



Comparative Study of the Antimicrobial Efficacy of Three Medicinal Plants against Dermatophytic Pathogens

Temitayo Veronica Folorunso^{1*}, Anthony Kayode Onifade²,
Kolawole Oladunmoye² and Bamidele Juliet Akinyele²

¹Department of Microbiology, Faculty of Science, Adekunle Ajasin University, P.M.B. 001, Akungba Akoko, Ondo State, Nigeria.

²Department of Microbiology, School of Life Sciences, Federal University of Technology, Akure, Nigeria.

Authors' contributions

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

Article Information

DOI: 10.9734/EJMP/2021/v32i1030423

Editor(s):

(1) Dr. Daniela Rigano, University Federico II of Naples, Italy.

(2) Prof. Marcello Iriti, Milan State University, Italy.

Reviewers:

(1) Rylosona Janarthini, Shanghai Jiaotong University, China.

(2) M. O. Chukwu, Sefako Makgatho Health Science University, South Africa.

Complete Peer review History: <https://www.sdiarticle4.com/review-history/75588>

Original Research Article

Received 11 August 2021
Accepted 23 October 2021
Published 29 October 2021

ABSTRACT

Bixa orellana, *Jatropha curcas* and *Cassia alata* are three of the prominent plants used for traditional medicine in Nigeria. Dermatophytosis also known as tinea or ringworm is the most frequent superficial fungal infections in Nigeria.

Objective: In this Present Study, We Aimed at Comparing The Phytochemical Components and the Antifungal Efficacy of these Medicinal against Selected Dermatophytes.

Study Design: Cross Sectional Study among a Particular Population.

Place and Duration of Study: Department of Microbiology, Federal University of Technology, Akure, Ondo State. Between March 2019 and September 2019.

Methods: The phytochemical contents of the plants were determined and the in-vitro antifungal activities of *Bixa orellana*, *Jatropha curcas* and *Cassia alata* were screened against seven species

of Trichophyton (*T. ajelloi*, *T. mentagrophytes*, *T. rubrum*, *M.gypseum* *T. soudanensis*, *T. tonsurans* and *T. verrucosum*) using agar dilution method.

Results: The phytochemical screening revealed the presence of flavonoid, saponin, phenol, steroids, glycoside, phytosteroids, alkaloids, terpenoid, tannin, and cardiac glycoside in various quantities. The findings from our study showed that the ethanol extracts of these medicinal plants have more antifungal activities than other solvents. However, the hexane and ethanol extracts of *Jatropha curcas* was observed to be significantly higher than other extracts. The zone of inhibition recorded ranges from 22 mm-32 mm and the minimum inhibitory concentration (MIC) of 12.5 mg/ml was recorded.

Conclusion: The ethanolic extract of *Jatropha curcas* showed broad effectiveness against the tested pathogens when compared to other plants and we conclude that the plants antifungal property is concentration dependent. However, we recommend further studies on these plants extracts using a large number of different isolates and solvents.

Keywords: Phytochemical screening; antimicrobial efficacy; medicinal plants and dermatophytes.

1. INTRODUCTION

The hunt for bioactive compounds from medicinal plants have been sponsored with persistent microbial resistance to existing chemotherapeutics agents. Medicinal plants are widely used in management of diseases all over the world [1]. In Nigeria, several thousands of plants species have been claimed to possess medicinal properties and employed in the treatment of many ailments [2] Plants can synthesize a wide variety of chemical compounds that are used to perform important biological functions. *Bixa orellana*, *Jatropha curcas* and *Cassia alata* are three of the prominent plants used for traditional medicine in Nigeria.

Bixa orellana is a perennial, tall shrub that can reach 6–10m (20–33ft) high. It bears clusters of 5 cm (2 in) bright white or pink flowers, resembling single wild roses that appear at the tips of the branches [3]. *Bixa orellana* (Achiote) is a shrub native to a region between northern South America and Mexico but it became cultivated in tropical regions of Asia, such as India, Sri Lanka, and Java mainly for the dye which the seeds yield [4]. The nickname, "lipstick tree", was derived from use of the dye as a cosmetic [5]. The fruits of the *Bixa orellana* are globular, ovoid capsules arranged in clusters resembling spiky looking red-brown seed pods covered in soft spines. Each capsule, or pod, contains 30-45 cone shaped seeds covered in a thin waxy blood-red aril. When fully mature, the pod dries, hardens, and splits open, exposing the seeds [4].

Bixa orellana is used in traditional medicine [6]. The tree has been used in Ayurveda, the folk

medicine practices of India, where different parts of the plant are thought to be useful as therapy. The ethanolic extracts of the leaves and seeds of *B. orellana* showed activity against all the Gram-positive and Gram-negative bacteria and the yeast-like fungus *C. albicans*. The activity appears to be more pronounced in the leaf extract [7]. The leaf extracts of *B. orellana* were reported to exhibit dose dependent antimicrobial activity against the tested pathogens from literatures. The antimicrobial properties of *Bixa orellana* various extracts is probably due to the presence of different phytochemicals compounds in varying proportion.

Jatropha curcas is another plant with a notable ethno-medicinal use. *Jatropha curcas* commonly called physic nut, purging nut or pig nut belong to the family *Euphorbiaceae* used in traditional folklore medicine to cure various ailments in Africa, Asia and Latin America. All parts of the plant: roots, stems, leaves seeds and fruits have been widely used in traditional folk medicine in many parts of West Africa [8].

J. curcas and their extracts are used in traditional medicine, such as antimicrobial [9], antiviral [10], acaricidal activity [11], insecticidal properties [12,13], coagulant and anticoagulant activities [14], antimolluscicidal [15] and anti-inflammatory [16]. Previous researchers have also reported that leaf extract of *Jatropha curcas* has significant wound healing activity [17], antidiabetic [18], and Immunomodulatory effect [19] among others. The potency of *J. curcas* extracts have been heavily linked to the presence of certain phytochemicals in different part of the plant body.

Cassia alata Linn commonly called Ringworm weed belongs to the family *Caesalpiniaceae*. It is an annual or biannual shrub with a nasty smell, 1–4m tall, preferring sunny and moist areas. The leaves are yellowish-green, broad, with 5–14 leaflet pairs, the distal ones often larger and with a notched apex [20].

Cassia alata is one of the most important species in the genus of *Cassia* [21] the potential of the plant has been exploited in medicinal system because of the presence of wide range of bioactive molecules, developing the rich resource of different types of medicines. *C. alata* contains rich amount of anthraquinones and polyphenols [22].

Ethnomedicinally, the leaves are specific for the treatment of ringworm and eczema, scabies, and athlete's foot [23]. Various extracts and different parts of *Cassia alata* have been reported to own many pharmacological activities such as laxative, antifungal [22,24], wound healing [25], antibacterial [26], antihelmintic [27], anti-inflammatory [28], abortifacient [29] and Antiplogenic activity [30].

Antimicrobial activity of the plant is associated with the presence of other phytochemical components such as phenols, tannis, saponins, alkaloids, steroids, flavonoids and carbohydrates [31].

Crushed leaves of this plant are effective in treating various skin diseases like ringworm, eczema, pruritis, itching, and scabies in humans [32]. Moreover, leaves were used to prepare herbal tea, herbal soaps, shampoos and skin care cosmetic products to cure dermatological skin diseases [33].

Dermatophytosis also known as tinea or ringworm is an infection caused by a group of keratinophilic fungi called dermatophytes. The three major genera causing tinea are Trichophyton, Microsporum and Epidermophyton [34]. Dermatophytes colonize the skin, nails, and hair of the humans. Majorly, it affects the keratinous tissues of humans and other vertebrates leading to superficial infections. Antimicrobial hunt from plants to cure infectious diseases is not a new concept as it is old as mankind. Herbal medicines have minimal side effects when compared to synthetic drugs. The alarming increase in dermatophyte infection, drug resistance, frequent remissions and relapses and the hepatotoxicity caused by some

oral synthetic antifungal agents are the matter of deep concern [34].

Dermatophytic infection is the most frequent superficial fungal infections in Nigeria. These fungal infections are not only causing primary diseases, but also responsible for various secondary ailments due to many predisposing factors. A widespread use of broad-spectrum antibiotics and immunosuppressive drugs for these ailments has led to an increase in the incidence of systemic fungal infections due to development of resistant strains of some of the fungi. The dermatophytes included in this study are seven species of Trichophyton which are *T. ajelloi*, *T. mentagrophytes*, *T. rubrum*, *M. gypseum*, *T. soudanensis*, *T. tonsurans* and *T. verrucosum*.

The medicinal plants included in this study have been independently reported to possess antifungal activities against some of these dermatophytes and several pathogenic organisms, but no study has directly compared the antifungal efficacy of *B. orellana*, *Casia alata* and *Jatropha curcas* extracts against the same selected isolates. Hence, in this present study, we compared the phytochemical components, antifungal efficacy and the minimum inhibitory concentration/ minimum fungicidal concentration of *B. orellana*, *Casia alata* and *Jatropha curcas* from various extracts with the aim to indicate the more potent plant between the three against the studied fungal isolates.

2. MATERIALS AND METHODS

The plants materials were collected from different locations in Akungba Akoko between 7:00am and 11:00 am and authenticated at the Herbarium of Plant Science and Biotechnology, Adekunle Ajasin University, Akungba Akoko, Ondo state.

Selected fresh plants materials collected were cleaned with 70% ethanol. They were air dried at 28°C for 10 to 15 days and grinded to powder form by grinder (Atlas, UK). 400grams of the leaf part of the plant was grounded into a 1.2 liters of ethanol solvents and covered in air-tight container with a cork, mixed together and left on the shaker at 37°C for 216hrs after which the filtrate was then evaporated with rotary evaporator at 77/78 revolution per minute (rpm) for 5 minute after which it was decanted. The extracts yielded a dark greenish residual mass. The extracts were then kept in sterile bottles until further experiment.

2.1 Collection of Clinical Samples

The nails, skin scrappings and hair samples were collected randomly from individuals suspected to have the symptoms of dermatophytosis of skin, nails and hair. The infected areas of the body were swabbed with cotton wool moistened with 70% ethanol to remove any dirt and bacteria contaminants.

Samples consisting of epidermal scales and infected hairs were scrapped from the scalp of lesions using a sterile blade or new hair brush. Also swab sticks moistened with sterile peptone water was used to collect pus from inflammatory lesions. The scrapings were collected on a piece of sterile brown envelope and labelled appropriately and kept in a black polythene bag. These samples were transported immediately to the Laboratory for microscopic and cultural examination after collection.

2.2 Preparation Of Culture Media

Glass Petri dishes and other glassware used were sterilized in the oven using dry air sterilization method at 160 °C for 1 h. All the media (Sabouraud Dextrose Agar (SDA) Oxoid, UK product, Dermasel Agar oxoid product and Christensen's Urea Agar Merck product) used in this study were prepared aseptically according to manufacturer's specification.

2.3 Direct Microscopic Examination

Each treated slide was examined microscopically under low (x10) and high (x40) objective for the presence of hyphae and chains of arthrospores. Microscopic examination was done according to [35].

2.4 Isolation of Dermatophytes from the Samples

The samples were inoculated on Sabouraud Dextrose Agar (SDA) containing antibiotics. Each sample was inoculated and incubated at 28 °C for four weeks; this was examined daily for fungal growth at 2-3- day intervals. Identification was done based on the growth characteristics and morphology of the fungal isolates. The macroscopic examination was performed for the rate of growth, colonial morphology, colour, presence of pigmentation in the medium and surface texture that is raised, folded, flat cotton, velvety.

2.5 Sub-Culturing of Fungal Isolates

Sterile Sabouraud Dextrose Agar was prepared and sub-culturing done according to [36]. Fungal isolates were identified using conventional and molecular techniques

2.5.1 Fungi DNA extraction protocol

The DNA extraction was done using the protocol reported by [37]. The supernatant from the procedure was discarded carefully with the DNA pellets intact. Trace of ethanol was removed by drying the pellets at 55°C for 1 minute. The dried pellets were then suspended in 50µl of Tris-EDTA (TE) buffer and stored at -20°C for further analysis.

2.5.2 Polymerase Chain Reaction (PCR) analysis

Polymerase chain reaction was done according to [37]. PCR was carried out in a GeneAmp 9700 PCR System Thermocycler (Applied Biosystem Inc., USA). The ITS gene for characterization of fungi, ITS universal primer set which flank the ITS1 (5.8S) and ITS2 region was used; ITS1: 5'TCC GTA GGT GAA CCT GCG G3'and ITS4: 5'TCC TCC GCT

TAT TGA TAT GC3'. PCR sequencing preparation cocktail consisted of 10µl of 5x GoTaq colourless reaction, 3µl of 25mM MgCl₂, 1µl of 10mM of dNTPs mix, 1µl of 10 pmol each of the ITS1 and ITS4 primers and 0.3units of Taq DNA polymerase (Promega,USA) made up to 42µl with sterile distilled water, 8µl DNA template. The conditions for amplification were as follows; initial denaturation at 94°C for 5min; followed by a 30 cycles consisting of 94°C for 30s, 30secs annealing of primer at 55°C and 72°C for 1 minute 30seconds; and a final termination at 72°C for 10minutes and chill at 4°C.

2.5.3 Phytochemical analysis

Phytochemical analysis; both quantitative and qualitative was carried out on the leaf extracts of the plants in order to determine the presence of secondary metabolites. Plant filtrates were prepared by boiling 20grams of the fresh plant in distilled water. The solution was filtered through a vacuum pump. The freshly prepared crude extracts were qualitatively tested for the presence of secondary metabolites such as

carbohydrates, alkaloids, glycosides, coumarins, tannins, phenols, flavonoids, saponins, and proteins by following the methods described in previous literature according to [38,39].

2.6 Antifungal Analysis of the Plant Extracts against the Isolated Dermatophyte

Muller-Hinton agar was sterilized and allowed to set, then the isolated organisms were incorporated into sterile 9.9 ml of sterile peptone water contained in bijoule bottles. Serial dilution of the inoculum was done up to 104 from which 0.1ml of isolated organism was inoculated onto the agar; the inoculum was introduced unto the solidified sterile Mueller Hinton agar by spread plate method in triplicate. Wells were bored on the inoculated plates using a sterilized stainless-steel cork borer of size 6mm diameter. The prepared extracts were inoculated into the bored wells while the control experiments were also carried out where the wells were filled with 30% Dimethyl sulfoxide (DMSO) and distilled water. The plates were incubated at 25°C for 72 h after which the results were read by measuring the diameters of zones of inhibition around the wells with the aid of a metric ruler and recorded. A larger zone of inhibition indicated that the test substances were active against the fungi, a small or no inhibition zone may mean no activity. The susceptibility resistance of the various isolates against each plant extract was determined by the Clinical Laboratory Standard Institute [40]

2.7 Determination of Minimum Inhibitory Concentration (MIC) and Minimum Fungicidal Concentration (MFC)

The MIC value of the plant extracts was determined as the lowest concentration that completely inhibited fungal growth after 72h of incubation at 28 ± 2°C. For the determination of the MIC, 200 mg of the plant extracts each were mixed with 2000 µl of DMSO for complete dissolution of the extract. The test-tubes were filled with 1.0 ml of Potato dextrose broth. 200 mg/1.0 ml, 100 mg/0.5 ml, 50 mg/0.25 ml, 25 mg/0.125 ml and 12.5 mg/0.0625 ml of the plant extracts was added to the test-tubes respectively and the last test tube without extract was used as negative control of the growth of the fungal in the medium. The MIC estimation of the crude extract was determined using the methods of [41,42,43]. Two-fold dilutions of the crude extract was prepared and 2 ml aliquots of different concentrations of the solution were added to 18 ml of pre-sterilized molten SDA for fungi at 40°C

to give final concentration regimes of 0.050 and 10 mg/ml. The medium was then poured into sterile Petri dishes and allowed to set. The surface of the medium was allowed to dry under laminar flow before streaking with 18 h old fungal cultures. The plates were later incubated at 25°C for up to 72 h for fungi, after which they were examined for the presence or absence of growth. The test tubes were incubated at 28 ± 2°C in an incubator for 48h. The lowest concentration that revealed no visible fungal growth after sub-culturing were taken as MFC (Minimum fungicidal concentration) values while the lowest concentration that prevented the growth of the tested fungus was taken as the minimum inhibitory concentration (MIC).

2.8 Statistical Analysis

All statistical analyses were computed using the program R 3.2.2 (R Development Core Team, 2017), using agricolae and nlstools packages. Data were analysed independently using three-way ANOVA with Extract, fungi isolate, concentration and plant as fixed factors. Turkey HSD multiple post-hoc tests were used to evaluate the significance of the differences among the means.

3. RESULTS AND DISCUSSION

3.1 Phytochemical Analysis

The results of the qualitative analysis of leave extracts from *B. orellana*, *J.curcas* and *Cassia alata* are presented in Table 1, 2, and 3. In this study, Phytochemical analysis of *B. orellana* leaf extracts revealed the presence of flavonoids, anthraquinones, terpenoid, phenol, steroids, saponin, phytosteroids and cardiac glycoside in varied quantity and absence of phlobatannins, proteins, fixed oil and phytosterons using various solvents (Table 1). Flavonoid was found to be significantly highest in ethanol and hexane extracts but not found in aqueous solvent. Tannin and phytosteroids were present in ethanolic extract but in low quality. Similar to this study, [44,45] reported that the aqueous extract of *Bixa orellana* contain aminoacids, carbohydrates, steroids, proteins, flavonoids, phlobatanins, terpenoids, cardiac glycosides and saponins while the ethanolic extract contained alkaloids, amino acids, carbohydrates, cardiac glycosides, steroids, proteins, and phlobatanins.

From this analysis, the phytochemical compounds present in the various extracts of

Cassia alata extracts include flavonoid, saponin, phenol, steroids, glycoside, phytosteroids, alkaloids, terpenoid, tannin, and cardiac glycoside as presented in Table 2. However, [31] earlier reported that the antimicrobial activity of Cassia alata is associated with the presence of phytochemical components such as phenols, tannins, saponins, alkaloids, steroids, flavonoids and carbohydrates.

In comparison to the other plants studied, the ethanolic extract of *J. curcas* revealed a significant higher quantity of flavonoids and glycosides, moderate quantity of coumarin, quinones & anthraquinones, and low quantity of tannins, terpenoids, phenol, steroids, phytosteroids, alkaloids, carbohydrates, & cardiac glycoside. While phlobatannins, fixed protein, phytosterols, and saponins were absent. Notably, the cold water extract of *J. curcas* showed large quantity of saponins [46] reported that saponin has antifungal properties. Its mode of action is by killing and inhibiting the excessive division of cells. Various researchers have previously reported the presence of these phytochemicals in similarity, with this study.

In a study by [47] the phytochemical screening of *Jatropha curcas* leaf revealed the presence of tannins, saponins, flavonoids, alkaloids, oxalates and cardiac glycosides while [48] reported that root and latex extracts of *J. curcas* plant contained phenolics, flavonoid and saponins. Other phytochemical screening reported by [49] indicated the presence of nine different phytochemicals from *Jatropha curcas* in varying degrees. The major phytochemical compounds from the methanolic and aqueous leaf extracts reported include phenols, flavonoids, and tannins. Significantly, the ethanolic extract of both plant (*Bixa orellana* and *Jatropha curcas*) showed a significant high quantity of flavonoid [50] opined that ethanol is a suitable solvent for the extraction process.

3.2 Antifungal Activities of Plant Extracts

The antifungal activity of the three plants (*Bixa orellana*, *Cassia alata* and *Jatropha curcas*) in varied concentration (12.5 mg/ml, 25 mg/ml, 50 mg/ml, 100 mg/ml and 200 mg/ml) are presented in Figs. (1, 2 and 3). However, the two most effective of the plants are *Bixa orellana* and *Jatropha curcas*. All the concentrations of *B. orellana* examined had significant effect against the isolates, the highest zone of inhibition was recorded with hexane extracts at 200 mg/ml against *T. verrucosum* while the same extract

had no significant effect on *M. gypseum* at 12.5 mg/ml (Fig. 3). The ethanolic extract obtained from *B. orellana* exhibited a zone of inhibition of 24 mm, 26 mm, and 22 mm on *T. ajelloi* at 12.5, 25.0 and 100mg/ml respectively. The *B. orellana* extract showed high zone of inhibition of 24mm, 26mm, 28 mm and 32 mm at 12.5 mg/ml, 50 mg/ml, 100 mg/ml and 200 mg/ml respectively against *T. verrucosum* while the least value 0.2mm at 50 mg/ml was recorded on *T. tonsurans* (Fig. 1, 2, 3). *Bixa orellana* extracts therefore had a zone of inhibition range of 22mm – 32mm against the tested dermatophytes. Similarly, [51] earlier reported an antidermatophytic activity of *Bixa orellana* against *T. rubrum* (12.66 ±1.15 mm), *M. gypseum* (10.33±1.15 mm), *T. tonsurans* (09.33±0.57 mm) and *T. mentagrophytes* (09.00±0.00 mm). Furthermore, [52] reported that the leaf extract of *B. orellana* at 1000 µg/ml concentration showed significant inhibition against all the tested bacteria and fungus, with highest inhibition zone (18±0.3 mm) against *S. typhi*, *Acinetobacter sp.*, *T. mentagrophytes* and *T. rubrum*. Minimum Inhibitory Concentration (MIC) of leaf extract was determined as 15.62 µg/ml - 31.25 µg/ml for the tested organism. Among the dermatophytes, 78.2% inhibition was reported for *T. mentagrophytes* & *T. rubrum*. Similar to our study, [52] reported that both leaf & seed extracts of *B. orellana* exhibited dose dependent antimicrobial activity against tested organism. From our study, *B. orellana* was found to contain saponins & steroids. It was reported that the mode of action of saponins against bacteria is due to its ability to cause leakage of proteins & certain enzymes from the cell [53].

For *Bixa orellana*, no zone of inhibition was recorded for cold water against *T. mentagrophyte*. In concentrations without antifungal activity, it could be that potent antifungal flavonoids didn't diffuse through the paper disc because of low diffusion rate [54,55]. It is desirable to note that the inhibitory activity of plant extract is largely dependent on the concentration, parts of the plant used and the microbes tested [56].

According to [57], researchers have extensively studied the biological properties of *Jatropha curcas* with evidence that the plant is ethno-medically valuable. The antifungal activity of *Jatropha curcas* extract using different solvents at a varied concentration (12.5 mg/ml, 25 mg/ml, 50 mg/ml, 100 mg/ml, and 200 mg/ml) is represented (Figs. 1, 2, and 3).

In this study, ethanol extracts of *Jatropha curcas* was observed to be significantly higher than cold extracts. However, highest mean of zone of inhibition was observed by the said extracts against *T. rubrum*, *T. soudanenses* and *T. tonsurans*. Unlike the result obtained from our study, [58] reported *T. rubrum* was resistant to *Jatropha* methanolic extracts with no reported zone of inhibition. The difference in our result could be due to the difference in the solvent used for the extraction. Different biochemical compounds are present in plant extracts from various solvent. The plants extract test at the different concentrations on fungal isolates exhibited varied degree antifungal activities. *T. soudanenses* and *T. verrucosum* were highly susceptible to all extract of *Jatropha curcas* used with a different activity. Ethanol extract inhibited *T. metagrophytes* and *M. gypseum* at 12.5mg/ml concentration. The diameter zone of clearance of ethanolic extract at 200mg/ml was higher when compared to other concentrations used. Hence, it could be said that the antifungal activity of the plant is concentration dependent. The high zones of inhibition (31mm and 30mm) of hexane and ethanolic extract against *T. soudanenses* and *T. verrucosum* were observed and recorded. Several previous studies are in agreement with this present study. [42] reported that the ethanolic extract of the stem bark of *J. curcas* inhibited *B. subtilis*, *E. coli*, *P. vulgaris* while [59] reported the inhibitory ability of the methanolic extract of in vivo leaves and in vitro derived callus (30 days old) of *J. curcas*.

Furthermore, [60] showed that the latex of *Jatropha curcas* had good antifungal activity against *Candida albicans* and *Tricophyton sp.* [43] reported that the zones of inhibition exhibited by the extracts against the test fungal species ranged between 0 and 20mm. Relatively, [56] indicated that the ethanolic extracts of *Jatropha curcas* inhibited pathogenic bacteria with zones of inhibition ranging from 30.6 to 38.5 mm. In a study by [60] the diameter of zones of inhibition reported ranged between 20 to 26 mm while it was reported that the methanolic extracts of the *Jatropha curcas* seed kernel showed the maximum zone of inhibition of 25.5mm [57].

The inherent ability of the *J. curcas* extracts to inhibit the growth of several bacterial and fungal revealed the broad spectrum antimicrobial potential of *J. curcas*, which enlist the plant as candidate for bioprospecting for antibiotic and antifungal drugs [42,61].

Some factors could affect the inhibitory activity of plant extracts which include the concentration, the accumulation and concentration of secondary metabolites which are responsible for inhibitory activity is varied according the plant parts [62]. This may be causal for the variation in the inhibitory activity of extracts of *J. curcas* [59]. The susceptibility of these fungi to *J. curcas* extracts is significant, because fungi have recently been implicated in cases of immuno-compromised patients who frequently develop opportunistic infections [63].

Table 1. Results of qualitative phytochemical analysis of *Bixa orellana* Leaves

Test	Ethanol	Hexane	Cold water	Hot water
Flavonoids	+++	+++	-	-
Tannins	+	-	-	-
Saponins	+	+	+	+
Quinones	+	++	-	-
Anthraquinones	++	+	+	++
Terpenoids	+	++	++	+
Coumarin	-	+	-	+++
Phenol	++	+	++	++
Steroids	+	++	++	++
Phytosteroids	+	-	-	-
Phytosterons	-	-	-	-
Glycosides	++	+	+	-
Alkaloids	+	-	-	+
Carbohydrates	+	-	+	-
Phlobatannins	+	-	+	-
Proteins	+	-	+	-
Fixed oil	-	-	-	-
Cardiac glycoside	+	-	+	++

- absent; + present in small quantity; ++ moderately present
+++ present in large quantity

Table 2. Results of phytochemical analysis of *Casia alata* leaves

Test	Ethanol	Hexane	Cold water	Hot water
Flavonoids	+++	++	+++	+++
Tannins	+	-	-	-
Saponins	+++	+	+	+
Quinones	-	++	-	-
Anthraquinones	+++	-	+	-
Terpenoids	+	-	+	+++
Coumarin	+++	-	++	+
Phenol	+	+	+	-
Steroids	+	+	+	-
Phytosteroids	+	+	-	-
Phytosterols	-	-	-	-
Glycosides	+	++	+	-
Alkaloids	+	-	+	+
Carbohydrates	+	-	+	-
Phlobatannins	-	-	-	-
Proteins	-	-	-	-
Fixed oil	-	-	-	-
Cardiac glycoside	+	-	-	-

- absent

+ present in small quantity

++ moderately present

+++ present in large quantity

Table 3. Results of qualitative phytochemical analysis of *Jatropha curcas*

Test	Ethanol	Hexane	Cold water	Hot water
Flavonoids	+++	+	-	-
Tannins	+	-	-	-
Saponins	+	-	+++	+
Quinones	++	+	+	+
Anthraquinones	++	-	-	-
Terpenoids	+	+	-	-
Coumarin	++	-	+	+
Phenol	+	-	+	+
Steroids	+	++	+	+
Phytosteroids	+	+	-	-
Phytosterols	-	+	+	+
Glycosides	+++	++	+	+
Alkaloids	+	++	-	-
Carbohydrates	+	+	+	+
Phlobatannins	-	-	-	-
Proteins	-	+	-	-
Fixed oil	-	-	-	-
Cardiac glycoside	+	+	-	-

- absent

+ present in small quantity

++ moderately present

+++ present in large quantity

M. gypseum was observed to be highly resistant to all the concentrations of *J. curcas* extracts, *T. mentagrophyte* was recorded to be susceptible to hot water and ethanol extracts in all concentration while all extracts at 200 mg/ml had the highest potency against *T. verrucosum* (Fig.

3). Thus, from the result from our study, *Jatropha curcas* is more potent against the majority of the tested organisms than *Bixa orellana* at similar concentration. It was evidently active against *T. rubrum*, *T. soudanenses*, *T. tonsurans*, *T. verrucosum*, and *T. megatrophites* which totally

resistant to *Bixa orellana* extracts with the highest zone of inhibition of 31mm. *T. ajellio* and *T. verrucosum* were the two mainly susceptible organisms to *Bixa orellana* extracts in this study with the highest zone of inhibition of 24mm.

For *Cassia alata*, various degree zone of inhibition was recorded on the fungal isolates using the plant extract at different concentrations. Hexane extract had the highest zone of inhibition against *T. soudanenses*, *M. gypseum*, *T. rubrum* and *T. tonsurans* while no zone of inhibition was recorded for ethanol and water extracts against *T. soudanenses*, *T. mentagrophyte* and *T. ajelloi*, respectively.

In agreement with our study, [64] reported that the ethanol extract of the *Cassia alata* showed high activity against fungi: *Trichophyton mentagrophytes* var *interdigitale*, *T. mentagrophytes* var. *mentagrophytes*, and *T. rubrum*. Similarly, [65,66] also reported that the aqueous extract of *C. alata* had antifungal effect on *A. flavus*, *A. parasiticus*, *F. oxysporum* and *C. albicans*.

Furthermore, [67] observed that the ethanol extracts of *C. alata* leaves exhibited high activity against dermatophytic diseases caused by *Rhizopus* spp, *P. oxalicum*, *A. tamari*, *A. niger*, *F. oxysporum* and *F. vacitilus*. According to [68], all the crude methanol and aqueous extract of the leaves inhibited the growth of *C. albicans*, *T. mentagrophytes*, *A. niger* as indicated by the zones of inhibition but show no activity against *Penicillium*.

In concordance with our study, identified *Jatropha curcas* as the most effective among the extracts studied particularly against *A. niger*. Similarly, [41] reported that *J. curcas* leaf extract when compared to other plants was more active against isolated microbes and in particular *Pseudomonas aeruginosa* known to be resistant to most synthetic antibiotics. The higher microbial activity of *J. curcas* may be due to its phytochemical compositions. The difference in the antifungal activities of the studied plants could be due to the variation in the phytochemical compounds earlier reported above.

The Minimum Inhibitory Concentrations (MIC) values of the *Bixa orellana*, *Cassia alata* and *Jatropha curcas* as shown in Fig. 4. The observed result on the minimum inhibitory concentration (MIC) of the extract agreed with

the report that organisms varied widely in the degree of their susceptibility [69,70]. From our findings, *J. curcas* has the lowest MIC, followed by *B. orellana* while the MIC value in *C. alata* was found to be highest. Ethanol extract of *J. curcas* had the lowest MIC value (12.5 mg/ml) for all the isolates with the exemption of *M. gypseum* which MIC value is 100 mg/ml.

[56] reported that the ethanolic extracts of *Jatropha curcas* inhibited pathogenic bacteria with the minimum inhibitory concentration (MIC) ranging from 2.2 to 10.0 mg/ml. In a study by [58], the minimum inhibitory concentration of 1.25-10 mg/ml and minimum fungicidal concentration of 2.5-20 mg/ml were reported from organisms which exhibited different degree of susceptibility to the inhibitory activity of the crude extracts. Similarly, [71] reported that the antibacterial activity of *Jatropha curcas* was also found to be effective at MIC value of 10mg/ml. Furthermore, [61] reported that the extracts of *Jatropha curcas* showed broad antifungal activity against the tested fungal isolates at a final concentration of 10 mg/ml. According to [56], the ability of the ethanolic extracts of the leaf and bark of *J. curcas* to inhibit growth of the test organisms is an indication of its antimicrobial potency which may be employed in treatment of microbial infections. Furthermore, [43] reported that the ethanolic extract of *Jatropha curcas* leaves have antidermatophytic activities with a minimum inhibitory concentration (MIC) between 19.95 and 79.43 mg/ml. The minimum inhibitory concentration of 30.20mg/ml was reported against *Trichophyton* spp. However, [69] reported that the ethanolic extract of *Jatropha curcas* had the lowest concentration of 0.5ml that reduced mycelia growth, while the other extracts had reduction of mycelia growth at 1.0ml and 2.0 ml respectively.

The MIC readings for *T. soudanenses* and *T. tonsurans* exposed to cold water of *B. orellana* is 100 mg/ml; *T. mentagrophyte* and *T. rubrum* exposed to cold water extract of *C. alata* is 100 mg/ml; *T. rubrum* and *M. gypseum* exposed to ethanol extract of *C. alata* is 100 mg/ml (Fig. 4).

This study reviewed that ethanol extract of the *Jatropha curcas* showed high activity of 12.5 mg/ml against all the fungi isolate tested except in *M. gypseum* that as a 100mg/ml. This also is similar to a case of *Trichophyton mentagrophytes* var *interdigitale*, *T. mentagrophytes* var. *mentagrophytes*, *T. rubrum* and *Microsporium gypseum* activity of MIC 12.5 mg/ml according to [64].

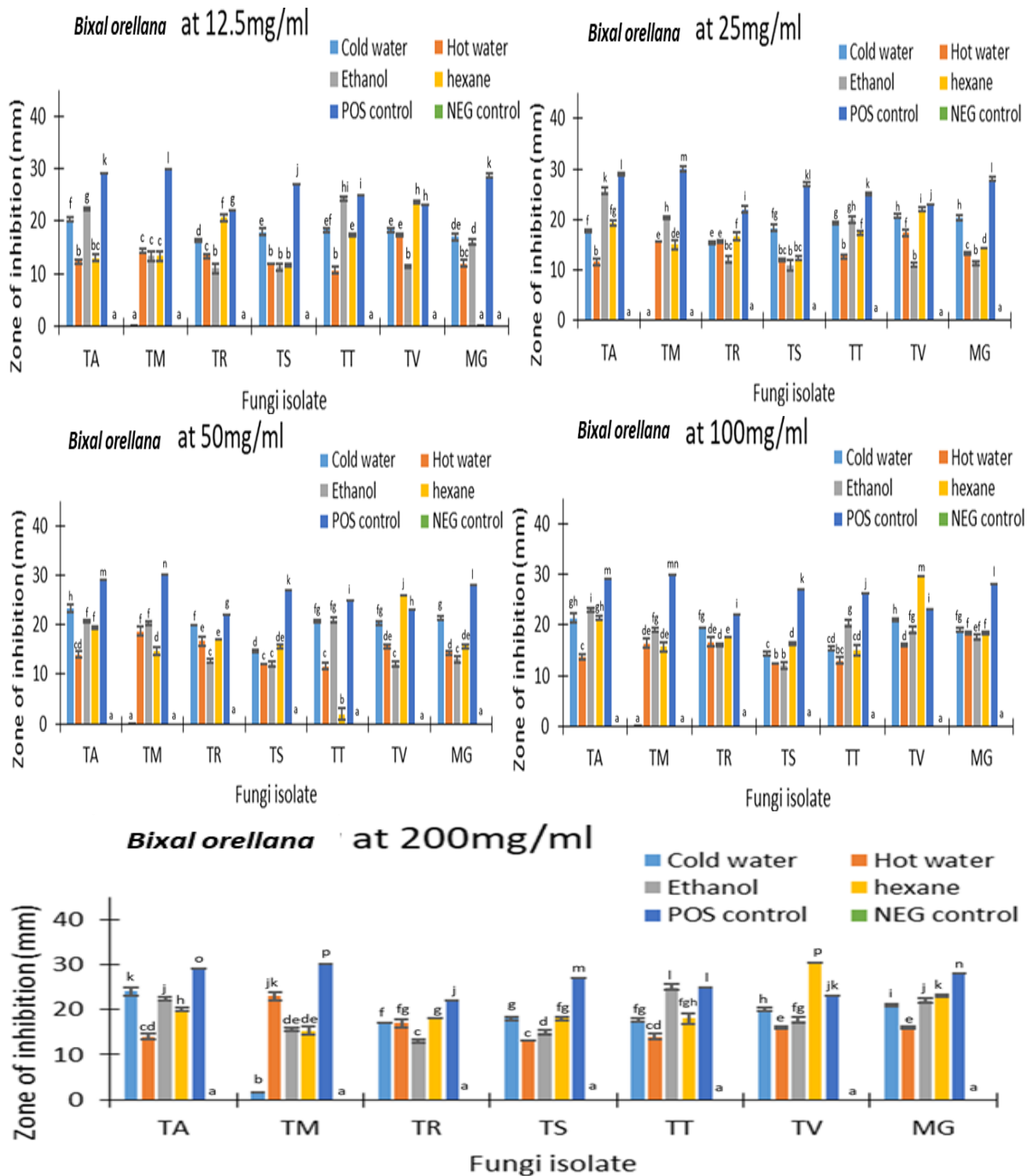


Fig. 1. Antifungal activity of B. orellana leaf extracts using different solvents at 12.5, 25 50, 100 and 200mg/ml

Data represents mean of the triplicate data and error bar represent standard error of the mean. (Different alphabet indicates significant difference, while $p = 0.05$). TA – *T. ajelloi*, TM - *T. metagrophytes*, TR - *T. rubrum*, TS - *T. soudanese*, TT - *T. tonsurans*, TV - *T. verrucosum*, MG - *M. gypseum*

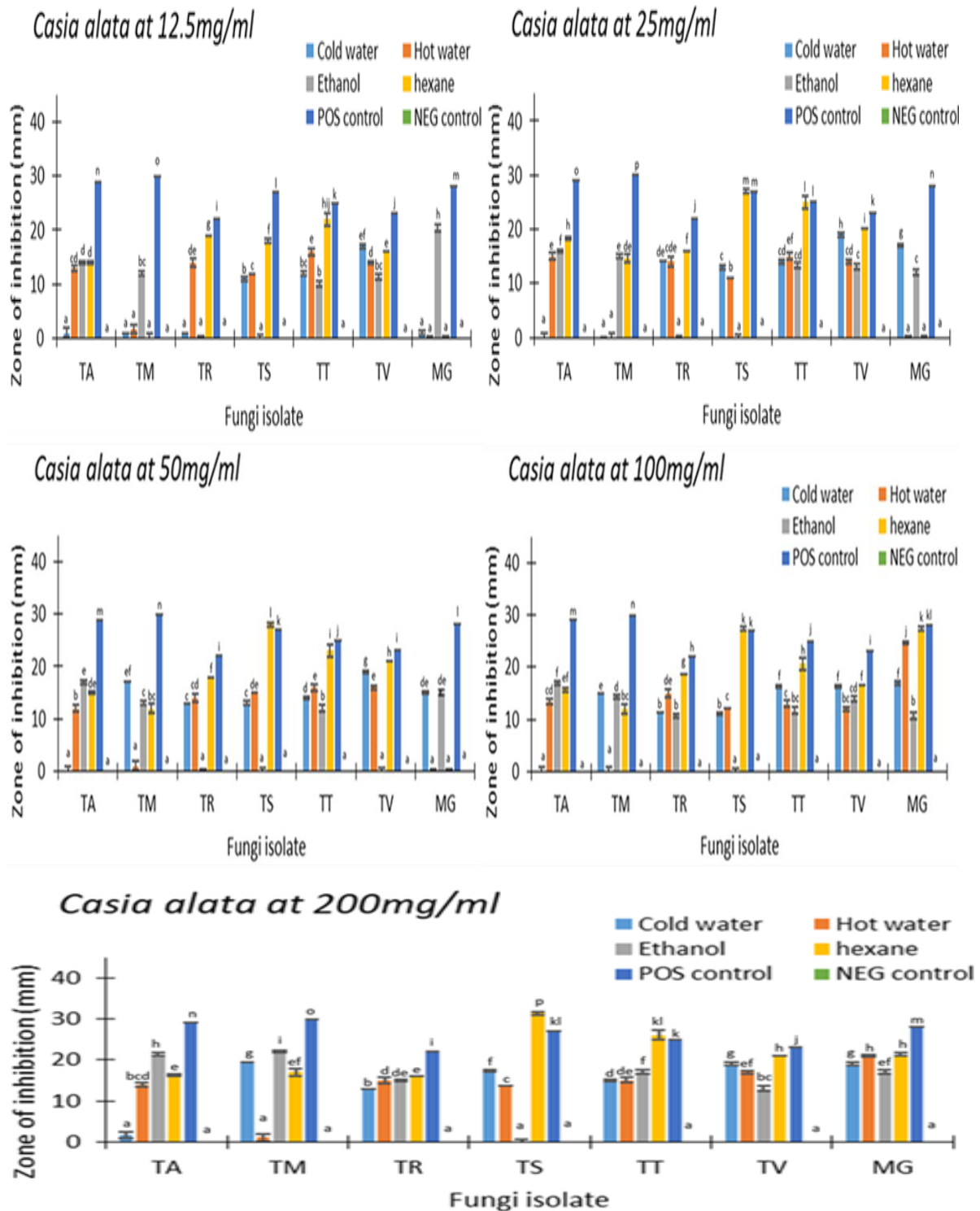


Fig. 2. Antifungal activity of *Casia alata* leaf extracts using different solvents at 12.5, 25, 50, 100 and 200 mg/ml

Data represents mean of the triplicate data and error bar represent standard error of the mean. (Different alphabet indicates significant difference, while $p = 0.05$).

TA – *T. ajelloi*, TM - *T. metagrophytes*, TR - *T. rubrum*, TS - *T. soudanese*, TT - *T. tonsurans*, TV - *T. verrucosum*, MG - *M. gypseum*

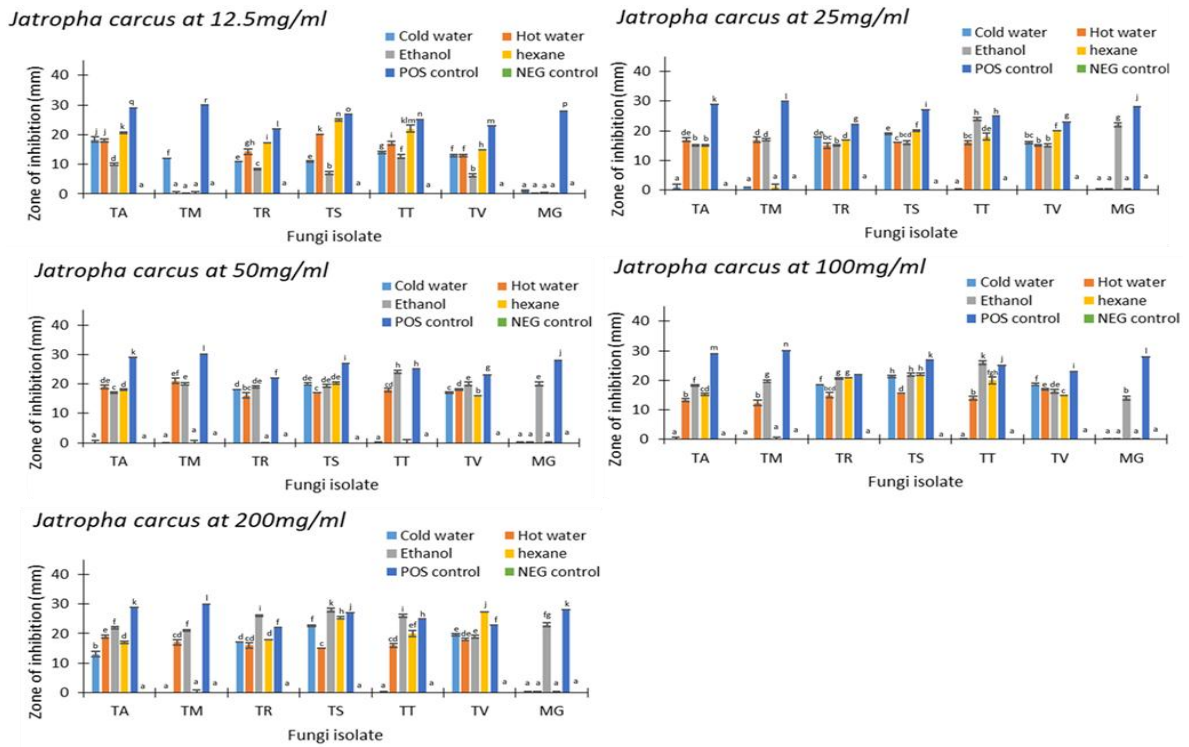


Fig. 3. Antifungal activity of J. curcas leaf extracts using different solvents at 12.5, 25, 50, 100 and 200mg/ml

Data represents mean of the triplicate data and error bar represent standard error of the mean. (Different alphabet indicates significant difference, while $p = 0.05$). TA – *T. ajelloi*, TM - *T. metagrophytes*, TR - *T. rubrum*, TS - *T. soudanese*, TT - *T. tonsurans*, TV - *T. verrucosum*, MG - *M. gypseum*

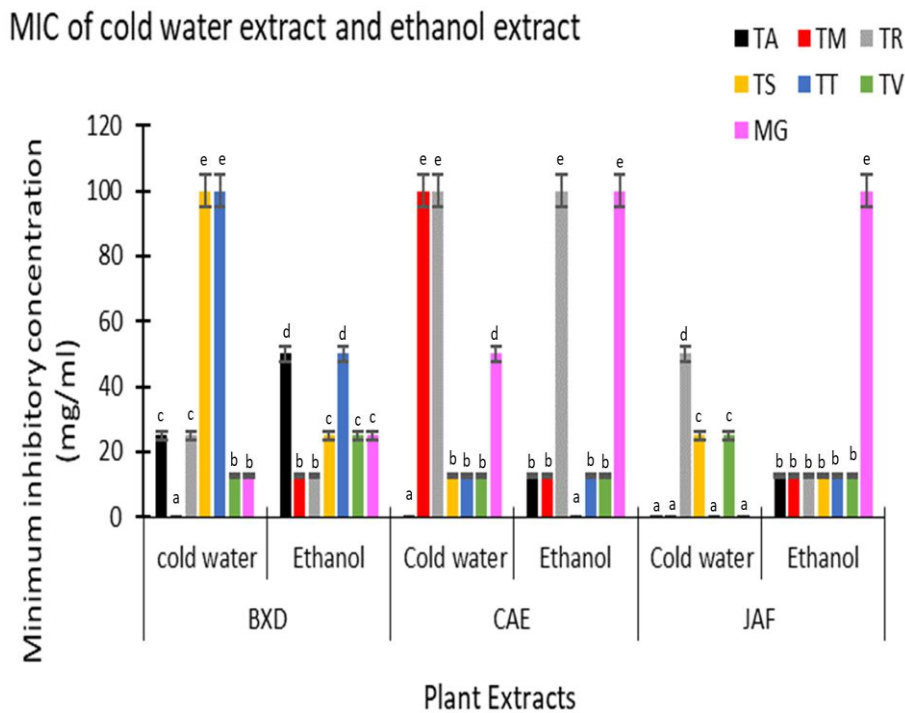


Fig. 4. Minimum inhibitory concentration of cold water and ethanol extracts of the plant leaves

Table 4. Minimum Fungicidal Concentration (MFC) of the Leaf Extracts on the fungal isolates

Isolates	<i>B. orellana</i>		<i>C. alata</i>		<i>J. curcas</i>	
	CW (mg/ml)	ET (mg/ml)	CW (mg/ml)	ET (mg/ml)	CW (mg/ml)	ET (mg/ml)
<i>T. ajelloi</i>	50	50	-	50	25	25
<i>T. mentagrophyte</i>	-	25	100	50	25	200
<i>T. rubrum</i>	50	25	25	200	25	50
<i>T. soudanenses</i>	200	50	50	-	50	25
<i>T. tonsurans</i>	25	50	25	50	25	25
<i>T. verrucosum</i>	25	100	50	25	50	50
<i>M. gypseum</i>	100	25	50	200	50	50

The result of the Minimum Fungicidal Concentration (MFC) is presented in Table 4. The two plants were screened for MFC against the isolates, cold water extract of *B. orellana* gave an averagely low MFC against *T. tonsurans* and *T. verrucosum* of 25 mg/ml and high MFC against *M. gypseum* and *T. soudanenses* of 100 and 200 mg/ml respectively. Ethanol extract of *B. orellana* had averagely strong MFC of 25 mg/ml against *T. mentagrophyte*, *T. rubrum*, *M. gypseum* and averagely weak against others. Cold water extract of *J. curcas* gave the better MFC results. These results were ranging between 25 mg/ml to 50 mg/ml with 4 isolates giving MFC of 25 mg/ml. Cold water extract of *J. curcas* was therefore the most potent and active extract amongst the tested extracts.

4. CONCLUSION

In our study, we determined and compared the phytochemical compounds, antimicrobial properties and the MIC/ MFC of *Bixa orellana*, *Jatropha curcas* and *Cassia alata* against seven dermatophytes. The phytochemical compounds present in the plants include anthraquinones, terpenoid, phenol, flavonoids, steroids, saponin, and others in different concentrations, the plant extracts were effective against at least three of the seven tested dermatophytes with a zone of inhibition range of 22mm-32mm. The ethanolic extracts *Jatropha curcas* showed broad effectiveness against the tested pathogens when compared to *Bixa orellana* and *Cassia alata*. The minimum inhibition concentration recorded for the plants extracts is 12.5mg/ml and we conclude that the plants antifungal property is concentration dependent. The comparative study of medicinal plants will provide evidence for the selective usage of these natural antidotes to pathogenic infection. Hence, we recommend that further studies should be done with these plants extracts using a large number of different

isolates. Also the ethanolic extract of the plants proved to be effective against all the isolate.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country.

CONSENT

All authors declare that 'written informed consent was obtained from the patient (or other approved parties) for publication of this case report and accompanying images.

ETHICAL APPROVAL

As per international standard or university standard written ethical approval has been collected and preserved by the author(s).

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Aliyu AB, Ibrahim MA, Ibrahim H, Dambatta MB, Oyewale AO. GC-MS Analysis of *Pavetta corymbosa* Lipophilic Extract and Its Antimicrobial Activity. *Ife Journal of Science*. 2017;2(19).
2. Okigbo RN, Mmeka EC. An Appraisal of Phytomedicine in Africa. *KMITL Science and Technology Journal*. 2006;6(2).
3. United States Department of Agriculture; 2019.
4. Ventosa E. *Bixa orellana* (annatto). Invasive Species Compendium. Wallingford, UK: CABI; 2018. DOI:10.1079/ISC.9242.20203482896

5. Julia J, Shen Maiken C, Arendrup Gregor BE, Jemec, Ditte Marie L, Saunte. Photodiagnosis and Photodynamic Therapy. 2018;102- 169.
6. Vilar DA, Vilar MS, Moura TF, Raffin FN, Oliveira M., Oliveira F., Petrônio F., Margareth F., Formiga M, José Maria B. Traditional Uses, Chemical Constituents, and Biological Activities of *Bixa orellana* L.: A Review; 2014.
7. Fleischer TC, Ameade EK, Mensah ML, Sawyer IK. Antimicrobial activity of the leaves and seeds of *Bixa orellana*. *Fitoterapia*. 2003;74(1–2):136–138.
8. Adamu H.M, Abayeh OJ, Ibok NU, Katsu SE. Antifungal activity of extracts of some *Cassia*, *Detarium* and *Ziziphus* species against dermatophytes. *Nat Prod Rad*. 2006;5:357–60.
9. Ravindranath N, Ravinder RM, Mahender G, Ramu R, Ravi KK, Das B. Deoxypreussomerins from *Jatropha curcas*: are they also plant metabolites? *Phytochemistry*. 2004;65:2387–2390
10. Matsuse IT, Lim YA, Hattori, M., Correa, M, Gupta MP. A search for anti-viral properties in Panamanian medicinal plants: The effects on HIV and its essential enzymes. *Journal of Ethnopharmacology*, 1998;64:15–22
11. Dimri, M, Sharma, C. Effects of sarcoptic mange and its control with oil of *Cedrus deodara*, *Pongamia glabra*, *Jatropha curcas* and benzyl benzoate, both with and without ascorbic acid on growing sheep: epidemiology; assessment of clinical, haematological, cell-mediated humoral immune responses and pathology. *Journal of Veterinary Medicine Series A*. 2004;51:71–78
12. Jing L, Fang Y, Ying X, Wenxing H, Meng X, Syed MN, Fang C. Toxic impact of ingested *Jatropherol-I* on selected enzymatic activities and the ultrastructure of midgut cells in silkworm, *Bombyx mori* L. *Journal of Applied Entomology*, 2005;129:98–104
13. Georges K, Jayaprakasam B, Dalavoy SS, Nair MG. Pest-managing activities of plant extracts and anthraquinones from *Cassia nigricans* from Burkina Faso. *Bioresource Technology*. 2008; 99:2037–2045
14. Osoniyi O, Onajobi F. Coagulant and anticoagulant activities in *Jatropha curcas* latex. *Journal of Ethnopharmacology*, 2003;89:101–105
15. Rug M, Ruppel A. Toxic activities of the plant *Jatropha curcas* against intermediate snail hosts and larvae of schistosomes. *Tropical Medicine and International Health*, 2000;5:423–430
16. Mujumdar V, Misar AV. Anti-inflammatory activity of *Jatropha curcas* roots in mice and rats. *Ethnopharmacol*. 2004;90(1):11-5.
17. Shetty S, Udupa SL, Udupa AL, Vollala VR. Wound healing activities of Bark Extract of *Jatropha curcas* Linn in albino rats. *Saudi. Med. J*. 2006;27(10):1473-6.
18. Mishra S, Vijayakumar M, Ojha S, Verma A. Antidiabetic effect of *Jatropha curcas* L leaves extract in normal and alloxan-induced diabetic rats. *Int J Biomed Sci*.2000; 2:482–487
19. Abd-Alla HI, Moharram FA, Gaara AH, ElSafty MM. Phytoconstituents of *Jatropha curcas* L leaves and their immunomodulatory activity on humoral and cell-mediated immune response in chick ;*Z Naturforsch* 2009;C64:7:8:495-501.
20. Hennebelle T, Weniger B, Joseph H, Sahpaz S. *Senna alata*. *Fitoterapia*. 2009;80(7):385-93
21. Meenupriya J, Vinisha Sahaya A, Priya P. *Cassia alata* and *Cassia auriculata* – Review of their bioactive potential. *World Journal of Pharmaceutical Sciences*; 2014. ISSN:(print) 2321-3310
22. Oke DG, Oluranti OO, Akande JA. Preliminary studies on the laxative properties of *Senna alata* L. and *Hollandia yoghurt*. *African Journal of Biotechnology*. 2018;17(13):429-432.
23. Srinivasan N. Pharmacognostical and phytochemical evaluation of *Cassia alata* Linn. *Journal of Medicinal Plants Studies*. 2018;6(5):69-77.
24. Adnaik RS, Bhagwat DA, Raut ID, Mohite SK, Magdum CS. Laxative and Anthelmintic Potential of *Cassia alata* Flower Extract. *Research J. Pharm. and Tech*. 2011;4(1):98-100.
25. Midawa SM, Ali BD, Mshelia BZ. “Cutaneous wound healing activity of the ethanolic extracts of the leaf of *Senna alata* L (Caesalpiniaceae). *Journal of Biological Science and Bioconservation*, 2010; 2.
26. El-mahmood AM, Doughari JH. Phytochemical screening and antibacterial evaluation of the leaf and root extracts of *Cassia alata* Linn. *African Journal of*

- Pharmacy and Pharmacology. 2008; 2(7): 124-9.
27. Suman K, Saptarshi R, Larisha M. Cassia alata L: potential role as anthelmintic agent against Hymenolepis diminuta. Parasitology Research. 2012;111(3):1187-1192.
 28. Villasenor IM, Sanchez AC. Cassiaindoline, a new analgesic and anti-inflammatory alkaloid from Cassia alata. zeitschrift für naturforschung C. 2009;64(5,6):335-338.
 29. Yakubu MT, Adeshina AO, Oladiji AT, Akanji MA, Oloyede OB, Jimoh GA. Abortifacient Potential of Aqueous Extract of Senna alata Leaves in Rats. Journal of Reproduction & Contraception. 2010;21(3):163-177.
 30. Jarinyaporn, Supaporn. Antilipogenic effect of Senna alata leaf extract in high-fat diet-induced obese mice. Asian Pacific Journal of Tropical Biomedicine. 2016;6(3):232-238.
 31. Makinde AA, Igoli J.O., Ta'ama L., Shaibu S.S., Garba A., Antimicrobial activity of Cassia alata. African Journal of Biotechnology. 2007; 6(13):1509-1510.
 32. Igoli JO, Ogaji OG, Igoli NP, Tor-Anyiin TA. Traditional medicinal practices among the Igede people of Nigeria (part II). African Journal of Traditional Compliment Alternative Medicine. 2005; 2(2): 134-152.
 33. Falodun A, Okunrobo LO, Uzoamaka N. Phytochemical screening and anti-inflammatory evaluation of methanolic and aqueous extracts of Euphorbia heterophylla Linn (Euphorbiaceae). African Journal of Biotechnology. 2006;5(6):529- 530.
 34. Meena R, Ramaswamy RS. Herbs for combatting dermatophytosis- a review. International Journal of Plant. 2014;1(6):373-79.
 35. Colin VL, Mario Domingo Baigorí MD, Licia María Pera LM. Tailoring fungal morphology of Aspergillus niger MYA 135 by altering the hyphal morphology and the conidia adhesion capacity: biotechnological applications. A.M.B. Express, 2013;3:27.
 36. Olutiola, P.O., O. Famurewa O. and Sonntag H.G., (2000). Introduction to General Microbiology: A Practical Approach. 2nd Edn., Bolabay Publications, Ikeja, Nigeria.
 37. Liu D, Coloe S, Baird R, Pedersen J. Rapid mini-preparation of fungal DNA for PCR. J. Clin. Microbiol. 2000;38:471.
 38. Ahmed S, Bin RF, Ahmed S, Mohammad SA. Insights into the bioactive compounds, antioxidant potential and TLC profiling of different extracts of Tomato plants. Jahangirnagar Univ. J. Biol. Sci. 2018;7(2):65–77
 39. Sofowora A. *Medicinal Plants and Traditional Medicinal in Africa*. 5th Ed. Sunshine House, Ibadan, Nigeria: Spectrum Books Ltd;. Screening Plants for Bioactive Agents. 2006;134–156
 40. Rizk AM. Constituents of plants growing in Qatar. 1. A chemical survey of sixty plants. Fitoterapia. 1982;53:35–44
 41. The Clinical and Laboratory Standards Institute (CLSI) Wayne, Pennsylvania, USA; 2012.
 42. Akinpelu DA, Kolawole DO. Phytochemical and antimicrobial activity of leaf extract of Piliostigma thonningii (Schum.). Sc. Focus. 2004;7: 64–70.
 43. Igbinosa OO, Igbinosa, EO, Aiyegoro OA. Antimicrobial activity and phytochemical screening of stem bark extracts from Jatropha curcas (Linn). African Journal of Pharmacy and Pharmacology. 2009;3(2):058-062
 44. Mbakwem-Aniebo C, Onianwa O, Okonko O. Effects of Ficus Exasperata Vahl on Common Dermatophytes and Causative Agent of Pityriasis Versicolor in Rivers State, Nigeria. American Journal of Dermatology and Venereology. 2012; 1(1):1-5
 45. Sangvikar S, Malgaonkar M, Sharma C, Kumar S, Murthy SN. comparative phytochemical screening of qualitative and quantitative parameters of Bixa orellana . World Journal of Pharmacy and Pharmaceutical Sciences. 2015;4(12):1001-1017.
 46. Prathima D, Sujitha A, Usha R. Phytochemical Screening and Antimicrobial Activity of Bixa Orellana Linn; 2016.
 47. Ugwuoke, Ukwueze. Efficacy of Some Leaf Extracts on Some Seed Borne Fungi of African Yam Bean (Sphenostylis stenocarpa Hochest Ex. A. Rich). Bio-Research, 2010;8(1):638 – 645.
 48. Oyama MO, Malachi OI, Oladejo AA. Phytochemical Screening and Antimicrobial Activity of Leaf Extract of

- Jatropha curcas. Journal of Advances in Medical and Pharmaceutical Sciences 2016;8(1):1-6.
49. Oskoueian E, Norhani A, Syahida A, Wan ZS, Abdul RO, Yin WH. Bioactive Compounds and Biological Activities of Jatropha curcas L. Kernel Meal Extract International Journal of Molecular Sciences 2011b;12:5955-5970
 50. Ahmed S, Bilkis MM, Ahmed S, Gomes JD, Abdullah MS. Comparative phytochemical, antioxidant, and antibacterial study of different parts of Doigota plants (*Bixa orellana* L.) Bulletin of the National Research Centre. 2020;44:9
 51. Obi VI, Onuoha C. Extraction and Characterisation Methods of Plants and Plant Products. In: Biological and Agricultural Techniques. Ogbulie, J. N. And Ojiaku, O. J. (eds). Websmedia Publication, Owerri. 2000;271–286.
 52. Vidyasagar. Antidermatophytic activity of young leaves methanolic extract of *Bixa orellana*. World Journal of Pharmaceutical and Medical Research; 2017.
 53. Tamil Selvi A, Dinesh MG, Satyan RS, Chandrasekaran B, Rose C. Leaf and Seed extracts of *Bixa orellana* L. exert antimicrobial activity against bacterial pathogens. Journal of Applied Pharmaceutical Science. 2011;01(09);116-120.
 54. Vila J, Marco F, Soler L, Chacon M, Figueras MJ. In vitro antimicrobial susceptibility of clinical isolates of *Aeromonas caviae*, *Aeromonas hydrophila* and *Aeromonas veronii* biotype *sobria*. Journal of Antimicrobial Chemotherapy. 2002;49(4):701-2.
 55. Namuli AN, Abdullah\ CC, Sieo Zuhainis SW, Oskoueian E. Phytochemical compounds and antibacterial activity of *Jatropha curcas* Linn. Extracts. Journal of Medicinal Plants Research. 2011;5(16):3982-3990
 56. Cushnie TP, Lamb AJ. Antimicrobial activity of flavonoids International Journal of Antimicrobial Agents. 2005;26(5):343-56.
 57. Ekundayo FO, Adeboye, Ekundayo EA. Antimicrobial activities and phytochemical screening of pignut (*Jatropha curcas* Linn.) on some pathogenic bacteria. Journal of Medicinal Plants Research 2011; 5(7):1261-1264
 58. Singh B, JR Nadkarni JR, Vishwakarma RA., Bharate SB., Nivsarkar M, Anandjiwala S. “The hydroalcoholic extract of *Cassia alata* (Linn.) leaves and its major compound rhein exhibits antiallergic activity via mast cell stabilization and lipoxygenase inhibition,” Journal of Ethnopharmacology. 2012;141(1):469–473.
 59. Egharevba HO, and Kunle OF. Broad Spectrum Antimicrobial Activity of Extracts of *Jatropha curcas*. J. App. Pharm. Sci., 2013;3(04):083-087 Broad Spectrum Antimicrobial Activity of Extracts of *Jatropha curcas*. Journal of Applied Pharmaceutical Science 3 (04);2013: 083-087
 60. Kalimuthu KS, Vijayakumar R, Senthil kumar. Antimicrobial Activity of the Biodiesel Plant, *Jatropha Curcas* L. International Journal of Pharma and Bio Sciences. 2010;1(3):1-5
 61. Oyi AR, Onaolapo JA, Haruna AK, Morah CO. Antimicrobial Screening and Stability Studies Of The Crude Extract Of *Jatropha Curcas* Linn Latex (Euphorbiaceae). Nigerian Journal of Pharmaceutical Sciences 2007;6(2):14-20
 62. Sarin R, Manvi M, Sapna B. Evaluation of antibacterial potential of stem bark of *M. oleifera* Lam. Biosc. 2010;1:89–94.
 63. Essawi T, Srouers M. Screening some Palestinian medicinal plants for antibacterial activity. J. Ethnopharmacol. 2000;70:343-349.
 64. Portillo A, Vila R, Freixa B, Adzet T, Canigueral S Antifungal activity of Paraguayan plant used in traditional medicine. J. Ethnopharmacol. 2001;76: 93-98.
 65. Ibrahim D, Osman H. Antimicrobial activity of *Cassia alata* from Malaysia. J. Ethnopharmacol. 1995;45(3):151-6.
 66. Abubakar MN, Ramanathan R, Senthil KT. In vitro antifungal activities of *Cassia alata* Linn. Flower extract. Natural Product Radiance. 2008;7(I): 6-9.
 67. Abubakar I, Mann A, Mathew J. Phytochemical composition, antioxidant and anti-nutritional properties of root-bark and leaf methanol extracts of *Senna alata* L. grown in Nigeria. African Journal of Pure and Applied Chemistry. 2015;9:91-97.
 68. Odunbaku OA, Lusanya OA. Synergistic effect of ethanol leaf extract of *Senna alata* and antimicrobial drugs on some pathogenic microbes. Adv. Environ. Biol. 2011;5:2162-2165.

69. Arekemase MO, Kayode MO, Ajiboye AE. Antimicrobial Activity and Phytochemical Analysis of *Jatropha curcas* Plant against Some Selected Microorganisms. *International Journal of Biology* 2011;3(3):52-598.
70. Willey JM, LM. Sherwood, Woolverton CJ. Prescott, Harley, and Klein's Microbiology. New York, McGraw Hill, 7th Ed. 2008;250-276.
71. Sangeetha JK. Divya MV, Prashanth A, Vamsikrishna G, Leela Rani. Anti inflammatory and Antibacterial Activity of *Jatropha curcas* Linn. *J.P.R.H.C.* 20209;2(3): 258-262.

© 2021 Folorunso et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:

The peer review history for this paper can be accessed here:
<https://www.sdiarticle4.com/review-history/75588>