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IN VITRO ANTIBACTERIAL, ANTIOXIDANT, CYTOGENOTOXIC AND NUTRITIONAL VALUE OF *ALOE BARBADENSIS* MILL. (ASPHODELACEAE)

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ABSTRACT

This study evaluated the *in vitro* antibacterial, antioxidant, nutritional and cytogenotoxic potentials of *Aloe barbadensis* (AB) root extract. The antimicrobial activity of AB was determined using agar well diffusion and microdilution techniques while the nutritional analyses were done using standard procedures. The antioxidant activities were evaluated via the lipid peroxidation, 1, 1, diphenyl-2-picrlhydrazyl (DPPH) and ferric reducing power (FRAP) assays. The cytogenotoxic effects were assessed using the *Allium cepa* and *Sorghum bicolor* assays. Five test bacterial isolates: *Escherichia coli, Klebsiella pneumoniae, Proteus mirabilis, Pseudomonas aeruginosa and Staphylococcus saprophyticus)* were found susceptible to the ethanolic extract of AB root. The extract at 50 mg/ml recorded the highest zone of inhibition (25.00±1.73 mm) against *S. saprophyticus*. The nutritional analysis gave a nitrogen free extract value of 24.66%, 30.05% crude fibre, 15.14 mg/kg of Na, 2.01 mg/kg of Mg, 37.42 mg/kg of Fe and 0.02 mg.kg of Zn. The fractions had antiradical activity in the lipid peroxidation, DPPH and FRAP assays. The ethanolic extract induced some chromosomal aberrations in the root tip cells of *Allium cepa*. The ethanolic extract had the highest (0.13±0.09 mm) antiproliferative activity at 20 mg/kg in the *Sorghum bicolor* assay. The relatively high nutrient content may be a possible mechanism for the aphrodisiac property of the plant. The results of the biological activities of AB root is an indication of its potential use in drug development for the management of infectious diseases and other related ailments.

Keywords: *Aloe barbadensis*, antibacterial, antioxidant, cytotoxicity, genotoxicity, microbial infections, phytochemicals

INTRODUCTION

Today medicinal plants are fast becoming an important key to unlock the ravaging effects of some diseases in the world. The World Health Organization (WHO) has supported traditional medicines, particularly in developing countries by promoting the incorporation of its useful elements into the national health care system (**Akerele, 1987**). This fortunately has led to the rising popularity of herbal medicine in developing countries (**Obici *et al*., 2008**). This affirms the belief that medicinal plants are nature's gift to mankind.

Aloe barbadensis (AB) of the family- Asphodelaceae has been reported ethnomedically to have several folks uses with some validated scientific information. Its ethnomedical use as an aphrodisiac was earlier reported by **Erhabor *et al*. (2013)** and this was validated *in vivo* using an animal model (**Erhabor and Idu, 2017**). Other previously reported biological activities of this plant include the use of its gel in wound healing (**Davis *et al*., 1994; Maenthaisong *et al*., 2007**), inflammatory problems (**Che *et al*., 1991; Robbers and Tylers, 1999**) as well as in moisturizing and anti-aging activities (**West and Zhu, 2003; Sahu *et al*., 2013**). The leaves have been used for treating fungal infections associated with superficial mycoses (**Shamim *et al*., 2004**). The antidiabetic property (**Tanaka *et al*., 2006; Yagi *et al*., 2006**) and its perceived reproductive functions in enhancing fertility (**Oyewopo *et al*., 2011; Iwu, 2014;**) have also been documented.

Some of the ethnomedical uses of *A. barbadensis* include its use in the treatment/management of intestinal ulcers (**Ross, 1999**), constipation, impotency, ringworm and eczema (**Adodo, 2012**). It is used as a purgative, appetite stimulant and emmenagogue, as well as in the treatment of colds, piles, asthma, cough and jaundice (**Joseph and Raj, 2010**). Interestingly, chemicals found in the plant have become known remedies for treating sexual disorders/reproductive diseases (**Yakubu *et al*., 2007; Joseph and Raj, 2010**), microbial infections (**Usman *et al*., 2005; Idu *et al*., 2011; Erhabor *et al*., 2013;; Mongalo *et al*., 2017; Motlhatlego *et al*., 2018;**), cancers (**Venables *et al*., 2016; Eboji *et al*., 2017**) and free radicals implicated diseases (**Otang *et al*., 2012; Sharma and Lall, 2014; Elisha *et al*., 2015; Motlhatlego *et al*., 2018**) as well as other relevant ailments/diseases. The medicinal value of plants lies in these chemical constituents or active ingredients and other nutrients for their actions (**Edeoga, 2000; Okigbo *et al*., 2008**). These plant chemicals have been reported to offer a boost of nutritional value thereby improving sexual performance and libido (**Yakubu *et al*., 2007; Sumalatha *et al*., 2010**). This enhancement in the general health can thus lead to a burst of energy which translates into increased sexual appetite, besides increasing blood flow and intensity of ejaculation accompanied by anabolic and growth hormone stimulating properties (**Smith *et al*., 2000**; **Ahmad *et al*., 2003**; **Yakubu *et al*., 2007**). These phytochemicals have also been reported to enhance erection and prolong ejaculatory latency in male albino rats (**Yakubu *et al*., 2005; Muanya and Odukoya, 2008; Sumalatha *et al*., 2010**). These bioactive substances from medicinal plants include saponins, alkaloids, flavonoids, tannins, sterols, protein, carbohydrates and several minerals (zinc, calcium, magnesium, potassium) (**Olowokudejo *et al*., 2008**). *A. barbadensis* following previous studies undertaken mainly on the leaves and gel has many of these bioactive phytochemical substances (**Ross, 1999; Joseph and Raj, 2010; Adodo, 2012; Sahu *et al*., 2013; Idu *et al*., 2014**). Among, these phytochemicals are those which may scavenge reactive oxygen species that have been implicated in damaging biological molecules (DNA, protein, carbohydrate and lipids), leading to cancer, infertility and other ailments (**Ashafa *et al*., 2010; Raghuveer and Vyas, 2010**). Despite the many merits of these phytochemicals, they are said to have a deleterious effect when not properly utilized or consumed. There is therefore a need to assess their safety levels either at the cell, tissue or organ levels.

In this study, our objective was to determine the antimicrobial, antioxidant, nutritional and phytochemical constituents of the root of *Aloe barbadensis* as a potential aphrodisiac. The cytotoxic and genotoxic effects of the plant were also investigated.

METHODS

Collection of plant sample

The roots of *A. barbadensis* were collected from Okene, Nigeria. It was authenticated by Mr. G. Ibhanesebhor of the Herbarium Unit of the Obafemi Awolowo University, Ile-Ife, Nigeria, with voucher number-IFE17004 where it was deposited.

Preparation of plant sample and extraction

AB roots were detached from the whole plant, rinsed in water, spread on laboratory tables and dried at room temperature. The roots were dried in an oven set at 40ºC for 10 minutes before being reduced to a fine powder using a grinder. Two kilograms (2kg) of the powdered plant material was extracted with ethanol (5 L) using a Soxhlet extractor. The extract was concentrated to dryness using a water bath (HH-S Water Bath; Searchtech Instruments) set at an average temperature of 50 ºC. The percentage yield of the ethanolic extract was determined using the formula (% yield= weight of extract/weight of powder material x 100/1).

Preparation of solvent fractions

Forty grams of the ethanolic extract was re-dissolved in 100 ml of 60 % methanol and partitioned exhaustively with chloroform (150 ml) in a separating funnel by decanting until a transparent layer was obtained. The lower layer-chloroform fraction (organic) was collected first and the upper layer- aqueous fraction (inorganic) was also collected. Water (70 ml) was added to the chloroform to remove compounds that entered the bottom layer due to agitation. The chloroform fraction was further concentrated to dryness using a water bath at a relatively low temperature. Ethyl acetate (300 ml) was added to the aqueous fraction and decanted appropriately. The ethyl acetate fraction, being less dense, was collected after decanting the aqueous fraction. Both fractions were then further concentrated using a water bath. The respective percentage yields of the fractions (chloroform, ethyl acetate and aqueous) were determined using the formula (% yield= weight of extracted fraction/weight of crude extract X 100/1).

Antimicrobial evaluation

Collection of semen samples

The semen samples were collected according to the method previously described by **Ekhaise and Richard (2008)**. The seminal fluid specimen was collected from five male patients attending a fertility clinic at two private hospitals in Benin City, Nigeria. The samples were collected from patients with secondary infertility using the masturbation method. The collected specimens were immediately transferred to the laboratory where they were further subjected to microbiological isolation and identification.

Isolation, Identification and Standardization of test organisms

The sperm specimens were streaked on both blood agar and nutrient agar. The plates were incubated at 37 ^{0}C for 24 hrs. After incubation, the bacterial species were identified via Gram staining and appropriate biochemical tests following standard procedures outlined by **Cheesbrough (2006)**. A loop full of stock culture of the organism was inoculated onto 5 ml of sterile Müller-Hinton agar and incubated for 24 hrs. Overnight culture of the organisms (0.2ml) was inoculated into 20 ml of sterile nutrient broth and incubated for 3-5 hrs. The turbidity of the culture was compared to that of a 0.5 Mac-Farland standard to standardize the culture to 10^6 cfu\ml.

Susceptibility Testing

According to the method described by **Emeruwa (1982)**, 0.5ml of the standardized culture was spread onto a sterile plate to achieve confluent growth. Fifteen milliliters of Müller-Hinton agar at 45 ^{0}C was added to each plate and the plates rocked for even spread and proper mixing of bacteria and agar. The content of the plates was allowed to solidify and wells approximately 6 mm in diameter were bored on the surfaces of the agar medium using a sterile cork borer and the bottom of the holes seeded with molten agar. The reconstituted extract (0.2 ml) at the test concentrations was dropped into the holes while an aqueous solution of the standard antibiotic (chloramphenicol) at the same concentrations as the extract was used as positive control. Sterile distilled water (0.2 ml) was used as negative control. The plates were allowed to stand for 30 mins for pre-diffusion of the extract to occur and then incubated at 37 ^{0}C for 24hrs and zones of inhibition were measured to the nearest cm using a meter rule. The final measurements were converted to mm. The mean of duplicate results was recorded.

Determination of Minimum Inhibitory Concentration (MIC)

Bacterial strains were cultured overnight at 37 ^{0}C on nutrient broth and were adjusted to a final density of 10^6 cfu\ml. The standardized bacterial inocula were used to inoculate the 96-well microtitre plates containing appropriate dilutions of the extract (12.5 - 0.10 mg/ml) under sterile conditions. The plates were incubated under aerobic condition at 37 ^{0}C and examined after 24 hrs. As an indicator of bacterial growth, 40 µl of 0.2 mg/ml p-iodonitrotetrazolium violet was added to each well and incubated for 30 mins at 37 ^{0}C. The colourless tetrazolium salt was reduced to a red coloured product by the biological activity of the organisms. Each treatment was performed in triplicate and complete suppression of growth at a specific concentration of extract as indicated by a clear solution was required for it to be recorded as active (**Eloff, 1998**). Chloramphenicol was used as positive control in the experiment with sample free solutions of 10% DMSO as the negative control.

Determination of Minimum Bactericidal Concentration (MBC)

The MBC was determined by removing a loopful of bacterial suspension from the MIC micro-titre plate that did not show any growth and sub -cultured into nutrient agar plates. The plates were incubated and the concentrations at which no visible growth was observed were recorded as MBC.

Antioxidant activity

Lipid peroxidation assay

This assay was carried out using a slightly modified method described by **Ohkawa *et al*. (1979)**. Liver homogenate was prepared from commercially available goat liver sourced from a local abattoir in Benin City, Nigeria. The liver was washed several times with ice-cold saline solution. Ten percent of the liver homogenate was prepared using ice-cold potassium chloride (KCl) (0.15M) in a blender. Lipid peroxidation was initiated in 1ml of tissue homogenate mixed with various concentrations of the ethanol extract (20, 40, 60, 80 and 100 µg/ml). This was followed by adding 0.1ml of ferric sulphate (2.5 mM), 0.1ml of ascorbate (100 mM) and 0.1ml of KH_3PO_4 (10 mM). The volume was made up to 3 ml with distilled water and incubated at 37 ^{0}C for one hour. One ml, each of 5 % trichloroacetic acid (TCA) and 0.6% thiobarbituric acid (TBA) was then added to the reaction mixture and the tubes boiled for 30 minutes in a water bath. The tubes were centrifuged at 3500 rpm for 10 minutes. The extent of inhibition of lipid peroxidation was evaluated by the estimation of thiobarbituric acid reactive substances (TBARS) level by measuring the absorbance at 532 nm. The lipid peroxidation inhibition percentage was calculated by using the formula below;

$$\text{Inhibition \%} = \frac{[A_{control} - A_{test}]}{A_{control}} \times 100$$

Where,
$A_{control}$ = Absorbance of control
A_{test} = Absorbance in the presence of the extracts
The experiments were repeated in triplicates. α- Tocopherol was used as standard antioxidant.

DPPH (2, 2-diphenyl-1-picryl hydroxyl) radical scavenging assay

The radical scavenging activity of the crude ethanol extract and fractions of *A. barbadensis* root against the DPPH radical (Sigma-Aldrich) was determined by a slightly modified method using UV (**Chidambara Murthy *et al*., 2002; Leong and Shui, 2002**). Concentrations of 20, 40, 60, 80 and 100 ug/ml of the crude extract and Vitamin C were prepared in methanol (Analytical grade). One ml of the extract was placed in a test tube, followed by 2 ml of 0.1mM DPPH in methanol. A control solution was prepared containing the same amount of DPPH and methanol. This procedure was similarly conducted to determine the DPPH scavenging activity of the various fractions (chloroform, ethyl acetate and aqueous). The radical scavenging activity was calculated using the following formula;

$$\% \text{ inhibition} = [A_b - A_a] / A_b \times 100$$

Where,
A_b = the absorbance of the blank sample
A_a = the absorbance of the extract and fractions

Ferric reducing power assay

The previously outlined method by **Kumar *et al.* (2005)** was used in assessing the ferric reducing power of the samples. This was determined by mixing various concentrations of the crude extract, fractions and standard ascorbic acid solutions (20, 40, 60, 80 and 100 µg/ml) in 1ml of methanol with phosphate buffer (2.5 ml, 0.2 M at pH 6.6) and potassium ferricyanide $K_3[Fe(CN)_6]$ (2.5 ml, 1%). The

mixture was incubated at 50 ^{0}C for 20 minutes. Then 2.5ml of 10 % trichloroacetic acid (TCA) was added to the mixture which was centrifuged at 3000 rpm for 10 minutes at room temperature. An aliquot of 2.5ml of the supernatant was mixed with 2.5 ml distilled water and ferric chloride ($FeCl_3$) (0.5ml, 0.1%) and the absorbance of the reaction mixture was measured at 700 nm to detect increased reducing power. All the tests were performed in triplicate.

Phytochemical and nutritional analysis

The qualitative phytochemical screening of the crude ethanol extract and fractions was carried out to determine the different metabolites (glycosides, cardiac glycosides, saponins, flavonoids, phenolic, tannins, phlobatannins, terpenoids, alkaloids, steroids, polysaccharides and reducing sugars) using the methods described by **Sofowora (1993), Trease and Evans (1996)**. The total phenolic and flavonoid content was determined using the method of **Lin (2007)** while the saponin content was done using the method of **Obadoni and Ochuko (2002); Ejikeme *et al*. (2014)**. The alkaloidal content was determined using the method of **Harborne *et al*. (1973)**. The proximate analysis (gross chemical composition) was determined by the recommended method of **Horwitz and Latimer (2000)**. The determined six food categories were moisture content, ash content, crude oil, crude fibre, crude protein and nitrogen free extract.
For the mineral element's composition, a weighed aliquot of 5 g of the powdered root was ashed at 550^{o}C in the Muffle furnace for 5 hours and the residue dissolved in 100 ml deionized water. Standard solutions of the minerals were prepared and used to calibrate the atomic absorption spectrophotometer (AAS) (model 969AA, Unicam Series) using acetylene-air flame at specific wavelengths. Aliquots of the ash solutions were injected into the AAS and from the standard curve the various concentrations were obtained.

Cytotoxic and genotoxic Study

***Allium cepa* assay**

The relatively same sized purple variety of onion bulbs were obtained commercially from a local market in Benin City, Edo State, Nigeria. The bulbs were stored under dry and well aerated condition for three weeks before use to prevent rot and enhance viability. The outer scaly leaves of the onion bulbs were carefully removed with hand while the dried roots were shaved off with a sharp razor blade to expose the fresh meristematic tissues. The bulbs were placed into freshly obtained distilled water to protect the primordial cell from drying off. Mouldy bulbs and those with shooting green leaves were discarded. The macroscopic evaluation (root growth inhibition) and microscopic analysis (induction of chromosomal aberrations) were subsequently done. Photomicrographs of the root tip cells were taken using an Olympus model microscope with a 5 mega pixel digital microscopic eyepiece camera**.** The standard procedures earlier described by **Rank and Nielsen (1994); Olorunfemi *et al*. (2011)** was adopted for this bioassay with the under listed parameters determined as displayed in the formulas below.

$$\text{Percentage(\%) root growth of control} = \frac{\text{overall mean root length of test solution}}{\text{overall mean root length of control}}\ x\ 100$$

Mitotic index was computed by determining the mitotic cell frequency at the tip as

$$\frac{\text{Number of dividing cells}}{\text{Total number of cells counted}}\ x\ 100$$

The mitotic inhibition was determined using the following formula:

$$\frac{\text{Mitotic index in control} - \text{Mitotic index in treatment}}{\text{Mitotic index in control}}\ x\ 100$$

Frequency of Chromosomal aberration:

$$\frac{\text{Number of Aberrant cells}}{\text{Total number of cells counted}}\ x\ 100$$

***Sorghum bicolor* assay**

The growth inhibitory effects of the ethanol extract of *A. barbadensis* was carried out following the method described by **Fajana (2013)**. Seeds of *Sorghum bicolor* (Guinea corn) were purchased locally in Edo State, Nigeria. A simple viability test was carried out by placing a handful of the seeds in distilled water. The viability of the seeds was determined by their ability to sink in water. Those that remained submerged in water were removed and air dried for use while those which floated were discarded. The viable seeds were sterilized with 96 % ethanol for 1 minute and then rinsed with distilled water. The, 10 ml different concentrations (5, 10, 15, 20, 25 and 30 mg/ml) of the ethanol extract containing 3.3% tween 80 in water were poured into 9 cm full Petri dishes lined with cotton wool and filter paper (Whatman No 1). Twenty (20) viable seeds were spread on each and incubated in the dark. The lengths (mm) of the radicles emerging from the seeds were taken at 24, 48, 72, and 96 hours. The control seeds were only treated with 3.3% Tween 80 in distilled water containing no extracts. The experiments were carried out in triplicates.

Statistical analysis

Data are shown as mean ± SEM and mean ± SD of the corresponding replicates. One Way ANOVA was done where applicable as well as a Duncan's multiple range test to analyse differences among different means and the interaction between the variables using SPSS 15.0 computer software package. Differences at $P<0.05$ or $P<0.01$ were considered statistically significant.

RESULTS

Percentage yield of extract and fractions

Two kilograms of the powdered root plant material yielded 124.65g (6.23%) of the ethanol extract. The fractionation of 40 g of the ethanol extract gave various yields of 10.96g (27.4%), 2.69g (6.73%) and 22.89g (57.23%) corresponding to the chloroform, ethyl acetate and aqueous fractions respectively (Tab1).

Table 1 Percentage extract/fractions yield of *A. barbadensis* root

Extract/ Fractions	Yield in (%) and (g)
Ethanolic extract	6.23% (124.65g)
Aqueous fraction	57.23% (22.89g)
Chloroform fraction	27.4% (10.96g)
Ethyl acetate fraction	6.73% (2.69g)

Antimicrobial effect of *A. barbadensis* root extract on test organisms

The susceptibility of the test organisms to the ethanolic extract of *A. barbadensis* root is shown in Tables 2 – 3. The tested concentrations of the extract of AB had a certain degree of inhibitory activity against the tested organisms with the highest zone of inhibition of 25.00±1.73 mm recorded against *Staphylococcus saprophyticus* at a concentration of 50 mg/ml (Tab 2). It was observed that the extract had a MIC of 6.25 mg/ml against *E. coli* and *S. saprophyticus* while the MIC was 12.5 mg/ml against *P. mirabilis, P. aeruginosa* and *K. pneumoniae*. The extract had the same MBC value of 12.5 mg/ml against the tested organisms (Tab 3). The positive control, chloramphenicol was active against all test organisms but significantly at the highest concentration of 50 mg/ml. The commercial antibiotic (chloramphenicol) was most active against *Staphylococcus saprophyticus* with an inhibitory diameter of 24 mm and least active against *E. coli* and *P. aeruginosa* with no measurable zone of inhibition (Tab 4).

Table 2 Effect of various concentrations of the ethanolic root extract of *Aloe barbadensis* on test organisms

Test organisms	Concentrations of Extract (mg/ml)		
	12.5	25	50
Escherichia coli	8.00±0.58	12.67±0.88	16.00±1.00
Klebsiella pneumoniae	4.67±0.88	6.67±0.88	15.00±2.00
Proteus mirabilis	3.00±0.58	4.33±0.88	12.00±1.00
Pseudomonas aeruginosa	4.33±0.67	8.00±0.58	14.67±1.45
Staphylococcus saprophyticu	9.70±0.88	13.33±1.20	25.00±1.73

Values are mean±SEM (zone of inhibition in mm); n = 3

Table 3 Antibacterial activity (MIC and MBC) of the ethanol extract of *A. barbadensis* root

Isolates	MIC (mg/ml)	MBC (mg/ml)
Escherichia coli	6.25	12.5
Klebsiella pneu	12.5	12.5
Prote	12.5	12.5
Pseuaomonas aeruginosa	12.5	12.5
Staphylococcus saprophyticus	6.25	12.5

Table 4 Antibiotic sensitivity of the test organisms with growth inhibitory zones

Organisms	Chloramphenicol		
	12.5 mg/ml	25 mg/ml	50 mg/ml
Escherichia coli	NMZI	NMZI	10.06±0.05
Klebsiella pneumoniae	4.00±0.04	10.02±0.07	18.00±0.03
Proteus mirabilis	NMZI	10.09±0.03	16.00±0.09
Pseudomonas aeruginosa	NMZI	NMZI	12.00±0.02
Staphylococcus saprophyticus	10.05±0.06	15.00±0.03	24.00±0.02

Legend: NMZI= No measurable zone of inhibition; n = 2; Values are mean±SEM (zone of inhibition in mm)

Antioxidant activities of *A. barbadensis* root extract

Lipid peroxide scavenging activity of *A. barbadensis* root extracts

The result of the lipid peroxide scavenging activity of the extract and fractions are presented in Figure 1. It was noticed that the ethanolic extract had a concentration-dependent activity. The chloroform fraction had a maximum inhibition of 48.28 ± 0.34%, followed by the crude ethanolic extract with 46.57 ± 0.69%, ethyl acetate had an inhibition zone of 30.58 ± 1.04%. The aqueous fraction had the least inhibition of 30.07 ± 1.89%. This scavenging activity happened at the same concentration of 100 µg/ml. The standard positive control, α-tocopherol had inhibition of 97.33 ± 0.54% at 100 µg/ml (Figure 1).

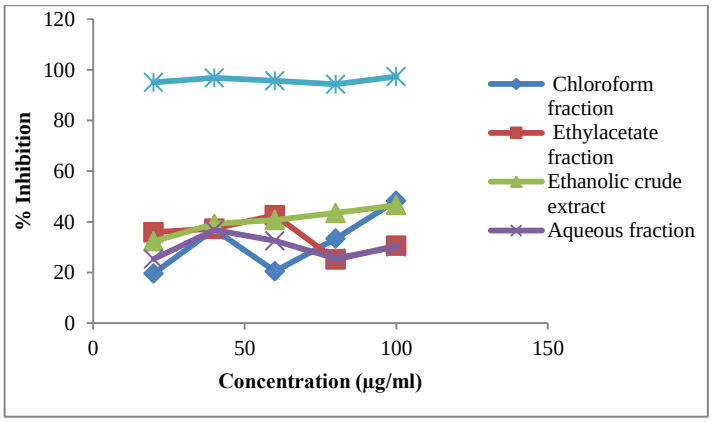

Figure 1 Lipid peroxide free radical scavenging activity of the crude ethanolic extract and fractions of *A. barbadensis* root

DPPH scavenging activity of *A. barbadensis* root extracts

The ethyl acetate fraction had the highest free radical scavenging ability with a percentage inhibition of 82.2 ± 0.44 % at 100 µg/ml while the chloroform fraction had the lowest inhibition of 27.52 ± 2.32 % at the same concentration of 100 µg/ml. The standard positive control (Vitamin C) had an inhibition percentage range of 93.39 % to 94.72 % (Figure 2).

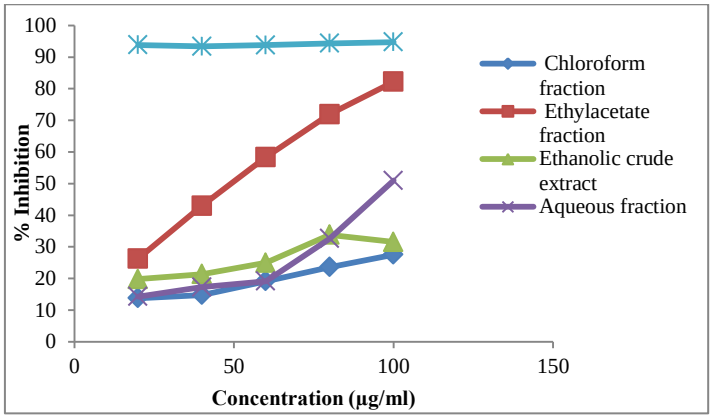

Figure 2 DPPH scavenging activities of crude ethanolic extract and fractions of *A. barbadensis* root with Vitamin C.

Ferric reducing power of *A. barbadensis* root extracts

The ferric reducing power potential of the ethanolic extract and fractions are shown in Figure 3. The highest reducing power activity was observed in the ethyl acetate fraction, with an average absorbance range of 0.145 ± 0.01 to 0.36 ± 0.00 nm. (Figure 3). The positive control (Vitamin C) had a concentration-dependent ferric reducing power within an absorbance range of 0.962 to 1.513 nm.

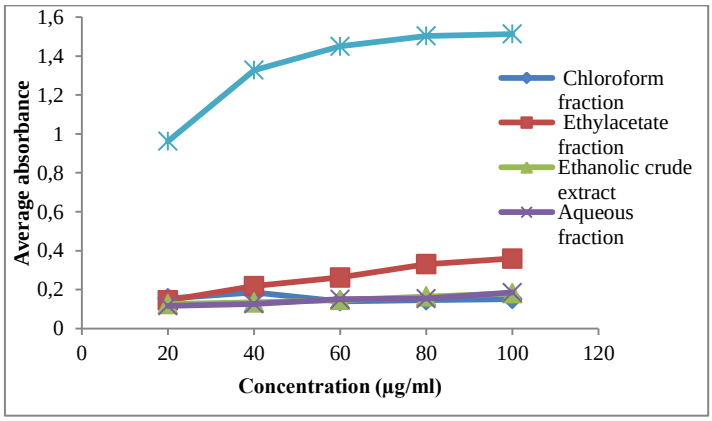

Figure 3 Ferric reducing powers of the crude ethanolic extract and fractions of *A. barbadensis* root

Preliminary phytochemistry and nutritional attributes of *A. barbadensis* root extract

Qualitative phytochemistry of ethanol root extract of *A. barbadensis* and its fractions

The prefatory phytochemical screening of the crude ethanolic root extract and fractions (chloroform, ethyl acetate and aqueous) of *A. barbadensis* revealed the presence of condensed tannins, phenolic compounds, flavonoids, glycosides and reducing sugars in all the tested extracts. Saponins were absent only in the chloroform extract while terpenoids were present in the chloroform and ethyl acetate fractions only. Alkaloids were present in the ethanol extract and ethyl acetate fraction only. Steroids were only present in the ethyl acetate fraction (Tab 5).

Table 5 Qualitative phytochemical composition of the ethanol extract and fractions of *A. barbadensis* root

Phytochemicals	Ethanol extract	Fractions		
		Chloroform	Ethyl acetate	Aqueous
Glycoside	+++	++	+	++
Cardiac glycoside	+	-	-	-
Saponin	++	-	+	++
Flavonoid	++	+++	+	+
Phenolic compound	++	++	++	+
Hydrolysable tannin	-	-	-	-
Condensed tannin	++	++	++	+
Phlobatannins	+	-	-	-
Terpenoid	-	++	++	-
Alkaloid	+	-	+	-
Polysaccharide/Starch	-	-	-	-
Reducing sugar	+++	++	++	++
Steroids	-	-	+	-

Legends: + + +; appreciable amount; + +: moderate amount; +: minute amount; -: not detected

Quantitative analysis of *A. barbadensis* root.

The percentage total phenolic content was found to be highest (2.12 ± 0.003) quantitatively of the secondary metabolites determined, closely followed by saponins (1.75 ± 0.001). Alkaloids had the lowest concentration of 0.121± 0.001% (Tab 6).

Table 6 Quantitative Composition of the ethanol extract of *Aloe barbadensis root*

Parameters	Composition (%)
Alkaloids	0.12 ± 0.001
Flavonoids	1.21 ± 0.002
Saponins	1.75 ± 0.001
Phenolics	2.12 ± 0.003

Values represent Mean ± SEM of three (3) determinations.

Proximate composition of *A. barbadensis* root

The percentage composition on a dry weight basis presented in Table 7 indicated that the root of *A. barbadensis* contained a moderate amount of crude fibre (30.05 ± 2.33%) and 24.66 ± 1.89% nitrogen free extract.

Table 7 Proximate Composition of the root of *Aloe barbadensis*

Parameters	Composition (%)
Moisture Content	10.23 ± 0.04
Ash Content	18.48 ± 0.32
Crude Oil	9.15 ± 1.34
Crude Protein	7.44 ± 0.62
Crude Fibre	30.05 ± 2.33
Nitrogen Free Extract (NFE)	24.66 ± 1.89

Values represent Mean ± Standard deviation of two (2) determinations on dry Weight basis.

Mineral element composition of the root of *A. barbadensis.*

The root extract was found to have a considerable concentration of iron (37.42 ± 0.001 mg/kg), sodium (21.01 ± 0.002 mg/kg) and potassium (15.14 ± 0.001 mg/kg) (Tab 8). The amount of zinc quantified was low (0.02± 0.003), while cadmium and lead were not detected.

Table 8 Minerals Composition of the root of *Aloe barbadensis*

Minerals	Concentration (mg/kg)
Potassium	15.14 ± 0.001
Calcium	3.12 ± 0.003
Sodium	21.01 ± 0.002
Magnesium	2.01±0.002
Iron	37.42 ± 0.001
Zinc	0.02± 0.003
Manganese	0.10±0.001
Lead	ND
Cadmium	ND

Legend: ND- Not Detected; Values represent Mean ± SEM of three (3) determinations on a dry weight basis.

Cytotoxic and genotoxic evaluation of AB root extract

Effect of AB root extract on root length and meristematic cells of *Allium cepa*

The highest growth rate (root length) was observed at 0.25 mg/ml with a root length of 1.74 ± 0.05 cm, while the lowest growth was observed at1 mg/ml with a corresponding root length of 0.76 ± 0.03 cm (Tab 9). The mitotic indices, types and numbers of aberrations of the different concentrations of the crude ethanol extract of *Aloe barbadensis* root were also recorded in this study. It was observed that the mitotic index decreased with an increase in concentration of the extract (Table 10) while several chromosomal aberrations were induced in the *Allium cepa* root cells when exposed to the ethanol extract of *A. barbadensis* (Figure 4).

Table 9 Mean root length of *Allium cepa* exposed to crude ethanolic extract of *A barbadensis root.*

Concentrations of extract (mg/ml)	Number of Roots	Mean root length (cm)	% Root Growth of control
Control	100	3.11 ±0.07	100.00
0.25	100	1.74 ± 0.05	55.95
0.5	90	1.15 ± 0.05	36.98
0.75	100	1.14 ± 0.05	36.66
1	93	0.76 ± 0.03	42.44

Values are Mean ± SEM of five replicates; Control – distilled water

Table 10 Cytological effects on *Allium cepa* root tip cells grown in various concentrations of ethanolic extract of *Aloe barbadensis* root

Concentrations of extract (mg/ml)	No. of dividing Cells	Total No. of cells counted	Mitotic Index± S.E (cm)	Mitotic Inhibition	Laggard	Polar deviation	Bridge	C – mitosis	Disturbed cell	Vagrant	Multipolar	Stickiness	% Aberrant Cell + S.E (cm)
Control	174	2113	8.63 ± 0.38	0.00	1		2			1		1	0.24 ± 0.25
0.25	155	2043	7.40 ± 0.42	13.79	4	7	5	5	3	5		5	1.66 ± 0.46
0.50	137	2145	6.29 ± 0.38	27.11	6	6	9	4	4	8		7	2.05 ± 0.71
0.75	128	2118	6.04 ± 0.43	30.01	6	8	9	6	4	8	1	5	2.27 ± 0.97
1	116	2045	7.36 ± 0.34	34.88	9	4	11	7	4	10	1	12	2.84 ± 1.39

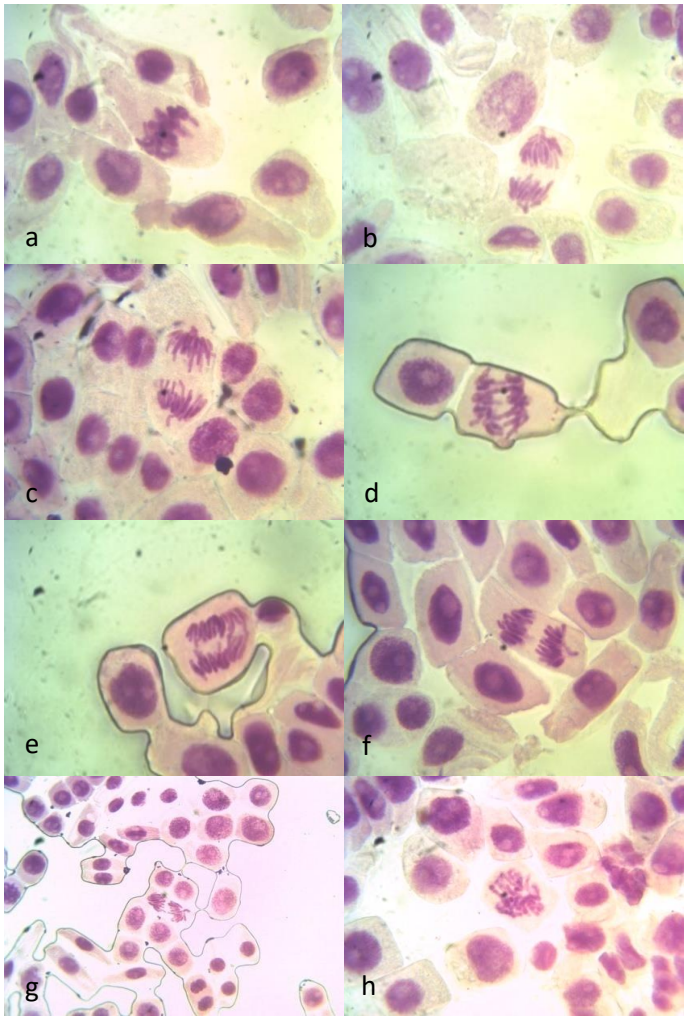

Figure 4 Chromosomal aberrations induced in *Allium cepa* root exposed to ethanol extract of *A. barbadensis* root (a) Vagrant + Polar deviation, (b) Sticky metaphase, (c) Vagrant cell on anaphase, (d) Multiple bridges, (e) Bridge, (f) Vagrant, (g) Polar deviation, (h) C- mitosis

Effect of AB root extract on root length of *Sorghum bicolor* (guinea corn)

A significant reduction (*P<0.01*) in the radicle length of *Sorghum bicolor* when exposed to the ethanolic extract of *A. barbadensis* during the entire period of the study was noticed (Tab 11). After 24 hr, a significant reduction in the average radicle length (0.13±0.09 mm) of the seeds of guinea corn was recorded at the 20 mg/ml concentration of the extract compared to an average length of 4.17±0.42 mm observed in the control. The effect of the extract on the radicle length of *S.bicolor* at all tested concentrations after 24 and 48 hrs (except 5 mg/ml) were not significantly different from each other but differed significantly *(P<0.01*) from the control.

DISCUSSION

Antimicrobial potential

Infectious diseases, according to **Okigbo and Ajalie (2005)**, are responsible for over one percent of deaths occurring in tropical countries. Infertility is one ailment that has been linked to these infections and accounts for two-thirds of the recorded infertility cases in Nigeria (**Okonofua *et al.*, 1995)**. Similarly, polymicrobial infections have been linked to many cases of erectile dysfunction (**Momoh *et al.*, 2011**). In this study, ethanolic root extract of *A. barbadensis* was active against the Gram-positive and Gram-negative bacterial species isolated from the semen samples with a zone of inhibition of ≥ 10 mm (**Andrews, 2005; Usman *et al.*, 2005**). The inhibitory activity of the extract was found to be concentration-dependent (Tab 2). The isolated bacterial species from this study were similar to species isolated from earlier work (**Ekhaise and Richard, 2008; Ibadin and Ibeh, 2008; Komolafe and Awoniyi, 2013**). This probably reflects the prevalence of these bacterial species associated with male infertility due to their ability to cause polymicrobial infections. The ethanolic extract of *A. barbadensis* at 50 mg/ml had the highest inhibitory activity against the isolated organisms. It is noteworthy that the extract suppressed the growth of the isolated Gram-negative bacterial species. This indicated a very good antimicrobial property of the extract which agrees with the report of **Ashafa *et al.* (2008)** on the ability of the stem extracts of *Felicia muricata* to inhibit all Gram-negative bacterial strains tested. Of the isolated bacterial species, *E. coli*, *P. aeruginosa* and *S. saprophyticus* had lower tolerance to the extract when compared to the control drug, at 50 mg/ml (Tab 2). The MIC and MBC values of the extract (Tab 3) suggested that the tested organisms had low resistance. According to the MIC result, the extract was most active against *E. coli* and *S. saprophyticus* with the same MIC value of 6.25 mg/ml compared to a MIC of 12.5 mg/ml obtained for the other organisms. The extract had bactericidal activity against all the tested bacteria with an MBC value of 12.5 mg/ml for all the isolated species. It can, therefore, be inferred that the bactericidal activity of the extract against the isolated organism may prevent the inhibition of spermatogenesis and sperm function impairment in infertile males (**Ibadin and Ibeh, 2008**).

Antioxidant study

Antioxidants are chemical substances used to prevent human diseases and disorders, including male infertility. They scavenge or inhibit the production of free radicals (reactive oxygen species), which are very toxic to different forms of biological molecules (DNA, lipids, protein and carbohydrates) (**Ashafa *et al.*, 2010; Raghuveer and Vyas, 2010**). In the antioxidant assays, the ethanol extract and fractions of *A. barbadensis* exhibited strong scavenging activity against the free radicals. **Prasad and Ramakrishnan (2012**) reported that the exposure of unsaturated lipids in cell membranes and liver tissues to reactive oxygen species (ROS) make them very susceptible to peroxidation. The crude extract and fractions from the results obtained from this assay displayed a significant antiperoxidant effect (Figure 1). The chloroform fraction had the highest scavenging/ inhibitory activity of 48.28±0.34 %, followed by the crude ethanolic extract (46.57±0.69 %). The antiperoxidant effect of the ethanolic extract which was observed to be concentration dependent was similar to the findings of a previous study elsewhere (**Prasad and Ramakrishnan, 2012)** where the lipid peroxide radical scavenging potential of *Rumex vesicarius* was investigated. The inhibitory lipid peroxidation potential of the extract and fractions reflects their ability to probably reduce the risk factors associated with the induction and development of atherosclerosis (**Godwin and Prabhu, 2006**); slow down the aging process and boost immune responses (**Prasad and Ramakrishnan, 2012)**. The lipid peroxidation inhibitory capacity of the extract and fractions is also very remarkable as it can be linked to the ability of the plant to prevent peroxidative damage to the plasma membrane of the sperm and loss of DNA structural integrity. This also ensures prevention of cell mortality and enhanced fertility (**Lombardo *et al.*, 2011**). The potent lipid peroxidation inhibitory effect of the extract and fractions can also be an indication that the plant may prevent loss of sperm membrane fluidity, which in turn improves sperm motility and sperm oocyte fusion (**Raghuveer and Vyas, 2010**). The lipid peroxidation inhibition capacity in this study supports the use of the plant in treating reproductive disorders/ailments, as previously reported by **Erhabor and Idu (2017)**. For the DPPH assay (Figure 2), the extract and fractions had a high scavenging activity against the reactive oxygen species produced.

Table 11 Growth inhibitory effects of the ethanol extract of *A. barbadensis* on the radicle length (mm) of guinea corn.

Groups	24hrs	48hrs	72hrs	96hrs	P-Value
Control	$4.17^{a\#}$±0.42	$16.63^{a\#\#}$±2.8	$22.83^{a\#\#\#}$±0.08	$30.47^{a\#\#\#\#}$±0.02	**P<0.01
5mg/ml	$0.30^{b\#}$±0.06	$7.57^{c\#\#\#}$±0.88	$2.63^{c\#\#}$±0.01	$3.33^{c\#\#}$±0.03	*P<0.05
10mg/ml	$0.20^{b\#}$±0.58	$0.65^{b\#}$±0.40	$1.03^{e\#\#}$±0.02	$2.03^{e\#\#\#}$±0.01	**P<0.01
15mg/ml	$0.33^{b\#}$±0.09	$1.23^{b\#\#}$±0.27	$3.03^{b\#\#\#\#}$±0.03	$2.03^{e\#\#\#}$±0.01	**P<0.01
20mg/ml	$0.13^{b\#}$±0.09	$0.27^{b\#}$±0.03	$1.41^{d\#\#}$±0.01	$2.50^{d\#\#\#}$±0.02	**P<0.01
25mg/ml	$0.80^{b\#}$±0.26	$1.63^{b\#}$±0.9	$3.11^{b\#\#}$±0.01	$3.78^{b\#\#\#}$±0.01	**P<0.01
P-Value	***P<0.01*	***P<0.01*	***P<0.01*	***P<0.01*	

Values are mean±SEM; n = 20, *Note: **P<0.01-Highly Significant, *P<0.05-Significant, P>0.05-Not Significant*
Different superscript letters (in columns) shows that the mean is significant from others.
Different$^{\#}$(in rows) shows there is a significant difference across the sampled means across the hours.

The ethyl acetate fraction had the highest activity compared to the crude extract and other fractions. The concentration-dependent scavenging activity of the ethanol extract until the penultimate concentration (80 µg/ml) was observed. It was also noticed that the fractions had a concentration-dependent scavenging activity. The ability of the extract to inhibit the oxidation of DPPH implies that it can donate protons and scavenge other stable radicals. This agrees with the findings of **Falodun and Irabor (2008)** where a concentration-dependent scavenging ability of the methanol extract of *Calliandria surinamensis* was observed against the DPPH radical. Within the concentrations used in the ferric reducing power assay, the ethanolic extract, fractions (ethyl acetate and aqueous) and Vitamin C (positive control) exhibited a reducing power that increased as the concentration increased (Figure 3). This reducing power may be attributed to the ability of the extract and fractions to donate hydrogen that can react with reactive oxygen species to stabilize and end radical chain reactions (**Sudha *et al*., 2011**). It may also be due to hydrophilic polyphenolic compounds in the extract and fractions (**Okolie *et al*., 2011; Yen *et al*., 1993**), which have been implicated as being effective hydrogen donors (good oxidants). These findings are in sync with previous reports (**Al-Fartosy, 2011; Okolie *et al*., 2011; Sudha *et al*., 2011**) were the reductive capacities (antioxidant potential) of the ethyl acetate extract of pepino fruit; ethanol root extract of *Rauwolfia vomitoria* and methanol extract of *Inula graveolus* increased proportionally to their respective concentrations.

Phytochemical and nutritional composition of AB root

Investigations into the phytochemical constituents of the ethanol extract and its fractions revealed that glycosides, flavonoids, phenols, tannins and reducing sugars were present in the extract and fractions (Tab 5). The saponin content of the crude extract may have been responsible for the earlier reported aphrodisiac property of *A. barbadensis* root (**Erhabor and Idu, 2017**). This is because saponin has been previously postulated as a possible constituent responsible for the aphrodisiac property of *Tribulus terrestris*. It has also been found to increase testosterone in the body by raising the concentrations of luteinizing hormone which helps to sustain testosterone concentrations (**Gauthaman, *et al*., 2002**). Also, the aphrodisiac potential of clove (*Syzygium aromaticum*) has been attributed to the presence of sterols or phenolic compounds from an earlier study (**Ahmad *et al*., 2004**). The crude extract from the phytochemical results contained phenols but lacked sterols which were present only in the ethyl acetate fraction. Alkaloids have been reported to increase the dilation of blood vessels in the male sex organ (**Zamblé *et al*., 2008**) and are said to be one of the mechanisms of action through which aphrodisiac substances act. The ethanol extract of *A. barbadensis* was also found to contain an alkaloid, which can be linked to the increased blood flow noticed in the histological studies (**Erhabor, 2015**). Therefore, phenols, alkaloids and saponins may synergistically or individually be responsible for the aphrodisiac activity of the ethanolic extract of *A. barbadensis*. This agrees with the assertion that phytoconstituents potentiates erection and prolong ejaculatory latency in male rats (**Muanya and Odukoya, 2008; Sumalatha *et al*., 2010; Yakubu *et al*., 2005**). The occurrence of phenol in the extract may be responsible for its probable cytotoxic and antiproliferative properties. This can be inferred from earlier studies adducing the cytotoxic and antiproliferative activity of phenols against melanocytes cell lines (**Ayinde and Agbakwuru, 2010**). **Hour *et al*. (1980)** also reported that saponins and flavonoids had antitumor and anticancer properties and as a result, their presence in *A. barbadensis* could be linked to its relatively toxic effect on cells. Again, phenols and flavonoids have been suggested to be responsible for the radical scavenging activities of most drugs (**Falodun *et al*., 2011**). Therefore, it is possible that these secondary metabolites can be responsible for the antioxidant property of the ethanolic extract and fractions of *A barbadensis*. However, the tannins in the root extract of *A. barbadensis* may cause its antimicrobial activity as earlier reported by **Sodipo (1991)**. Flavonoids which have also been reported to have antimicrobial properties by **Yebpella *et al*. (2011)** are one of the chemical constituents in the extract of *A. barbadensis*. It is pertinent to note that the percentage composition of alkaloids, flavonoids and phenols (0.12%; 1.21% and 2.12% were lower than those recorded for the leaves of *Cymbopogon citratus* which had concentrations of 1.38%, 4.76% and 3.58% respectively. The saponin content (1.75%) was, however greater than that of *C. citratus* leaves (1.25%) (**Uraku *et al*., 2016**).

The proximate constituents of a plant indicate nutrients available in the plant. The nitrogen free extract (NFE) in *A. barbadensis* root (Tab 7) may have provided the required energy for cellular activities such as arousal of the penis when metabolized in the body as reported by **Odesanmi *et al*. (2012)** on the nutritional prospect of *Microdesmis keayana*. The NFE of *A. barbadensis* was found to be lower than that of *Acorous calamus* (75.03%) as reported by **Barua *et al*. (2015)**. The root of *A. barbadensis* also has protein which may form a pool for amino acids such as arginine which have been suggested to potentiate the aphrodisiac properties of medicinal plants (**Sumalatha *et al*., 2010**). The protein value from this present study was found to be lower than that reported by **Kibar and Temel (2016); Kibar and Kibar (2017)** for some wild edible plants. The moisture and ash contents are two values that can show the stability, quality and purity of a powdered crude drug. The moisture and ash contents of *A. barbadensis* root (Table 7) were relatively high when compared to that of *Microdesmis keayana* root (**Odesanmi *et al*., 2012**). It was also observed that the ash content of *A. barbadensis* root (18.48%) was higher than that of *C. citratus* leaves (7.15%) as reported by **Uraku *et al*. (2016)**. Interestingly, *A. barbadensis* root collected from the wild was also found to have higher ash content than the values reported by **Kibar and Temel (2016); Kibar and Kibar (2017)** for some wild edible plant species. The high values can be linked to its ability to store water for a long time, being a drought-resistant plant.

Micronutrients play vital roles in metabolism and enzymatic reactions when they function as co-factors and co-enzymes. The magnesium concentration in the root sample of *A. barbadensis* (Tab 8) can be described as low (2.01±0.002 mg/kg) when compared to that of *Microdesmis keayana* root (458.8±5.44 ppm). However, magnesium is useful in the biosynthesis of sex hormones such as androgen, oestrogens and neurotransmitters (dopamine and norepinephrine which aid in sex modulation) (**Odesanmi *et al*., 2012**). Potassium which function as a major cation of intercellular fluid, regulate the electrode potential and concentration of aldosterone and cell membrane permeability as reported by **Robert *et al*. (2003)** and was detected in moderate amount (15.14±0.001mg/kg) in the root of *A. barbadensis*. The concentrations of iron (37.24±0.001 mg/kg) and sodium (21.01±0.002 mg/kg) were higher than those of *Microdesmis keayana* which recorded values of 5.62±0.48 ppm and 5.68±0.25 ppm. The quantification of calcium in the root of *A. barbadensis* was very important as it helps in protecting the structural integrity of the skeleton and provides energy for contraction via a complex reaction involving vital enzymes regulating metabolism at the intermediary level (**Breslau, 1991**). Another mineral element, zinc, functions in boosting the immune system and is necessary for the effective synthesis of testosterone and as such can prevent or be used in managing male infertility (**Odesanmi *et al*., 2012**). The presence of zinc maybe responsible for the ability of the plant to boost the immune system and increased testosterone which supports the aphrodisiac use of the plant.

Cytotoxic and genetoxic effects of *A. barbadensis* root

Earlier report by **Grant (1994)** reported the ability of the meristematic cells of *Allium cepa* root tip to interact with mutagenic substances and as a result, these are used to detect the cytotoxic and genotoxic effects of harmful chemicals. The macroscopic evaluation of the *A. cepa* root showed that the extract at all concentrations decreased the root growth of the tested onion bulbs when compared to the control (distilled water) (Tab 9). This finding implied that the extract may be relatively toxic and can inhibit tumour-producing cells. From an earlier study, inhibition of the root growth of *A. cepa* suggests a reduction in the number of dividing cells (**Olorunfemi *et al*., 2011**), which reflects the mitodepressive effect of the extract on the ability of *A. cepa* cells to divide. Thus, the extract may have the ability to inhibit the synthesis of DNA and nucleus – proteins, a process that had been reported with extract of other plants (**Stephen, 1980; Timothy *et al*., 2014**). These results support report in this study where the effect of the extract inhibited the radicle growth of *Sorghum bicolor* seeds.

Following the previous report by **Smaka-Kincl *et al*. (1996)**, a decrease in the rate of mitotic index is one acceptable measure for determining the cytotoxic effect of harmful substances on all living organisms. The ethanol extract of *A. barbadensis* showed a concentration dependent decrease in mitotic index (MI) which implies a cytotoxic effect (Tab 10). This is consistent with an earlier report by **Timothy *et al*. (2014)** where the extract of *Icacina trichantha* decreased the MI of *A. cepa* root cells. Also, **Olorunfemi *et al*. (2014)** stated that chromosomal aberration and abnormalities are useful parameters for assessing genotoxic activity of a substance. These aberrations include sticky chromosomes, vagrant chromosomes, bridges, polar deviations and spindle disturbance. It was observed that the extract induced certain levels of chromosomal aberrations at all the tested concentrations (Figure 4). This concentration-dependent activity of the extract on the chromosomes of the *A. cepa* root cells implies a cyto-genotoxic effect of the extract. Therefore, it can be inferred that the extract has the functional therapeutic ability to destroy cancerous cells (**Dahof, 1983**). This finding also agrees with previous studies by **Timothy *et al*., (2014)** were the cyto-genotoxic activity of the leaf extract of *Icacina trichantha* on *A. cepa* root cells was tested.

It has been reported that meristematic tissues of seeds can rapidly multiply when exposed to favourable conditions. The level of proliferation is reflected in an increase in the radicle length of the seeds during a defined period (96 hrs) (**Ikpefan *et al*., 2013**). Also, germination inhibition and subsequent growth of the *Sorghum* spp radicle have been suggested as a measure to determine the herbicidal potential of a plant extract or its ability to induce dormancy and retard the growth of tumor-producing cells (**Sogbaike *et al*., 2002**). Administration of the ethanol extract of *A. barbadensis* at all concentrations and within the tested period significantly ($p<0.01$) inhibited the growth of *Sorghum bicolor* radicle (Tab 11). The extract displayed a concentration-dependent reduction in the radicle length of *S. bicolor* seeds when compared to the control. The extract of *A. barbadensis* root exerted the greatest significant ($p<0.01$) inhibitory effect against the growth of the *S. bicolor* radicle at 20 mg/ml. The result implies that the extract possessed an anti-proliferative activity which suggests that it can be used to retard the growth of cells that produce tumours. This finding agrees with earlier reports by **Fajana (2013)**, **Ikpefan *et al*. (2013)** on the growth inhibitory activity of the methanol extract of *Tridax procumbens* and *Cnidoscolus*

acontifolius on the radicle length of guinea corn seed. This *in vitro* cytotoxic result does not, however, agree with previous findings by **Erhabor and Idu (2017)** on the *in vivo* acute toxicological effect of the *A. barbadensis* root extract within the utilized dose range (100 to 400 mg/kg).

CONCLUSION

This study revealed the potential of *A. barbadensis* root extract as a strong scavenger of free radicals with relatively good antimicrobial activities though at moderately high concentrations. Lead and cadmium were not detected in the plant which probably showed lack of contamination of the plant material. It should be noted, however, that the fractions of AB root were not assessed for antimicrobial, quantitative phytochemical, cytotoxic and genotoxic activities due to insufficient samples. The moderately high nutrient content of the extract may be a possible mechanism for the aphrodisiac property of the plant. The results of the biological activities of *A. barbadensis* root is an indication of its potential use in drug development for managing infectious diseases and other related ailments. Although the results suggest that the extract may be cytotoxic, there is a need to assess the plant in other *in vitro* and *in vivo* assays using relevant cell lines, and in long term animal model studies.

List of abbreviations: AB: *Aloe barbadensis*, **DPPH**: 1, 1, diphenyl-2-picrlhydrazyl, **FRAP**: ferric reducing power, **MIC**: minimum inhibitory concentration, **MBC**: minimum bactericidal concentration.

Conflict of interest: The authors declare that no conflict of interest is in existent.

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REFERENCES

ADODO, A. (2012). *Nature power: A Christian Approach to Herbal medicine.* (3rd ed.). Ewu, Edo State, Nigeria: PAX Herbal Clinic & Research Laboratories.

AHMAD, S., LATIF, A., & QASMI, I. A. (2003). Aphrodisiac activity of 50% ethanolic extracts of *Myristica fragrans* Houtt.(nutmeg) and *Syzygium aromaticum* (L) Merr. & Perry.(clove) in male mice: a comparative study. *BMC Complement Altern Med.*, *3*(1), 6. https://doi.org/10.1186/1472-6882-3-6

AHMAD, S., LATIF, A., & QASMI, I. A. (2004). Effect of 50% ethanolic extract of *Syzygium aromaticum* (L.) Merr. & Perry.(clove) on sexual behaviour of normal male rats. *BMC Complement Altern Med.*, *4*(1), 17. https://doi.org/10.1186/1472-6882-4-17

AKERELE, O. (1987). The best of both worlds: bringing traditional medicine up to date. Soc Sci Med., *24*(2), 177-181. https://doi.org/10.1016/0277-9536(87)90250-4

AL-FARTOSY, A. J. M, (2011). Antioxidant properties of methanolic extract from *Inula graveolens* L. *Turk J Agric For.*, *35*(6), 591-596. doi:10.3906/tar-1010-1268

ANDREWS, J. (2005). BSAC standardized disc susceptibility testing method (version 4). *J Antimicrob Chemother.*, *56*(1), 60-76. https://doi.org/10.1093/jac/dki124

ASHAFA, A., GRIERSON, D., & AFOLAYAN, A. (2008). Antimicrobial activity of extract from *Felicia muricata* Thunb. *J. Biol. Sci*, *8*(6), 1062-1066. https://doi.org/10.3923/jbs.2008.1062.1066

ASHAFA, A., GRIERSON, D., & AFOLAYAN, A. (2010). *In vitro* antioxidant activity of extracts from the leaves of *Felicia muricata* Thunb. an underutilized medicinal plant in the Eastern Cape Province, South Africa. *Afr J Tradit Complement Altern Med*, *7*(4). https://doi.org/10.4314/ajtcam.v7i4.56695

AYINDE, B., & AGBAKWURU, U. (2010). Cytotoxic and growth inhibitory effects of the methanol extract *Struchium sparganophora* Ktze (Asteraceae) leaves. *Pharmacogn Mag*, *6*(24), 293. https://doi.org/10.4103/0973-1296.71795

BARUA, C. C., BORA, M., SAIKIA, B. N., HAZARIKA, M., & BARUA, I. C. (2015). Quantitative Analysis of Proximate and Mineral Composition of A Few Important Medicinal Plants of North East Region. *Int J Appl Biol Pharma Techn*. 2015; 6:188-193.

CHE, Q.-M., AKAO, T., HATTORI, M., KOBASHI, K., & NAMBA, T. (1991). Isolation of a Human Intestinal Bacterium Capable of Transforming Barbaloin to Aloe-Emodin Anthrone1. *Planta Med*, *57*(01), 15-19. https://doi.org/10.1055/s-2006-960007

CHEESBROUGH, M. (2006). *District laboratory practice in tropical countries*: Cambridge university press, United Kingdom.

CHIDAMBARA MURTHY, K. N., SINGH, R. P., & JAYAPRAKASHA, G. K. (2002). Antioxidant activities of grape (*Vitis vinifera*) pomace extracts. *J Agric Food Chem.*, 50(21), 5909-5914. https://doi.org/10.1021/jf0257042

DANHOF, I. E. & MCANALLEY, B. H.. (1983). Stabilized Aloe vera: Effect on Human skin cells. . *Drug and Cosmetic Industry, Danhof, I. E. and McAnally, B. H. (1983) Stabilized Aloe vera: Effect on Human skin cells. Drug Cos. Indus. 133:52-106.*, 52 - 106.

DAVIS, R. H., DONATO, J., HARTMAN, G. M., & HAAS, R. C. (1994). Anti-inflammatory and wound healing activity of a growth substance in *Aloe vera*. *J Am Podiatr Med Assoc.*, *84*(2), 77-81. https://doi.org/10.7547/87507315-84-2-77

EBOJI, O., VENABLES, L., SOWEMIMO, A., SOFIDIYA, M., KOEKEMOER, T., & VAN DE VENTER, M. (2017). *Burkea africana* Hook (Caesalpiniaceae) ethanolic extract causes cell cycle arrest at M phase and induces caspase dependent apoptosis. *S Afr J Bot*, *112*, 361-367. https://doi.org/10.1016/j.sajb.2017.06.013

EDEOGA HO, G. A. (2000). Nutritional values of some non-conventional leafy vegetable in Nigeria. *J Econ Tax Bot.*, *24*, 7-13.

EJIKEME, C. M., EZEONU, C. S., & EBOATU, A. N. (2014). Determination of physical and phytochemical constituents of some tropical timbers indigenous to Niger Delta area of Nigeria. *European Scientific Journal, ESJ*, *10*(18).

EKHAISE, F., & RICHARD, F. (2008). Common bacterial isolates associated with the semen of men complaining of infertility in University of Benin Teaching Hospital (UBTH), Benin City, Nigeria. *World J Med Sci.*, *3*(1), 28-33.

ELISHA, I. L., DZOYEM, J.-P., BOTHA, F. S., & ELOFF, J. N. (2015). The efficacy and safety of nine South African medicinal plants in controlling *Bacillus anthracis* Sterne vaccine strain. *BMC Complement Altern Med*, *16*(1), 5. https://doi.org/10.1186/s12906-015-0980-1

ELOFF, J. N. (1998). A sensitive and quick microplate method to determine the minimal inhibitory concentration of plant extracts for bacteria. *Planta Med*, *64*(08), 711-713. https://doi.org/10.1055/s-2006-957563

EMERUWA, A. (1982). Antibacterial substance from *Carica papaya* fruit extract. *J Nat Prod*, *45*(2), 123-127. https://doi.org/10.1021/np50020a002

ERHABOR, J. O. (2015). *Aphrodisiac potential and biosafety evaluations of the ethanolic extract of Aloe barbadensis Mill. Root.* (Ph.D), University of Benin, Benin City, Nigeria.

ERHABOR, J., IDU, M., & UDO, F. (2013). Ethnomedicinal survey of medicinal plants used in the treatment of male infertilty among the IFA Nkari People of Ini Local Government area of Akwa Ibom State, Nigeria. *Res. J. Recent Sci., ISSN*, *2277*, 2502.

ERHABOR, J., OSHOMOH, E., TIMOTHY, O., OSAZUWA, E., & IDU, M. (2013). Antimicrobial activity of the methanol and aqueous leaf extracts of *Emilia coccinea* (Sims) G. Don. *Niger. J. Biotechnol.*, *25*, 37-45

ERHABOR, J. O., & IDU, M. (2017). Aphrodisiac potentials of the ethanol extract of *Aloe barbadensis* Mill. root in male Wistar rats. *BMC Complement Altern Med*, *17*(1), 360. https://doi.org/10.1186/s12906-017-1866-1

FAJANA, A. (2013). cytotoxic and growth inhibitory effects of the methanol extract of *Tridax procumbens* linn (Asteraceae). *J Pharmacogn Phytochem.*, *2*(1).

FALODUN, A., & IRABOR, E. E. (2008). Phytochemical, proximate, antioxidant and free radical scavenging evaluations of *Calliandria surinamensis*. *Acta Poloniae Pharma Drug Res*, *65*, 571-575.

FALODUN A, U. A., ODIM EE, OGUAZU E. . (2011). Phytochemical and antioxidant properties of Pericarp of *Garcinia kola* extract. *Bayero J Pure Appl Sci.*, *4*(1), 105-109. https://doi.org/10.4314/bajopas.v4i1.23

GAUTHAMAN, K., ADAIKAN, P., & PRASAD, R. (2002). Aphrodisiac properties of *Tribulus Terrestris* extract (Protodioscin) in normal and castrated rats. *Life Sci*, *71*(12), 1385-1396. https://doi.org/10.1016/s0024-3205(02)01858-1

GODWIN, A., & PRABHU, H. R. (2006). Lipid peroxidation of fish oils. *Indian J Clin Biochem*, *21*(1), 202-204. https://doi.org/10.1007/bf02913098

GRANT, W. F. (1994). The present status of higher plant bioassays for the detection of environmental mutagens. *Mut Res*, *310*(2), 175-185. https://doi.org/10.1016/0027-5107(94)90112-0

HARBORNE, J., GREENHAM, J., & WILLIAMS, C. (1973). Phytochemical analysis. *Chapman and Hall company Ltd, London*, *1*, 5-6.

HORWITZ, W., & LATIMER, G. (2000). Official Methods of Analysis of AOAC International, Gaithersburg MA, USA. *Association of Official Analytical chemist.*

HOUR S.S, A. E., CARTE RD. . (1980). Concentration of water melon juice. *Journal of Food Science*, *45*, 718-719.

IBADIN, O., & IBEH, I. (2008). Bacteriospermia and sperm quality in infertile male patient at University of Benin Teaching Hospital, Benin City, Nigeria. *Mala J Microbiol*, *4*(2), 65-67. https://doi.org/10.21161/mjm.12108

IDU, M., ASOWATA, I. AND ERHABOR, J.O. . (2011). Preliminary antimicrobial and Phytochemical study of the aqueous, ethanol, methanol and chloroform extracts of the leaves of *Napoleona imperialis* P. Beauv. (Lecythidiaceae). *Journal of Plant Development Science*, *3*, 225 – 231.

IDU, M., ERHABOR, J. O., & OVUAKPORIE-UVO, O. (2014). Ethnomedicinal plants used by the Idoma people-Benue State, Nigeria. *American Journal of Ethnomedicine*, *1*(1).
IKPEFAN, E. O., AYINDE, B., & GITA, T. (2013). In vitro comparative cytotoxic and growth inhibitory effects of the methanol extracts of the leaf, stem and root barks of Cnidoscolus acontifolius (Mill.) Johnst (Euphorbiaceae). *International Journal of Bioassays*, *2*(2), 445-449.
IWU, M. M. (2014). *Handbook of African Medicinal Plants*: CRC press.
JOSEPH, B., & RAJ, S. J. (2010). Pharmacognostic and phytochemical properties of *Aloe vera* linn an overview. *International journal of pharmaceutical sciences review and research*, *4*(2), 106-110.
KIBAR, B., & KIBAR, H. (2017). Determination of the nutritional and seed properties of some wild edible plants consumed as vegetable in the Middle Black Sea Region of Turkey. *S Afr J Bot.*, 108, 117-125. https://doi.org/10.1016/j.sajb.2016.10.011
KIBAR, B., & TEMEL, S. (2016). Evaluation of mineral composition of some wild edible plants growing in the eastern Anatolia region grasslands of Turkey and consumed as vegetable. *J. Food Process Pres. 40*(1), 56-66.
KOMOLAFE, O.I, AWONIYI, A.O. (2013). The prevalence of microbial isolates associated with infertility in men attending clinic in OAUTHC, Ile – Ife. *International Journal of Microbiology Research and Reviews 1*(5), 88-91.
KUMAR, R., SIVAKUMAR, T., SUNDERAM, R., GUPTA, M., MAZUMDAR, U., GOMATHI, P., . . . MURUGESH, K. (2005). Antioxidant and antimicrobial activities of *Bauhinia racemosa* L. stem bark. *Braz J Med Biol Res*, *38*(7), 1015-1024. https://doi.org/10.1590/s0100-879x2005000700004
LEONG, L., & SHUI, G. (2002). An investigation of antioxidant capacity of fruits in Singapore markets. *Food Chem*, *76*(1), 69-75.
LIN, J. Y. &. TANG., C. Y. (2007). Determination of total phenolic and flavonoid contents in selected fruits and vegetables, as well as their stimulatory effects in mouse splenocyte proliferation. *Food Chemistry*, 101, 140 - 147. https://doi.org/10.1016/j.foodchem.2006.01.014
LOMBARDO, F., SANSONE, A., ROMANELLI, F., PAOLI, D., GANDINI, L., & LENZI, A. (2011). The role of antioxidant therapy in the treatment of male infertility: an overview. *Asian J Androl.*, *13*(5), 690. https://doi.org/10.1038/aja.2010.183
MAENTHAISONG, R., CHAIYAKUNAPRUK, N., NIRUNTRAPORN, S., & KONGKAEW, C. (2007). The efficacy of aloe vera used for burn wound healing: a systematic review. *Burns*, *33*(6), 713-718. https://doi.org/10.1016/j.burns.2006.10.384
MOMOH, A. R. M., IDONIJE, O. B., OKOGBO, F., OKHIAI, O., OKOLO, P. O., TURAY, A. A., & MOMOH, A. A. (2011). A monomicrobial infection of *Staphylococcus aureus* associated with Erectile Dysfunction-A CASE REPORT.
MONGALO, N., MCGAW, L., FINNIE, J., & VAN STADEN, J. (2017). Pharmacological properties of extracts from six South African medicinal plants used to treat sexually transmitted infections (STIs) and related infections. *S Afr J Bot.*, *112*, 290-295. https://doi.org/10.1016/j.sajb.2017.05.031
MOTLHATLEGO, K., NJOYA, E. M., ABDALLA, M., ELOFF, J., & MCGAW, L. (2018). The potential use of leaf extracts of two Newtonia (Fabaceae) species to treat diarrhoea. *S Afr J Bot.*, 116, 25-33. https://doi.org/10.1016/j.sajb.2018.02.395
MUANYA, C., & ODUKOYA, O. (2008). Lipid peroxidation as index of activity in aphrodisiac herbs. *Plant Sci*, *3*(1), 92-98. https://doi.org/10.3923/jps.2008.92.98
NA, B. (1991). Calcium Homeostasis. In *Textbook of Endocrine physiology* (2nd edition ed., pp. 276-308). Dallas: Griffin and Ojeda.
OBADONI, B., & OCHUKO, P. (2002). Phytochemical studies and comparative efficacy of the crude extracts of some haemostatic plants in Edo and Delta States of Nigeria. *Global J Pure Appl Sci.*, *8*(2), 203-208. https://doi.org/10.4314/gjpas.v8i2.16033
OBICI, S., OTOBONE, F. J., DA SILVA SELA, V. R., ISHIDA, K., DA SILVA, J. C., NAKAMURA, C. V., CORTEZ, D.A.G. & AUDI, E. A. (2008). Preliminary toxicity study of dichloromethane extract of *Kielmeyera coriacea* stems in mice and rats. *J Ethnopharmacol.*, *115*(1), 131-139. https://doi.org/10.1016/j.jep.2007.09.013
ODESANMI, O. S., OJOKUKU, S. A., APENA, A., BIKOMO, O. E., & LAWAL, R. A. (2012). Nutritional prospect of an aphrodisiac *Microdermis keayana*. *J Med Plants Res.*, *6*(7), 1187-1190. https://doi.org/10.5897/jmpr11.919
OHKAWA, H., OHISHI, N., & YAGI, K. (1979). Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. *Anal Biochem.*, *95*(2), 351-358. https://doi.org/10.1016/0003-2697(79)90738-3
OKIGBO, R., & AJALIE, A. (2005). Inhibition of some human pathogens with tropical plant extracts–*Chromolaena odorata* and *Citrus aurantifolia*, and some antibiotics. *International Journal of Molecular Medicine and Advance Sciences*, *1*(1), 34-40.
OKIGBO, R., EME, U., & OGBOGU, S. (2008). Biodiversity and conservation of medicinal and aromatic plants in Africa. *Biotechnol. Mol. Biol. Rev*, *3*(6), 127-134.
OKOLIE, N., ISRAEL, E., & FALODUN, A. (2011). *In-vitro* evaluation of antioxidant potential of *Rauwolfia vomitoria* root extract and its inhibitory effect on lipid peroxidation as indication of aphrodisiac properties. *Pharm Chem J.*, *45*(8), 476-480. https://doi.org/10.1007/s11094-011-0660-5
OKONOFUA, F. E., AKO-NAI, K., & DIGHITOGHI, M. (1995). Lower genital tract infections in infertile Nigerian women compared with controls. *Sex Transm Infect.*, *71*(3), 163-168. https://doi.org/10.1136/sti.71.3.163
OLORUNFEMI, D., OGIESERI, U., & AKINBORO, A. (2011). Genotoxicity screening of industrial effluents using onion bulbs (*Allium cepa* L.). *J Appl Sci Environ Manage*, *15*(1). https://doi.org/10.4314/jasem.v15i1.65700
OLORUNFEMI, D. I., LELEKUMO, E., AND UBAH, M. (2014). Toxicity assessment of Bioremediated contaminated domestic water sources in Niger-Delta by a phytogentoxicity test. . *Nigerian Journal of Life Sciences, 4* (2), 140 – 150.
OLOWOKUDEJO, J., KADIRI, A., & TRAVIH, V. (2008). An ethnobotanical survey of herbal markets and medicinal plants in Lagos State of Nigeria. *Ethnobotanical leaflets*, *2008*(1), 116.
OTANG, W. M., GRIERSON, D. S., & NDIP, R. N. (2012). Phytochemical studies and antioxidant activity of two South African medicinal plants traditionally used for the management of opportunistic fungal infections in HIV/AIDS patients. *BMC Complement Altern Med.*, 12(1), 43. https://doi.org/10.1186/1472-6882-12-43
OYEWOPO, A., OREMOSU, A., AKANG, E., NORONHA, C., & OKANLAWON, A. (2011). Effects of Aloe Vera (Aloe barbadensis) aqueous leaf extract on testicular weight, sperm count and motility of adult male Sprague-Dawley rats. *Journal of American Science*, *7*(4), 31-34.
PRASAD, P. S. H., & RAMAKRISHNAN, N. (2012). *In vitro* lipid peroxidation assay of *Rumex vesicarius* L. *Int J Pharm Pharm Sci*, *4*(Suppl 1), S368-S370.
RAGHUVEER, C., CHAWALA, V. K., SONI, N. D., JAYANT, K., AND, & VYAS, R. K. (2010). Oxidative stress and rile of antioxidants in male infertility. *Pakistan Journal of Physiology*, *6*(2), 54-59.
RANK, J., & NIELSEN, M. (1994). Evaluation of the *Allium* anaphase-telophase test in relation to genotoxicity screening of industrial wastewater. *Mut Res*, *312*(1), 17-24. https://doi.org/10.1016/0165-1161(94)90004-3
ROBBERS, J. E., & TYLER, V. E. (1999). *Tyler's herbs of choice. The therapeutic use of phytomedicinals*: Haworth Press Inc.
ROBERT, K., GRANNER, D. K., & MAYES, P. A. M. (2003). Harper's Illustrated Biochemistry.
ROSS, I. (1999). Medicinal plants of the world, Part 1: Chemical constituents, Traditional and Modern medicinal uses. *New Jersery: Humana Press Inc*, 119-125.
SAHU, P. K., GIRI, D. D., SINGH, R., PANDEY, P., GUPTA, S., SHRIVASTAVA, A. K., . . . PANDEY, K. D. (2013). Therapeutic and medicinal uses of *Aloe vera*: a review. *Pharmacology & Pharmacy*, *4*(08), 599.
SHAMIM, S., AHMED, S. W., & AZHAR, I. (2004). Antifungal activity of *Allium*, *Aloe*, and *Solanum* species. *Pharm Biol.*, *42*(7), 491-498. https://doi.org/10.3109/13880200490891845
SHARMA, R., & LALL, N. (2014). Antibacterial, antioxidant activities and cytotoxicity of plants against *Propionibacterium acnes*. *South African Journal of Science*, 110(11-12), 01-08.
SMAKA-KINCL, V., STEGNAR, P., LOVKA, M., & TOMAN, M. J. (1996). The evaluation of waste, surface and ground water quality using the *Allium* test procedure. *Mut Res*, *368*(3-4), 171-179. https://doi.org/10.1016/s0165-1218(96)90059-2
SMITH, H., EDUFUNKE, I., & ADEYEMI, R. (2000). Toxicity test of water homogenate of *Cissus populnea* stem in mice and its effects on weights of some organs, serum gonadotropins, lipids and uric acid in male albino rats. *Nig Q J Hosp Med.*, *10*(3), 224-227. https://doi.org/10.4314/nqjhm.v10i3.12475
SODIPO OA, A. M., KOLAWOLE, FB, ODUTUGA AA. . (1991). Saponin is the active antifungal principle in *Garcinia kola* Heckle seed. . *Bioscience Research Communication*, *3*, 171.
SOFOWORA, A. (1993). *Medicinal Plants and Traditional Medicine in Africa.* . Ibadan, Nigeria: Spectrum Books Limited.
SOGBAIKE, D., OGUNDAINI, A., & ADESANYA, S. (2002). The effects of some synthesised stilbene analogues on *Artemia salina* naupalii and germination of *Sorghum bicolor* seeds. *Niger J Nat Prod Med*, *6*(1), 19-25. https://doi.org/10.4314/njnpm.v6i1.11686
STEPHEN, J. (1980). Adriamycin induced genetic toxicity as demonstrated by the *Allium* test. *Cytologia*, *45*(4), 769-777.
SUDHA, G., PRIYA, M. S., SHREE, R. I., & VADIVUKKARASI, S. (2011). *In vitro* free radical scavenging activity of raw pepino fruit (*Solanum muricatumaiton*). *Int J Curr Pharm Res*, *3*(2), 137-140.
SUMALATHA, K., KUMAR, S., & LAKSHMI, S. M. (2010). Review on natural aphrodisiac potentials to treat sexual dysfunction. *Int J Pharm Ther*, *1*, 10-18.
TANAKA, M., MISAWA, E., ITO, Y., HABARA, N., NOMAGUCHI, K., YAMADA, M., . . . INAGAKI, M. (2006). Identification of five phytosterols from *Aloe vera* gel as anti-diabetic compounds. *Biol Pharm Bull*, 29(7), 1418-1422. https://doi.org/10.1248/bpb.29.1418
TIMOTHY, O., IDU, M., OLORUNFEMI, D., & OVUAKPORIE-UVO, O. (2014). Cytotoxic and genotoxic properties of leaf extract of *Icacina trichantha* Oliv. *S Afr J Bot.*, *91*, 71-74. https://doi.org/10.1016/j.sajb.2013.11.008

Trease, G. (1996). Trease and Evans pharmacognosy 14th edition wb Ssuvnder company limited. London *Pp191-293.*

URAKU A.J., O. A., OGBANSHI ME, ONUOHA SC. . (2016). Nutritive and anti - nutritive potentials of *Cymbopopgon citratus* leaves. *American Journal of Food and Nutrition*, *20166*(1), 14 -22.

USMAN, H., HARUNA, A., AKPULU, I., ILYAS, M., AHMADU, A., & MUSA, Y. (2005). Phytochemical and antimicrobial screenings of the leaf extracts of *Celtis integrifolia* Lam. *J. Trop. Biosci*, *5*(2), 72-76.

VENABLES, L., KOEKEMOER, T., VAN DE VENTER, M., & GOOSEN, E. (2016). Isoalantolactone, a sesquiterpene lactone from *Artemisia afra* Jacq. ex Willd and its *in vitro* mechanism of induced cell death in HeLa cells, *S Afr J Bot*., *103*, 216-221. https://doi.org/10.1016/j.sajb.2015.08.016

WEST, D. P., & ZHU, Y. F. (2003). Evaluation of *Aloe vera* gel gloves in the treatment of dry skin associated with occupational exposure. *Am J Infect Control*., *31*(1), 40-42. https://doi.org/10.1067/mic.2003.12

YAGI, A., SATO, Y., MIWA, Y., KABBASH, A., MOUSTAFA, S., SHIMOMURA, K., & EL-BASSUONY, A. (2006). Ribosomal DNA sequence analysis of different geographically distributed *Aloe vera* plants: Comparison with clonally regenerated plants. *Saudi Pharmaceutical Journal*, *14*(3/4), 208.

YAKUBU, M., AKANJI, M., & OLADIJI, A. (2005). Aphrodisiac potentials of the aqueous extract of *Fadogia agrestis* (Schweinf. Ex Hiern) stem in male albino rats. *Asian J Androl*., *7*(4), 399-404. https://doi.org/10.1111/j.1745-7262.2005.00052.x

YAKUBU, M., AKANJI, M., & OLADIJI, A. (2007). Male sexual dysfunction and methods used in assessing medicinal plants with aphrodisiac potentials. *Pharmacognosy Reviews*, *1*(1), 49.

YEBPELLA, G., MM, A. H., HAMMUEL, C., MAGOMYA, A., AGBAJI, A., & OKONKWO, E. (2011). Phtyochemical screening and comparative study of antimicrobial activity of *Aloe vera* various extracts. *Afr J Microbiol Res*., *5*(10), 1182-1187. https://doi.org/10.5897/ajmr10.818

YEN, G. C., DUH, P. D., & TSAI, C. L. (1993). Relationship between antioxidant activity and maturity of peanut hulls. *J Agric Food Chem*., *41*(1), 67-70. https://doi.org/10.1021/jf00025a015

ZAMBLÉ, A., SAHPAZ, S., BRUNET, C., & BAILLEUL, F. (2008). Effects of *Microdesmis keayana* roots on sexual behavior of male rats. *Phytomedicine*, *15*(8), 625- 629. https://doi.org/10.1016/j.phymed.2007.10.002