysis, vitamin B3, B6, B12 analysis and GC-MS were carried out on the plant samples; *J. tanjorensis*, *J. carnea*, *S. bicolor* and *T. alnifolia*.

**Results:** Phytochemical screening showed the presence of Triterpenoids, Flavonoids, Tannins, saponins, Phlobatamins, Anthraquinones, Cardiac glycosides, Steroids, Carbohydrates, and proteins. GC-MS analysis revealed some compounds.

**Conclusion:** A comparison of the concentrations of the mineral and vitamins analysed revealed that *J. tanjorensis* had high concentrations of copper, iron and zinc, but was not statistically significant compared to the other plants investigated. The results obtained from the analysis of the vitamins, showed that *Sorghum bicolor* had high concentrations of vitamins: B3, B6 and B12 to the plants. Also, vitamin B3 was significantly higher at  $p < 0.05$  in *Sorghum bicolor*. The findings from this study showed that the investigated plants are suitable candidates for the development of blood building medications and their use in traditional settings as blood tonics is warranted. Furthermore, *Sorghum bicolor* is the most promising of the four medicinal plants as a candidate for further therapeutic studies.

### More Information

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### Keywords:

Haemopoietic,  
*Jathropha tanjorensis*,  
*Justicia carnea*,  
*Sorghum bicolor*,  
*Tetracera alnifolia*



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### Introduction

Anaemia, one of the oldest, most common and widespread blood disorder, is a public health problem in both developing and developed countries.<sup>1</sup> About half a billion women between the ages of 15–49 years and 269 million children between 6–59 months of age worldwide are affected by anaemia. The WHO in 2019, recorded that 30% (539 million) of non-pregnant

women and 37% (32 million) of pregnant women suffered from anaemia. Although anaemia affects all individuals at all stages of life cycle, preschool children and women of reproductive age are the most vulnerable population groups.<sup>1</sup>

At least 59% of children between 6 and 59 months and 40% of women of reproductive age are anemic. The regional WHO data on anaemia, reported that Africa

and South-East Asia were the most affected with an estimated 106 million women and 103 million children affected by anaemia in Africa, while 244 million women and 83 million children were anaemic in South-East Asia [1].

Iron deficiency which is a result of low intake of bioavailable dietary iron, is considered as the major cause of anaemia in developed countries. However, iron deficiency accounts for 50% of anaemia in developing nations. Other causes include; infection with hookworms, schistosomiasis, Malaria and HIV, other micronutrients deficiency, trauma and blood loss [2].

Medicinal plants have long been employed in the treatment of various ailments and even in the case of anaemia, botanical medicines need to be explored as conventional medicines for the treatment of anaemia are grossly expensive and lies beyond the reach of the poor masses who are at high-risk of dying from anaemia, that may well be prevented.

This scenario is further justified as herbal medicines have proven effective in the treatment of various diseases but since the emergence of orthodox medicine, the use of herbs for therapeutic purposes has suffered a major setback. Hence, this research was designed to address selected medicinal plants employed indigenously for the treatment of anaemia and compare their potency with an Orthodox medicine with similar properties.

For the purpose of this research, *Jathropa tanjorensis*, *Sorghum bicolor*, *Justicia secunda*, and *Tetracera alnifolia* were investigated for their blood building parameters and compared with a clinically-used blood tonic.

***Tetracera alnifolia***; is a species of flowering plant in the family Dilleniaceae. It is commonly known as the "saddle leaf" or "saddle vine" due to the shape of its leaves, which resemble saddles. This plant is native to Southeast Asia and is commonly found in the forests and riverbanks of Malaysia, Thailand, and Indonesia. *Tetracera alnifolia* bark is traditionally used in some cultures in the treatment of diarrhea, cough, STDs, malaria, stomach problems, inflammation and pain [3-8]. In the Southwest of Nigeria, decoctions of the leaves sheaths are used in the treatment of anaemia [9].

The bark contains compounds such as alkaloids, flavonoids, and tannins that have been shown to have pharmacological properties [10]. These compounds are believed to stimulate the production of red blood cells and increase the concentration of hemoglobin in the blood. However, more research is needed to fully understand the mechanism of action and potential side effects of *Tetracera alnifolia* bark as an anti-anaemic agent.

Presently, *Tetracera alnifolia* Willd is one of the herbal components in the improved Congolese drug, "Tetra" used for pain and inflammation management [11].

***Jathropa tanjorensis***; This is a perennial herb that belongs to the family Euphorbiaceae; its common names include; Catholic vegetables, Hospital too far, lapalapa and Iyana Ipaja in Yoruba language. The leaves are employed traditionally in the treatment of anaemia, cardiovascular diseases and diabetes. It is a traditionally used medicinal plant in South-eastern Nigeria with many claims from local consumers that it possesses blood replenishing properties. It has been reported that *Jathropa* leaves are rich in beta blockers, anti-cancer agents [12], anti-anemic [13], anti-microbial activities [14] anti-plasmodial [14] and anti-oxidant effect against oxidative stress induced by malaria parasite [15-17].

***Justicia carnea***; Brazilian plume flower family Acanthaceae is a flowering plants consisting about 600 species that are widely distributed in the tropics. In the review of the genus: *Justicia* in 2011, this specie was not documented to be pharmacologically active.<sup>18</sup> However, over the last decade there have been several publications on this specie. In Nigeria, the plant is commonly known as "Blood of Jesus leaf" and known indigenously as "Ogwu obara" by the Igbo tribes of Nigeria. It is popularly used as a blood tonic, especially by pregnant women due to its ability to ameliorate anaemic conditions [19] Numerous species of the genus; *Justicia* have documented use in the treatment of arthritis, rheumatism, malaria, kidney infections, fever, cancer, HIV, epilepsy, mental ill-health, inflammation, whooping cough, anaemia, tumour, liver disease, hepatitis, diabetes, fertility control, typhoid and bronchitis [18]. Other uses of the species include; insecticidal, allelopathy and ornamental uses [18].

The Modulatory activity of *Justicia carnea* in Plasmodium Infected Mice has been studied; the results obtained showed the plant possessed antimalarial activities [20]. Studies on the nutritive properties of aqueous extract of *Justicia carnea* leaves and its effects on the haematological and some biochemical indices of anaemia induced male wistar albino rats, have been undertaken [21]. A similar study on the Haematological and biochemical studies on *Justicia carnea* leaves extract in phenylhydrazine induced-anemia in albino rats have been reported [22]. The findings from these studies validated the plant's traditional use as a blood building therapy. The Antibacterial Activity of the aqueous extracts of *J. carnea* on Some Isolated Bacterial was recently reported [19]. The Anti-obesity, antioxidant and *in silico* evaluation of *Justicia carnea* methanol extract demonstrated that the plant possessed potential inhibitors of an enzyme linked with obesity [23]. Phytochemical evaluation of *J. carnea* revealed the



presence of alkaloids, carbohydrates, flavonoids, phenols, terpenoids, tannins, glycosides and reduced sugars. The following vitamins: A, B1, B6, B9, B12, C and E have been reported in the extracts of the plant. Similarly, the mineral content evaluation of *J. carnea* documented iron and calcium as the most abundant minerals in the plant, while copper, manganese and zinc had low concentrations [21].

***Sorghum bicolor***; also known as sorghum, is a cereal grain crop that is widely cultivated for its edible seeds. While it is not commonly known for its blood forming ability, sorghum has been found to contain several bioactive compounds that may have potential health benefits, including the promotion of blood health. *Sorghum bicolor* has a variety of uses in traditional medicine. Ethnopharmacological documentations showed that the plant is an all-purpose medicinal plant with notable uses such as anticancer, anti-inflammatory, anti-irritant and anti-epilepsy. The plant is also used as an antidote to gastro-intestinal problems, as a diuretic, antioxidant and an emollient for skin [24]. Sorghum has been in use traditionally in developing countries such as Nigeria for combating anaemia, and in the management of sickle cell diseases in several indigenous communities [25,16], and in the treatment of viral and infectious diseases [27]. The plant is rich in minerals and bioactive compounds.

One of these bioactive compounds is iron [28], which is an essential nutrient for the production of red blood cells. Iron is involved in the formation of hemoglobin, the protein in red blood cells that carries oxygen from the lungs to the tissues. Additionally, sorghum is a good source of folate [29,30], which is also important for blood health as it helps the body produce and maintain new cells, including red blood cells.

It has been reported that Sorghum contains more antioxidants in comparison to other cereals [31,32]. Thus, regular consumption of sorghum has immense potentials in curbing the rise in cardiovascular diseases. Some of the antioxidants contained in the plant are anthocyanins and phenolic acids, which have been shown to have anti-inflammatory and anti-cancer properties [33,34]. Inflammation can impair blood formation, so the antioxidant properties of sorghum may help promote healthy blood formation. Again, sorghum has been reported from studies to demonstrate significant hypoglycemic activity comparable to glibenclamide [35], and also found useful in mitigating against gastrointestinal discomfort experienced by patients with celiac disease [36]. Further studies have demonstrated the anti-inflammatory, antinociceptive [37], hematopoietic and hepatoprotective [26] activities, including the plant's extract haemoglobin-boosting [38,39] and immunity enhancement through the regulation of splenocytes formation [40], have been documented.

In this study some blood-building parameters were investigated. These include some minerals- Copper, Iron, manganese and zinc, vitamins, such as Vitamins B3, B6, B12 and the possible nonpolar compounds which may be implicated in the process of blood building.

## Methods and Materials

### Collection of Plant Materials

The plant samples were obtained from Rumuomasi market in Obio/Akpor Local Government Area, Rivers State and were identified by a botanist; Dr. Suleiman Mikailu of the Department of Pharmacognosy and Phytochemistry, University of Port Harcourt. The plant materials were dried for a period of 14days after which they were pulverized.

### Extraction of the Plant Materials

Using the American National Cancer Institute (NCI) method of extraction [41], the crude extracts were obtained from 100g of each plant sample which was macerated in organic solvents - Methanol and dichloromethane combined in a 1;1 ratio, after which they were concentrated using a rotary evaporator, with the water bath temperature set at 40 °C. The extracts were further dried in a desiccator to remove any trace of solvent. The extract was weighed, stored in airtight container and preserved in refrigerator for further analysis.

### Phytochemical Tests

Preliminary phytochemical screening was carried out on all the crude leaf extracts (dichloromethane and methanol) using standard procedures as described by Trease and Evans [42], Sofowora [43] and Harborne [44].

### Materials and Apparatus

Grinding machine, weighing balance, measuring cylinders, wide mouth bottles, spatula, foils, rotary evaporator, water bath, desiccator, airtight container, separating funnel etc.

### Drugs, Chemicals and Reagents

Dichloromethane (Sigma-Aldrich brand), methanol (Sigma-Aldrich brand), water, n-hexane (Sigma-Aldrich brand), hydrochloric acid, magnesium metal, sodium hydroxide, ferric chloride, dragendorff's reagent, wagner's reagent, olive oil, hydrochloride acid, chloroform, acetic anhydride, sulphuric acid, Fehling's solution A + B, molisch reagent, million's reagent, glacial acetic acid, picric acid solution, distilled water.

### GC-MS Analysis

The gas chromatography mass spectrometry (GC-MS) analysis of the crude methanol/dichloromethane extract of *Jathropa tanjorensis*, *Justicia carnea*, *Sorghum bicolor* and *Tetracera alnifolia* were quantitatively determined using an Agilent 7890B GC system coupled with an Agilent 5977A MSD with a Zebron-5MS column (ZB-5MS 30 m × 0.25 mm × 0.025 µm) (5%-phenylmethylpolysiloxane). The GC-grade



helium served as the carrier gas at a constant flow rate of 2 mL/min. The crude extract was dissolved with ethanol and filtered before use. The column temperature was maintained at 60°C and gradually increased at 10°C per minute until a final temperature of 300°C was reached. The time taken for the GC-MS analysis was 30 min. The compounds were identified based on computer matching of the mass spectra with the NIST 11 MS library (National Institute of Standards and Technology library).

#### Methods for the Elemental Analysis

Heavy metal analysis was conducted using Agilent FS240AA Atomic Absorption Spectrophotometer according to the method of APHA 1995 (American Public Health Association) [45].

Working principle: atomic absorption spectrometer's working principle is based on the sample being aspirated into the flame and atomized when the AAS's light beam is directed by means of the flame into the monochromator, and onto the detector that measures the amount of light absorbed by the atomized element in the flame. Since metals have their own characteristic absorption wavelength, a source lamp composed of that element is used, making the method relatively free from spectral or radiational interferences. The amount of energy of the characteristic wavelength absorbed in the flame by the sample is proportional to the concentration of the element in the sample.

#### Sample Digestion Method

Sample Digestion was carried out, prior to the AAS analysis according to the method of [46].

Procedure:

Weigh out approximately 2g of the dried sample material into the digestion flask, followed by the addition of 20ml of the acid mixture (comprising of 650ml Conc HNO<sub>3</sub>; 80ml perchloric acid; 20ml Conc H<sub>2</sub>SO<sub>4</sub>).

The flask is heated, until a clear digest is obtained.

Dilute the digest with the addition of distilled water to the 100ml mark.

#### Preparation of Reference Solutions

Preparation of several standard metal solutions in the required concentration range is done, followed by the preparation of the reference solutions by diluting the single stock element solutions with water containing 1.5 mL concentrated nitric acid/liter. Calibration blank solution was prepared using all the reagents except for the metal stock solutions.

Calibration curve for each metal was prepared by plotting the absorbance of standards versus their concentrations.

#### Methods for Vitamin B Analysis

##### Determination of Vitamin B3:

- A 5g of sample was dissolved in 20ml of anhydrous glacial acetic acid and warmed slightly.
- 5ml of acetic anhydride was added and mixed.

- 2-3 drops of crystal violet solution were added as indicator.

Titrate with 0.1M perchloric acid to a greenish blue colour.

Calculation:

$$\text{Vitamin B3} = \frac{\text{titer value} \times 0.0122}{0.1} \quad (1)$$

##### Determination of Vitamin B6:

- 5g of sample was dissolved in a mixture of 5ml of anhydrous glacial acetic acid and 6ml of 0.1m mercury II acetate solution.

- 2 drops of crystal violet were added as indicator.

- Titrate with 0.1m perchloric acid to a green colour end point.

- Calculation: each meal of 0.1M perchloric acid is equivalent to 0.02056g of C<sub>8</sub>H<sub>11</sub>NO<sub>3</sub>HCl

##### Determination of Vitamin B12:

Spectrophotometric determination of cyanocobalamin in serum preparations by coupling reactions with pyridine.

Sample preparation: Weighed equivalent of 0.1ml of sample was taken into the separator. In the separator, 5 ml of water was added, mixed well and extracted with 5 ml chloroform. The water layer was discarded, while the chloroform layer was poured into a dry 50 ml volumetric flask by-passed through anhydrous sodium sulphate and made up to 50 ml with chloroform.

Procedure: 2ml sample and blank solution was taken into test tube. In each test tube, added 2 ml of 0.2%-solution of phenyl hydrazine (in hydrochloric acid and alcohol in ratio of 1:5 v/v) and mixed well. Followed by heating on water bath to almost dryness and cooled at room temperature. 2 ml solution mixture (ammonia and alcohol in ratio of 1:1) was added in each test tube to 1 ml pyridine. The absorbance recorded at 635 nm against blank. Standard cobalamin was also analyzed and treated same as sample. Calibration curve was plotted, and the concentration of sample extrapolated.

#### Comparison of Sample Parameters with a Known Blood Tonic

The Parameter values obtained from the samples were compared to a known anti-anaemic Orthodox medication- Ferglobin syrup which contains; Iron 100ppm, zinc 30ppm, Cyanocobalamin 5microgram, zinc sulphate 5mg.

#### Statistical Analysis

Each test was carried out in triplicate. The values were expressed as mean ± standard error of mean (SEM). The Dunnett one-way analysis of variance (ANOVA) was used to determine the significant differences among all the plants extracts against the control and the *P* value < 0.05 was considered as significant. All statistical analysis was performed using Graph Pad Prism version 8.0 software.



## Results

Table 1 revealed the presence of some chemical classes of the bioactive components in the organic extracts of the investigated plants. Alkaloids, tannins, saponins,

steroids, triterpenoids, carbohydrates and proteins were detected in all the plants. The absence of cardiac glycosides was observed in *J. tanjorensis* which was the only plant with flavonoids present.

**Table 1: Phytochemical Screening Results**

| Tests              | Sample A | Sample B | Sample C | Sample D |
|--------------------|----------|----------|----------|----------|
| Alkaloids          | +        | +        | +        | +        |
| Tannins            | +        | +        | +        | +        |
| Saponins           | +        | +        | +        | +        |
| Flavonoids         | —        | —        | +        | —        |
| Antraquinones      | +        | —        | +        | +        |
| Plobatamines       | —        | +        | —        | —        |
| Steroids           | +        | +        | +        | +        |
| Triterpenoids      | +        | +        | +        | +        |
| Cardiac glycosides | +        | +        | —        | +        |
| Carbohydrates      | +        | +        | +        | +        |
| Protein            | +        | +        | +        | +        |

**Note:** A= *Tetracera alnifolia*, SAMPLE B= *Justicia carnea*, SAMPLE C= *Jathropha tanjorensis*, SAMPLE D= *Sorghum bicolor*, presence=+, absence = -

Documented phytochemical evaluation of *J. carnea* revealed the presence of alkaloids, carbohydrates, flavonoids, phenols, terpenoids, tannins, glycosides and reduced sugars [21]. Similarly, the presence of alkaloids, flavonoids, tannins and saponins have been reported in *J. tanjorensis* [47].

Previous phytochemical investigations on *Tetracera alnifolia* by Gbadamosi et al., reported the presence of

saponins and cardiac glycosides [9]. In this research more phytoconstituents were detected.

*Sorghum bicolor* extracts did not show the presence of flavonoids. This finding is in agreement with an earlier study which detected the presence of alkaloids, cardiac glycosides, saponins, steroids, terpenoids and quinones in the seed oil of *sorghum bicolor* [48].

The results of the elemental analysis of the plants' extracts are presented in Table 2.

**Table 2: Results of Elemental Analysis**

| Parameters      | Sample A | Sample B | Sample C | Sample D | Feroglobin syrup |
|-----------------|----------|----------|----------|----------|------------------|
| COPPER (PPM)    | 0.282    | 0.178    | 0.378    | 0.288    | 2.00             |
| IRON (PPM)      | 0.1583   | 1.677    | 1.723    | 1.487    | 100.00           |
| ZINC (PPM)      | 0.385    | 0.433    | 0.585    | 0.332    | 30.00            |
| MANGANESE (PPM) | 0.444    | 0.544    | 0.844    | 0.944    | 2.50             |

**Note:** A= *Tetracera alnifolia*, SAMPLE B= *Justicia carnea*, SAMPLE C= *Jathropha tanjorensis*, SAMPLE D= *Sorghum bicolor*,

The findings from this investigation revealed that the plants all had the key elements required for blood building. A graphical illustration of the values of the minerals is contained in Figure 1.

It could be observed that *J. tanjorensis* had high concentrations of copper, iron and zinc compared to the plants. The plant had slightly less concentration of manganese in comparison to *sorghum bicolor*. The blood tonic: Feroglobin had more concentrations of the elements than the plants' extracts.

The vitamin B analysis is shown in Table 3.

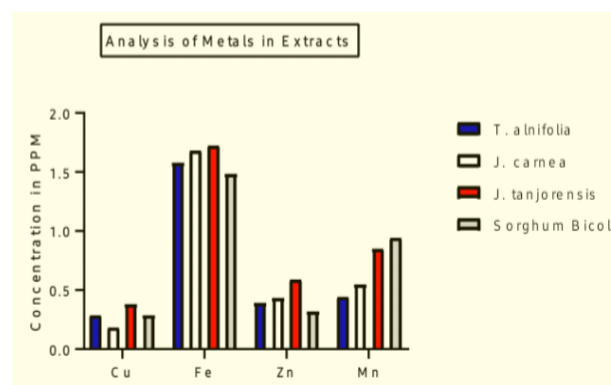


**Figure 1: Histogram Showing Results of Elemental Analysis**



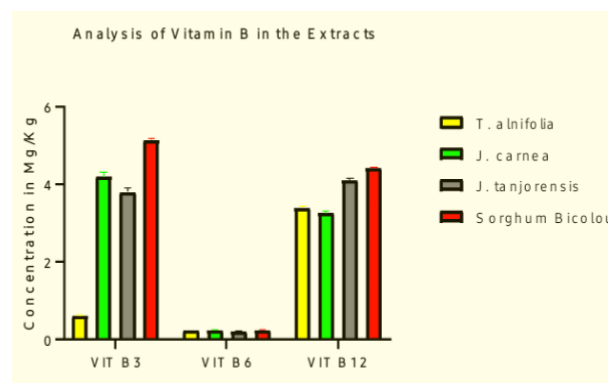
**Table 3. Results of Vitamin B Analysis**

| Parameters            | Sample A | Sample B | Sample C | Sample D | Feroglobin syrup |
|-----------------------|----------|----------|----------|----------|------------------|
| VITAMIN B3 (mg/100g)  | 0.5857   | 0.5124   | 0.4636   | 0.6283*  | 10.00            |
| VITAMIN B6 (mg/100g)  | 0.2320   | 0.2490   | 0.2100   | 0.2360   | 1.00             |
| VITAMIN B12 (mg/100g) | 3.3985   | 3.2620   | 4.0905   | 4.4255   | 0.10             |

**Note:** A= *Tetracera alnifolia*, SAMPLE B= *Justicia carnea*, SAMPLE C= *Jathropa tanjorensis*, SAMPLE D= *Sorghum bicolor*,  $P < 0.05$

The results showed that all the plants had the required vitamins for blood building. While Feroglobin had higher concentrations of Vitamins B3 and B6. It had an extremely low concentration of Vitamin B12. Vitamin B12 plays a vital role in the blood building process. A comparison of the concentrations of the vitamins contained in the plants' extracts is displayed in Figure 2.

The breakdown of the comparison showed that *S. bicolor* had high concentrations of all the vitamins than the other plants. A similar study on the aqueous extract of *J. carnea* leaves, revealed that the leaves are rich in vitamins with vitamin C, B2, and B1 as the most abundant vitamins. The researchers observed that vitamins B6 and B12 were the least available vitamins in comparison with the other vitamins. While this similarity in their findings is observed with respect to vitamin B6, vitamin B12 is the most abundant vitamin in comparison to the other vitamins analysed in this present study which employed the use of organic extracts [21].

**Figure 2: Histogram Showing Results of Vitamin B Analysis**

The GC-MS identified compounds in the plant extracts are presented in Table 4. The compounds identified were unconnected with the blood building activities of these plants.

**Table 4: Gas Chromatography-Mass Spectroscopy Result**

|          | Library ID                        | Area percentage | Retention time |
|----------|-----------------------------------|-----------------|----------------|
| Sample A | • Diaziridine-3-ethyl-3-methyl-   | 12.9412         | 13.6865        |
|          | • 1-Octene                        | 7.9047          | 17.9455        |
|          | • 6-methyl-                       | 7.90            | 17.924         |
| Sample B | • 2-Hexene-6-Nitro-               | 18.3738         | 23.8139        |
|          | • Oxirane-2,2-(1,4butandiyl) bis- | 4.2892          | 38.247         |
| Sample C | • Allylmethallyl Ether            | 3.9888          | 17.9427        |
| Sample D | • 1,8-Nonadiene-2,8-dimethyl-     | 14.4109         | 5.1520         |
|          | • Oxalic acid, cyclobutylundecyl  | 15.7077         | 13.7174        |

A previous study has documented the presence of Cyclononene, in the GC-MS analysis of the sorghum seed oil, which is a close derivative of 1,8-Nonadiene-2,8-dimethyl- observed in sorghum bicolor extract analysed in this present study [48].

### Discussion

#### Results from Phytochemical tests

From the phytochemical test conducted, it was observed that the important bioactive classes of compounds were present in these plants' extracts. Again, alkaloids, tannins, saponins, steroids,

triterpenoids, carbohydrates and proteins were detected in all the plants. The absence of cardiac glycosides was observed in *J. tanjorensis*, which was the only plant that had the presence of flavonoids detected.

#### Results from Elemental Analysis

The results from Elemental Analysis showed that all four samples had high iron content, However, *Jathropa tanjorensis* has the most favourable haemopoietic parameter qualities when compared to the other samples, Except for Manganese where



*Sorghum bicolor* had a higher value of 0.944ppm as against 0.844ppm of *Jathropha tanjorensis*.

#### Results from Vitamin B3, B6, B12 Analysis

Based on the results from the vitamin B analysis, it was observed that *Sorghum bicolor* had the highest concentration of all vitamins; including vitamin B12 which is necessary and directly involved in the formation of red blood cells. The plant also recorded a statistically significant  $P < 0.05$  value in vitamin B3 in comparison to the other plants.

#### Comparison of Sample Parameters with a Known Blood Tonic

The Parameter values obtained from the samples were compared to a known anti-anaemic Orthodox medication- Feroglobin syrup which contains; Iron 20mg, Cyanocobalamin-10microgram, zinc-6mg, manganese-0.5mg, copper-0.4mg, vitamin B6-2mg, vitamin B3-20mg. The values were converted to parts per million and it was observed that although Feroglobin syrup had a higher concentration of the elements and Vitamin B3 and B6; It had very minute amount of Cyanocobalamin (B12) when compared with the plants, which was statistically significant at  $p < 0.001$ . This implies that the plants have enhanced blood building potentials as Vitamin B12 is a major contributor to blood formation and is essential for the stimulation of red blood cells growth.

#### GC-MS Results

Based on the GC-MS results, some nonpolar compounds were detected in the samples. However, none of the compounds were found to have any contribution to anti-anaemic effect of the plant samples.

#### Statistical Analysis of Results

A statistical analysis was done on the results of the Elemental and Vitamin B analysis and it was observed that for the Elemental Analysis, all the plants extracts showed no statistical difference, while for the vitamin B analysis, only vitamin B3 showed statistical relevance at  $p < 0.05$  as B6 and B12 had no difference in  $p$  values. In comparison with Feroglobin syrup. It was observed that the plants extracts had statistically significant difference in the amount of Vitamin B12 at  $P < 0.001$  than the orthodox blood tonic.

#### Conclusion

From the experiment carried out, it can be concluded that *Sorghum bicolor* is the best ethnomedicinal remedy when compared to the other samples; this is due to its high iron, copper, and zinc content as well as its high concentration of Vitamin B12. However, from the comparison with feroglobin, it could be stated that Feroglobin has far less Cyanocobalamin (B12), which is essential for the stimulation of red blood cells growth than the plants, which further supports the anti-anaemic activities of these plants.

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