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# Comparative Studies of Therapeutic Effect of Leaves, Stem Bark and Root Bark Extracts of Afzelia africana (Smith) in Mice Challenged with Trypanosoma brucei brucei

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## Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

#### Article Information

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## ABSTRACT

**Background:** Traditional and complimentary health care is inarguably the system most close to homes, accessible and affordable. It is also culturally acceptable and trusted by large numbers of people. The affordability of most traditional medicines makes them all the more attractive at a time of soaring health-care costs, neglect of orphaned/non profitable diseases and

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nearly universal austerity.

**Aim:** Aqueous leaf, stem bark and root bark extracts were evaluated for their anti trypanosomal effect in experimental trypanosomiasis with a view to come up with a phytomedicine that is efficacious, available, accessible and non-toxic to both humans and animals.

**Study Design:** Complete randomized clinical trial design was used in the experiment. **Methodology:** Ninety five (95) mice were grouped into three (I, II, III) of thirty mice each (with sub groups A, B, C, D, E, and F consisting of five mice each) to which the leaf, stem bark and root bark extracts were administered at a dose of 100, 200, 300, 400 mg/Kgbw, while the remaining five mice served as the control for all the groups.

**Results:** The aqueous leaves extract at doses of 100, 200 and 300 mg/Kg bw portrayed very low activities except for the 400 mg/Kg bw that displayed a sustained Trypanostatic effect. The aqueous stem bark extract, at doses of 100 and 200 mg/Kg bw portrayed trypanostatic effect while doses of 300 and 400 mg/Kg bw effectively cleared the parasites from circulation on the 13<sup>th</sup> and 17<sup>th</sup> days into the treatment respectively. Three and two of treated mice survived and remained apparasitaemic for up to 120 days and beyond in the group treated with 300 and 400 mg/Kg bw respectively. In the group treated with the root bark extract, the mice on a dose of 100 mg/Kg bw died some few days into the experiment (6<sup>th</sup> day) while the dose of 200 mg/Kgbw sustained the animals until the 19<sup>th</sup> day. Doses of 300 and 400 mg/Kg bw were observed to clear the parasites in circulation after sustained administration for 23 and 16 days respectively.

**Conclusion:** This study has demonstrated the potency of the stem bark and root bark crude extracts of *Afzelia africana* in treating experimental trypanosomiasis and can thus be further purified and packaged as phytomedicine against this dreaded but neglected disease.

Keywords: Trypanosomiasis; trypanosomes; Afzelia africana; phytomedicine; chemotherapy.

#### 1. INTRODUCTION

Trypanosomiasis is one of the most important serious diseases of livestock and humans worldwide which continue to cause morbidity and mortality on a large scale in domestic/wild animals and human beings in sub sahara Africa and and South America [1,2]. The etiologic agents of the disease are flagellated protozoa that belong to the genus Trypanosoma: Trypanosoma brucei rhodesiense and T. b. gambiense cause human sleeping sickness and T. b. brucei (which is morphologically and biochemically indistinguishable from the two other subspecies), T. congolense and T. vivax cause nagana in livestock (cattle, sheep and goats) [3]. The parasites are transmitted between vertebrate hosts by the tsetse fly (Glossina spp.) [4,5]. Two other important livestock trypanosome Trypanosoma species are evansi and Trypanosoma equiperdum causing surra and dourine which are transmitted by biting flies or during coitus, respectively [6]. Sleeping sickness is again endemic in over 30 African countries threatening over 60 million people and has reached epidemic proportions in some countries, such as Angola, southern Sudan, Uganda and the Democratic Republic of Congo. Countries with high level of endemicity include Cameroun, Congo, Cote D'ivoire, Central African Republic, Guinea, Mozambique, Tanzania, and Chad.

Because of poor epidemiological information in Burundi, Botswana, Ethiopia, Liberia, Namibia, Rwanda, Senegal and Sierra Leon the situation there is poorly understood [7,8]. Almost 45000 cases of Human African Trpanosomiasis (HAT) were reported in 1999 but the World Health Organization estimates that the actual number of cases is between 300 000 and 500 000, since only 3–4 million people at risk of infection are under surveillance with regular examination or access to health centers [9,10].

The significance of trypanosomiasis to human health, nutrition and economy as noted by [11enormous. thereby necessitating 131 is continuous research for better ways of eliminating the disease. Unfortunately, the scarcity of compounds, the high incidence of side effects, and the emergence of resistance strains rendered existing chemotherapy, have inadequate [14,15]. Therefore, there is need to explore other agents, especially of plant origin for new generations of anti-trypanocidal agents that are more effective, less toxic, and readily available at cheaper prices [16].

It is estimated that up to 40% of all pharmaceuticals in industrialised countries are derived from natural sources. In the USA about 2% of prescriptions written by healthcare providers are for drugs that have natural ingredients, are synthetic copies or have artificially modified forms of natural chemicals [17]. The search continues for more therapeutically active plant-sourced materials, not always to the satisfaction of host communities [18].

Afzelia Africana (fabaceae/leguminosea) is one of the major and most widely distributed species in Africa. It is found in Senegal in West Africa to the Sudan in the North, Uganda and Tanzania in Eastern Africa. It is also found in South Asia e.g. India and is grown occasionally in other tropical countries as an ornamental plants. The plant species can grow into a large tree up to 20-30 m in height, with a spreading and open crown and large branches. The bark is dark grey, fissured and layered and peels off to reveal pale gray patches with granular pink brown slash. The leaves are alternate, peripinnate and up to 30 cm long. There are 3-8 pairs of shiny black leaflets widely spaced on the ranches. The fragrance white flowers often have purple markings and consist of 3 ecliptic upper petals, 10-12 cm long and lower petals with two divergent round lobes. The fruit is flattened, straight woody pod 10-15 cm long, 6-8 cm wide and 2-5 cm thick, each pod contains 7-10 seeds [19,20]. Roots, bark, leaves and fruits are used in traditional medicine. Root decoctions or macerations are used to treat stomach complaints, convulsions, trypanosomiasis and hernia, and as antidote. Root powder is applied externally to treat rheumatism. The roots have also been used in mixtures to prepare arrow poison. Bark decoctions and macerations are administered in the treatment of constipation, fever, vomiting, oedema, tachycardia, hypertension, bronchitis, lung complaints and bleedings during pregnancy, and as anodyne, diuretic, galactagogue and aphrodisiac. Bark ash is applied externally to treat lumbago and bark powder to wounds and swellings. The bark is also used as fish poison. Leaf decoctions and macerations are taken or externally against dysmenorrhoea, applied epilepsy, oedema, migraine, stomach-ache, asthenia, trypanosomiasis and as anodyne. Fruit preparations are taken to treat lung complaints and as aphrodisiac. Fruit ash is applied against leprosy, and as soap substitute. Twigs are used as chewing sticks [21,22], Previous studies have reported the plant to exhibit anti-inflammatory and analgesic bioactivities [23]. Powdered root of the plant mixed with millet beer has been used as treatment for hernia among some tribes in Cote d'Ivoire [24,25]. Most importantly, cattle pastoralists believe that the herd having

occasional inclusion of *A. africana* foliage while foraging tend to be more immune to Tse-tse fly infection than those not having access to it (Conducted field ethnobotanical survey on this study).

According to the World Health Organization, one third of the global population has no regular access to essential modern medicines, and in parts of Africa, Asia and Latin America, about half of the population faces shortage of minimum healthcare [26]. Studies on public health in the developing world repeatedly point to inadequacies in health care financing by the states which has led to a situation of highly limited material and human resources for healthcare services [27]. WHO, [28] cites the density of physicians of modern medicine per 100,000 persons in various countries as on year 2010-11 as: Rwanda 1.9, Ethiopia 2.9, Uganda 4.7. Benin 5.8. India 51.3 and China 164.2. This is in contrast to countries such as Australia and the USA where the figures are 249.1 and 548.9 respectively. This reveals the glaring inequities in health care delivery in developing countries. [29].

This research work therefore aims to comparatively evaluate the therapeutic potentials *(In vivo)* of the aqueous roots, stem and leaf extract of *Afzelia africana* in experimental trypanosomiasis in order to scientifically validate the traditional claim on this plant as a protection against trypanosomal infection.

## 2. MATERIALS AND METHODS

#### 2.1 Background on the Choice of Sample

The choice of the sample is an outcome of ethnopharmacological survey conducted in three villages of Koro, Awuru and Guffanti in Borgu Local Government Area of Niger State, Nigeria, which has large concentration of Fulani herdsmen with abundance flora. The survey was in form of personal contact with purposive approach (taking into cognisance the high level of illiteracy among the target subjects)

## 2.2 Plant Collection Identification and Preparation

The leaves, Stembark, and Root samples of the plant were obtained in the month of March, 2015, from the plantation of the Federal College of Wildlife Management, New Bussa, Niger State, North Central Nigeria. The sample was identified Garba et al.; BJMMR, 13(5): 1-14, 2016; Article no.BJMMR.23033

by the taxonomist in the Department of Wildlife and Ecotourism of the Institution and a specimen voucher number of WL/2213 was deposited. The samples (i.e. the leaf, stem bark and root bark) were harvested and dried seperately at room temperature. were pulverised They into powdered form using pestle and mortar and sieved as described by Onyeyili et al. [30]. Fifty (50 g) of each of the powdered sample was placed in a big conical flask and 400 ml of distilled water was added and allowed to stand in the laboratory for 24 hours with intermittent shakings at 4 hours interval. Thereafter, the extract was filtered (in each case) using a clean Muslin cloth. The filtrate was stored while the Marc was resuspended in another 350 ml of distilled water for another 24 hours and finally filtered. Steam bath was used to evaporate the water molecules in order to obtain the extract. The extract was then transferred into a sterile universal bottle and stored at 4°C until required for use. Percentage yields of 4.6, 9.4 and 7.6 g Kg<sup>1</sup> was obtained for the leaf, stem bark and the root bark respectively.

## 2.3 Choice of Strain (breed) of the Test Organism

The real strain of choice would have been *Trypanosoma brucei gambiense* which affect both Man and Livestock. Nevertheless this strain produces low parasitemia in small laboratory animals except in Hamster and the latter is not available in Nigeria [31]. Alternatively, a stabilate of pleomorphic *Trypanosome brucei brucei* strain 8/18 was obtained from the laboratory of the Malaria and Trypanosomiasis Research Unit of the Federal University of Technology, Minna, Nigeria and maintained in our laboratory by serial passage into the healthy mice.

## **2.4 Experimental Animals**

Albino mice were purchased from the Department Pharmacology, Ahmadu Bello University, Zaria, Nigeria. The mice were subsequently bred and multiplied in the Biochemistry/Nutrition laboratory of Animal Production Technology Department in the Federal College of Wildlife Management, New Bussa, Nigeria and used in all the subsequent experiment. The experiment were conducted in strict compliance with the internationally accepted principle for laboratory animal use and care as contained in the Canadian council on animals care [32] as reported by [33,34].

# 2.5 Antitrypanosomal Activity of the Leaf, Stem and Root Bark Extracts

In order to determine the activities of the extracts, ninety five (95) mice of mixed sexes and average weight of 38.6 g were grouped into three (i.e. GROUP I, II and III) for the Leaf, Stem and Root extracts respectively. Thirty (30) mice of mixed sexes were randomly allotted to each group while the remaining five mice were used as control (infected not treated INT) for all the test goups (I, II and III). Each group was further sub grouped into GROUPS (1A. 1B. 1C. 1D. 1E and 1F), (IIA, IIB, IIC, IID, IIE and IIF) and (IIIA, IIIB, IIIC, IIID, IIIE and IIIF) each containing five (5) mice. To each subgroup, the mice were intraperitoneally administered the extract at doses of 100, 200, 300 and 400 mgKg<sup>-1</sup>bw (i.e. A-D). Subgroups E – F in each case were treated with the extract but not infected (TNI) and not infected but administered Dimethylsulfoxide (DMSO) as placebo respectively.

# 2.6 Blood and Cerebrospinal fluid Infectivity Test

# 2.6.1 Blood infectivity test

One of the surviving mouse after treatment in each group was sacrificed four (4) weeks post treatment and 0.02ml of blood sample was drawn from the heart of the mice. It was sub-inoculated into two (2) clean parasite-free mice and the parasitaemia was monitored every 48 hours over 4 weeks period. A rapid "matching" method for estimating the host's parasitaemia was carried out as described by Herbert and Lumsden [35].

## 2.6.2 Cerebrospinal fluid infectivity test

Inoculation of clean parasite-free mice with CSF obtained from second surviving mouse was done as described by Nok, et al. [36] and improved upon by Garba et al. [33] and Ogbadoyi, et al. [37]. Briefly, the hair on the back of the mouse was shaved and it was positioned such that its head touches the hind limb. This positioning is necessary in order to make the vertebrae conspicuous. The lumber was the punctured by the insertion of a clean needle to obtain the clean clear and transparent fluid (CSF) that gushed into the needle (this was done with anaesthesia). Thereafter, two clean parasite- free mice were each sub- inoculated with 0.02 ml of the CSF and the parasitaemia was monitored every 48 hours for 4 weeks.

#### 2.7 Haematocrit Determination

This was done on two days interval and small volume of blood was collected from the tail (presterilised with methylated spirit) of the experimental animals into a heparinised capillary tube, one end of which was sealed with plasticine and then spun for 5 min in a Micro-haematocrit centrifuge (Hawksley & Sons Ltd, UK). The packed cell volume (PCV) was determined with the aid of Hawksley Micro haematocrit reader which gave reading in percentage.

#### 2.8 Prophylactic Activity Test

The test for prophylactic activity was done as described by Garba, et al. [33] and O'Neil et al. [38]. Five mice were each treated with the highest dose of the extract (400 mg/kg body weight) for five consecutive days before being infected with  $1 \times 10^6$  trypanosomes cells in 0.2mls inoculum. They were then routinely monitored for establishment of parasites.

## 2.9 Phytochemical Screening

The crude extract used (obtained by using water as solvent) was screened for the presence of tannins, saponins, alkaloids, phlobatanins, cardiac glycosides etc. as described by Tiwari et al. [39] using simple chemical tests.

## 2.10 Total Phenolic Contents (TPC)

TPC of *Afzelia africana* were determined by the Folin-Ciocalteu colorimetric method using gallic acid as a standard, and the absorbance was measured at 765 nm in a spectrophotometer

(HITACHI. Model: U-1100 573伊415). Results were expressed as gallic acid equivalent (GAE) mg/g of dried extract.

# 3. RESULTS

## 3.1 Trypanocidal Activity of the Aqueous Extracts

The aqueous leaves extract at doses of 100, 200 and 300 mg/Kgbw portrayed very low trypanocidal activities as the animals died on the  $6^{\text{th}}$ ,  $7^{\text{th}}$ , and  $13^{\text{th}}$  days into the experiment, except for the 400 mg/Kgbw that displayed a sustained Trypanostatic effect. In the case of the aqueous stem bark extract, doses of 100 and 200 mg/Kg bw portrayed trypanostatic effect with the parasitaemia in 200 mg/Kg bw being lower compared to the latter, while doses of 300 and 400 mg/Kg bw effectively cleared the parasites from circulation on the 13<sup>th</sup> and 17<sup>th</sup> days into the treatment respectively. Three and two of treated mice survived and remained apparasitaemic for up to 120 days and beyond in the group treated with 300 and 400 mg/kg bw respectively, while the remaining two and three in each (though apparasitaemic) died before then,. In the group treated with the root bark extract, the mice on a dose of 100 mg/Kg bw died some few days into the experiment  $(6^{th} day)$  while the dose of 200 mg/Kgbw sustained the animals until the 19th day with relatively low level of parasitaemia compared to the control and the 100 mg dose. Doses of 300 and 400 mg/Kg bw were observed to clear the parasites in circulation after sustained administration for 23 and 16 days respectively (Figs. 1a-c).



Fig. 1a. Trypanocidal activity of various doses of the leaf extract



Fig. 1b. Trypanocidal activity of various doses of stem bark extract



Fig. 1c. Trypanocidal activity of various doses of the root extracts

## 3.2 Packed Cell Volume

The result obtained for percentage PCV revealed a drop during the first seven days of the treatment but this was reversed in the subsequent days, except in the negative control group (Fig. 2).

#### 3.3 Blood and CSF infectivity test

The blood and the CSF drawn from the cured mice and inoculated into the healthy mice did not induce/cause the development of infection six weeks after the sub inoculation.

## 3.4 Prophylactic Activity of Extract

The animals administered the effective doses of 300 and 400 mg/kg body weight in groups II and III (i.e. stem bark and root bark extracts) for five consecutive days prior to infection were observed to develop infection 72 h post infection (Fig. 3). This indicated the inability of the extract to protect mice against infection.

## 3.5 Phytochemical Compositions

The phytochemical analysis (qualitative) carried out revealed the presence of some phytochemicals (Tables 1 and 2). Garba et al.; BJMMR, 13(5): 1-14, 2016; Article no.BJMMR.23033



Fig. 2. Mean PCV in group of mice treated with various doses of the leaf, stembark and root extracts and standard drug (Beneril)

# 4. DISCUSSION

The result obtained from this study have demonstrated the ability of leaf extract at a dose of 400 mg/kg to sustain the experimental animals up to the  $40^{th}$  day despite the presence of the parasite in the blood (though at a low level). This clearly portrays the possible trypanostatic activity of the extract. A plant part showing such a trend may possibly when further purified exhibit trypanocidal effect. The stem bark extract appears to display even more promising antitrypanosomal effect, as doses of 100 and 200 mg/kgbw were able to sustain the experimental animals beyond the experimental period to the 40<sup>th</sup> day and beyond while doses of 300 and 400 mg/kg interestingly cleared the parasites in the experimental animals on the 13<sup>th</sup> and 17<sup>th</sup> days into the experiment respectictively. Atawodi, [13] has also observed a drastic decrease (in vitro) in the activities of Trypanosoma brucei treated with aqueous stem bark extract of A. Africana, while O'Neil et al. [38] reported that the aqueous extract of A. aficana possesses bark antihyperglycemic properties. In addition, the extract could prevent various compications of diabetes as well as improving some haematological parameters. The root bark extract were interestingly observed to clear the parasites from circulation at the doses of 300 and 400 mg/kg with clearance achieved earlier in the former than latter. This is exciting due to the fact that in the elaborative in vitro screening of variety

of savannah plants carried out by Atawodi, [13], Tiwari, et al. [39], Oyedemi et al. [40], Atawodi et al. [41], and Hopp et al. [42], only the leaf and the stem bark extracts of *Afzelia africana* were tested, whereas the potency of the root extract has not (for unknown reason) been tested for its trypanocidal effect.

It is difficult to speculate the mechanism by which these extracts exhibit their antitrypanosomal activity since the active ingredient(s) were not isolated. However, accumulated evidence [43,44] suggest that many natural products exhibit their antitrypanosomal activity by virtue of their interference with the redox balance of the parasites acting either on the respiratory chain or on the cellular defenses against oxidative stress. This is because natural products possess structures capable of generating radicals that may cause peroxidative damage to trypanothione reductase that is very sensitive to alterations in redox balance. It is also known that some agents act by binding with the kinetoplast DNA of the parasite. The antitrypanosomal principles of the plant tested in this study is unknown, until further studies are carried out.

Although, our investigation did not involve structural elucidation, literature search revealed that extracts showing potent trypanocidal activity, have also been reported by Wilkinson and Kelly [45] to contain either alkaloids, flavonoids, phenolics and/or terpenes. Sara et al. [46],

reported the presence of similar phytochemical compounds obtained in our study in the crude extract of A. africana and these include tannins, flavonoids, steroids, alkaloids and saponins. These phytochemical compounds are known to be biologically active and thus aid the antimicrobial activities of Α. africana. Phytochemicals exert antimicrobial activity through different mechanisms; tannins for example, act by iron deprivation, hydrogen bounding or specific interactions with vital proteins such as enzymes [47,48] in microbial cells. Herbs that have tannins as their component are astringent in nature and are used for treating intestinal disorders such as diarrhoea and dysentery [49], thus exhibiting antimicrobial activity. The presence of tannins in A. aficana supports the traditional medicinal use of this plant in the treatment of different ailments [47,50]. AbdulHamid et al. [51] and Amani et al. [52] revealed the importance of tannins for the treatment of inflamed or ulcerated tissues. Brian, et al. [53] and Li et al. [54] reviewed the biological activities of tannins and observed that tannins (whether total or pure compound) have remarkable activity in cancer prevention and as anticancer agent [55]. This implies that *A. aficana* can serve as a source of drug for the treatment and prevention of cancer. In addition to its antimicrobial and anticancer activities, tannins have roles such as stable and potent antioxidants [47,54].

Alkaloid is another phytochemical compound observed in the stem bark extract of *A. aficana*. Alkaloids have been associated with medicinal uses for centuries and other possible roles have not been examined. One of the most common



Fig. 3. Prophylactic activity of various effective doses of stem and root barks extracts

S/no	Test	Leaf extract	Stem bark extract	Root bark extract
1	Phlobatannin	+	+	+
2	Flavonoid	+	+	-
3	Cyanoglycoside	-	+	+
4	Tannin	+	+	+
5	Sapponin	+	+	+
6	Steroid	+	+	+
7	Alkaloid	+	+	+
8	Phenol	+	+	+

Table T. FIIVIUCHEIIIICai CUIIDUSIIIUII UI IEAI. SIEIII DAIK AHU TUUI EKITACI UI AIZEIIA AIIICA	Table 1. Phytochemical composition of leaf, stem bark and root	extract of Afzelia	Africana
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KEYS: + = Present, - = Absent

biological properties of alkaloids is their toxicity against cells of foreign organisms. These activities have been widely studied for their potential use in the elimination and reduction of human cancer cell lines [56]. In addition, anti-inflammatory, alkaloids possess antiasthmatic, and anti- anaphylactic properties with consequences of altered immunological status in vivo [57-60]. Furthermore, alkaloid, which is one of the largest groups of phytochemicals in plants, has amazing effect on humans and this has led to the development of powerful pain killer medications [61]. Flavonoids, which are also one of the constituents of A. aficana stem bark extract, exhibit antimicrobial, anti-inflammatory, anti-angionic, analgesic, anti-allergic, cytostatic and antioxidant properties [62,47,63]. The ability of flavonoids to scavenge hydroxyl radicals, superoxide anion radicals and lipid peroxyradicals highlights some of their healthpromoting functions in organisms, which are important for prevention of diseases associated with oxidative damage of membrane, proteins and DNA [64,47]. Flavonoids in the human diet may reduce the risk of various cancers, as well as preventing menopausal symptoms [65,63]. All these facts support the usefulness of A. aficana in folklore remedies and is one of the reasons this plant is widely used for the treatment of many diseases among many tribes in Africa. A. aficana exhibited antimicrobial properties and flavonoids act in this way. In addition to the antimicrobial activities exhibited by flavonoids, thev also exhibit antitrypanosomal and antileishmanial activities [66]. Epidermiological studies suggest that the consumption of flavonoids is effective in lowering the risk of coronary heart diseases [67,68]. All these facts suggest that A. aficana can as well be used to treat coronary heart disease. Furthermore, several flavonoids exhibit antiviral activities [69]. Lastly, saponins, which are responsible for numerous pharmacological properties [68], also tested positive in A. aficana stem bark extract. Saponins are considered key ingredients in traditional Chinese medicine and are responsible for most of the observed biological effects in medicinal plants [70,71]. Saponins are known to produce an inhibitory effect on inflammation [72] [73-75]. These observations cited on phytochemical compounds support our findings on the usefulness of A. aficana in traditional medicament. Therapeutic effects of some medicinal plants commonly used in folklore remedies can therefore be attributed to the antioxidant and antimicrobial properties of their phytoconstituent.

Table 2. Total phenol	ic content of leaf, stem
bark and root bark ext	racts of Afzelia africana

Extracts	Concentration (µg/mg GAEq)			
Leaf	55			
Stem bark	105			
Root bark	85			
CAEq: Callia agid aquivalant				

GAEq: Gallic acid equivalent

Anaemia is one of the established major pathological features of African trypanosomiasis [34,36]. Therefore, the presence and severity of anaemia are good indicators of disease status. Control of anaemia is an integral part of disease management and so, any drug used for the treatment of African trypanosomiasis would have an added advantage if it is also effective in controlling the associated anaemia [33,34]. It is apparent from Fig. 2 that administration of the extract led to remarkable reduction in the severity of anaemia initially observed (except in 400 and 200 mg/kg bw of the leaf and root bark extracts) as evidence from the significant difference (P = .05) in the levels of the PCV of the treated animals and those which were not treated. This further enhances the efficacy of the extract and it is a big boost to the potentials of the effective extracts as an anti-sleeping sickness agents. Injuries sustained by red blood cell (RBC) membranes caused by the flagella and microtubule reinforced body of the organisms erythrophagocyosis greatly enhanced of damaged RBC by the MPS [76].

Traditional and Complimentary Medicine (T & CM) is widely used around the world and valued for a number of reasons. At the International Conference on Traditional Medicine for South-East Asian Countries in February 2013, the WHO Director-General, Dr Margaret Chan, stated that "traditional medicines, of proven quality, safety, and efficacy, contribute to the goal of ensuring that all people have access to care. For many millions of people, herbal medicines, traditional treatments, and traditional practitioners are the main source of health care, and sometimes the only source of care. This is care that is close to homes, accessible and affordable. It is also culturally acceptable and trusted by large numbers of people. The affordability of most traditional medicines makes them all the more attractive at a time of soaring health-care costs and nearly universal austerity. Traditional medicine also stands out as a way of coping with the relentless rise of chronic non-communicable diseases." Despite the prevailing arguments and counter arguments on the prospects and seeming constraints on the adoption of T&CM [77], there is little doubt that interest has grown, and will almost certainly continue to grow, around the world, so long, the drugs against some tropical "orphaned diseases" such as Trypanosomiasis continue to remain unavailable, inaccessible and expensive.

# 5. CONCLUSION

Conclusively, the result of this study taken together reveals that the aqueous leaf extract at dose of 400 mg/kg a higher bw has trypanostatic effect while lower doses of 100 and 200 mg/kg bw of the stembark extract also has similar effect. The Stem bark and Root bark extract at doses of 300 and 400 mg/kg bw were observed to have trypanocidal effect with the former having lower clearance period than the latter. The observed erythrolytic effect exhibited by doses of 200 and 400 mg/kg bw of the leaf and Root extract might probably be due high Saponin content. The plant can therefore be exploited to obtained alternative trypanocidals that are cheap, readily available, and accessible.

# CONSENT

It is not applicable.

## ETHICAL APPROVAL

All authors 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.

# **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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