

IN- VITRO EVALUATION OF ANTIFUNGAL EFFECTS OF *EUPHORBIA ABYSSINICA*
(DESERT CANDLE) EXTRACT

Running Title: anti-fungal effects of *Euphorbia abyssinica* extracts

ABSTRACT

Aim: This study was to evaluate extracts of *Euphorbia abyssinica* a cactus-like plant of the family *Euphorbiaceae* commonly called desert candle or Kechieh bih by the kendem people of south-West Cameroon, for anti-fungal activity.

Study Design: The Randomized Complete Block Design was used in the study, and the means that were significantly different were separated using Duncan's New Multiple Range Test.

Place and Duration of Study: This study was carried out in the Department of Microbiology, University of Nigeria Nsukka, Enugu State, Nigeria, between April 2011 and August 2012.

Methodology: The stem-bark and latex of this plant were extracted using 50%-methanol, water and absolute methanol. Antifungal effect of the extracts was evaluated using agar, and macro-broth dilution methods as well as the Time kill assay.

Results: Strains of *Aspegillus*, *Rhizopus*, *Mucor*, *Rodotorula*, *Geotricum*, *Basidiobolus*, *Trichophyton*, *Microsporum*, *Epidermophyton* and *Candida* species were used as test fungi for the study. The extraction of the stem-bark yielded 18%, 15% and 25% for absolute methanol, water and 50% methanol, respectively, while the latex yielded 13%, 12% and 15% for absolute methanol, water and 50% methanol extracts respectively. The growth inhibition of the fungi tested lasted for 7-14 or more days of incubation on Sabouraud dextrose agar plates. MIC range was 0.39-50.0mg/ml for the stem bark extracts and 1.95-50.0 for the latex extracts. There was a significant difference in the growth inhibition by the 50% methanol extracts of the stem-bark and latex ($P=0.5$) with significant means of 5.361 and 7.1086 respectively. *Candida albicans* was the most susceptible of the yeasts tested (MIC₉₀ 0.313mg/ml) and significant mean of, 0.896 and *M. gypseum* the least susceptible of the dermatophytes tested (significant mean 14.641). In the time kill assay, the results showed that *T. mentagrophytes*, *M. gypseum* and *E. floccosum* cells were killed by the higher concentrations (4MIC and 2MIC) of the plant extracts.

Conclusion: The plant extracts showed broad spectrum of activity against a fungi tested. Results authenticated the ethno-medicinal applications of *Euphorbia abyssinica* for the treatment of skin infections.

Key words: evaluation, antifungal, effects, *Euphorbia abyssinica* extracts.

1 INTRODUCTION

The diseases caused by multidrug resistant fungal pathogens (e. g dermatophytes like *Trichophyton*, *Microsporum* and *Epidermophyton* as well as opportunistic fungi like *Aspergillus species*, *Candida albicans*, *Mucor* and *Rhizopus species*) are becoming the world's leading cause of health complications if not death, especially in immunosuppressed patients [1]. Though fungal diseases (especially superficial and systemic) appear to be rare, they are not easily treated when contacted and thus most often, some cases of HIV AIDs, Hepatitis B, Cancer etc. and other immunosuppressive diseases become complicated by some of these fungal infections [2]. The treatment of immunocompromised, AIDS and cancer patients becomes sometimes difficult due to this problem [3]. This is even worst in under developed countries like Nigeria, where healthcare facilities are not easily assessable by the vast majority of the population. A greater part of the population in the interior areas of these countries therefore, rely on plant sources for treatment of skin diseases and other mycotic infections. The investigation of these plants used in folklore medicine for skin infections could invariably be a source of the antimycotic agents urgently needed in less developed world today.

Some plants of the genus *Euphorbia*, have been used in folklore medicine from creation till today e. g *Euphorbia kamerunica* and other plants belonging to the family *Euphorbiaceae* have been used to treat skin infections such as ulcers, warts, cancers, tumors [4, 2]. *E. tirucalli* has been used for treatment of swelling, asthma, cough, skin problems and rheumatism. Also its latex is used as treatment for sexual impotence, warts, toothache, hemorrhoids, and snake bites. *Euphorbia granulate* has also shown inhibitory activity against Human immunodeficiency virus (HIV-1) protease [5].

This study therefore seeks to explore the largely unexplored rich natural constituents of *Euphorbia abyssinica*, with the aim of evaluating the antimycotic activity.

This plant (*Euphorbia abyssinica*) is an erect, eight-angled branched tree with deep angles edged with a border of closely packed paired spines (about 1cm long [6]. It belongs to succulent *Euphorbia taxa* (*Euphorbiaceae*) and is commonly found in most African countries; E. Sudan, Erithrea, Nigeria, N. Ethiopia, Somalia, habiting well drained stony soils of hills and slopes in montane evergreen wood-lands

and bush lands at 1300 to 2200m in attitude. Among the Anyang-Kendem people of Mamfe, south West Cameroon it is known as Kechieh Bih. Euphorbias are recorded in international trade first place from Madagascar to France and Candle wax produced from *E. antisiphilitica* in Northern Mexico is internationally traded under CITES. *E. tirucalli* Linne¹⁷⁵³ and *E. milii* are extensively used for hedging purposes in the tropics, Africa and Brazil [7] Some of the non-succulent Euphorbias, for example, *E. pulcherrima* (Poinsettia, flame leaf, Christmas star) native to Southern Mexico and Central America are used as ornamental plants [8].

2 MATERIALS AND METHOD

2.1 Collection of Plant Materials

The plant parts used for this study were *Euphorbia abyssinica* stem bark and the latex. These were collected from Kendem in the Mamfe, south West region of Cameroon. In the Department of Botany, University of Nigeria Nsukka the plant was authenticated and the voucher specimen also deposited. The latex or sap of *Euphorbia abyssinica* was collected by cutting open, parts of the bark on the stem and branches. A container was connected to the bottom of the opening from which the latex dripped into the container. It was then allowed to dry in the water bath at 56°C and stored in a close capped bottle pending its use.

The plant stem-bark was rinsed thoroughly in running tap water, cut into tiny pieces and air dried in the dark. The dried material was then ground to powder in a mortar, weighed and stored in plastic bags in the dark.

2.1.1 Extraction of Plant Materials

Approximately 400 ml of solvent (absolute methanol, water or 50% methanol in water) in a 1 L conical flask was used to soak a 100 g weight of powdered plant material and then covered with cotton wool plugs. After vigorous shaking, the flask was intermittently shaken for 24 hours leaving it in a water bath maintained at 40°C between the intervals of shaking. Three layers of clean muslin cloth were first of all used to filter the mixture before passing it through Whatman no 1 filter-paper. The filtrates were

evaporated to dryness in a water bath at 56°C and the percentage yields of the crude extract determined [9].

2.2 Test organisms

Trichophyton mentagrophytes, *Trichophyton tonsurans*, *Trichophyton violaceum*, *Microsporum gypseum*, *Microsporum canis*, *Epidermophyton floccosum*, *Candida albicans*, species of *Aspergillus*, *Basidiobolus*, *Rodotorula*, *Mucor*, *Rhizopus* and *Geotrichum*, all collected from the Microbiology laboratory of the University of Nigeria Teaching Hospital Enugu, Nigeria, were the fungi used for the study.

They were cultured on Sabouraud Dextrose Agar plates at 25-35°C for 48 hours or more and the resultant pure mature colonies were sub-cultured on Sabouraud Dextrose Agar slants and stored as stock cultures.

Standardization of Fungal Inocula

Fungal stock cultures were sub cultured on Sabouraud Dextrose Agar and incubated at 25-35°C for 7 to 14 days. The matured fungal growths were covered with 2ml of distilled water and gently probed with a sterile loop or the tip of a Pasteur pipette. The resulting suspensions were transferred to sterile test tubes and allowed to settle for about 3-5 minutes. The resultant supernatant suspensions were drained into sterile bottles and the colony forming units (CFU) in the suspensions counted using a haemocytometer. The suspensions were then diluted with Sabouraud dextrose broth to correspond to the final standard inocula suspension (spores or yeast cells) of approximately 1×10^5 colony forming unit per ml [10]

2.3 Susceptibility Testing of Fungi by Pour-Plate Method

The susceptibility testing of fungi was done using pour-plate method of [10]. A 2.0ml amount of each reconstituted plant extract at the concentration of 1000mg/ml was pipetted into sterile glass test tube containing 18mls of molten Sabouraud Dextrose Agar (at about 45°C). The mixtures were swirled carefully for the contents and agar to be thoroughly mixed. Then 100µl of the standard fungal inocula were seeded onto each of the tubes. Again they were thoroughly mixed and poured into each of the plates and allowed to set. They were then incubated at about 25-35°C. A sterile plate without the extract served as the positive control for growth while another plate containing 2.0ml of 16µg/ml voriconazole as

the negative control. As soon as growth was observed at the positive control plates the test plates were checked for growth daily and the period of inhibition of growth was recorded in days.

2.4 Determination of Minimum Inhibitory Concentration (MIC) of the Plant Extracts on the Fungi by Macro-Broth Dilution Method

A two-fold serial dilution of the plant extract was carried out in tubes of Saboraud Dextrose broth to obtain dilutions ranging from 200mg/ml down to 0.39 mg/ml. The 11th tube, which was used as positive growth control culture, did not contain any extract. The control antimycotic agent, voriconazole, was similarly serially diluted but to attain concentrations ranging from 0.125-128µg/ml. Each dilution was seeded with 100µl of the standardized suspensions of the test fungal spores and incubated at 25-35°C for ≥48 hours. The lowest dilutions without visible growth in the tube cultures, compared to the positive and negative controls were considered as the MICs. The tests were carried out in quadruplets and the means of the MICs calculated [10].

2.5 Determination of Minimum Concentration at which 90% Fungal Growth Inhibition was Observed (MIC₉₀) from Broth Dilution Tubes

A 100µl volume of the fungal test suspensions from all the tubes showing no visible fungal growth was sub-cultured on Sabouraud Dextrose agar plates and incubated at 25-35°C. Positive growth control plates were also included and checked simultaneously in comparison with the test plates for fungal growth. The Minimum Fungicidal Concentration (MFC) was considered as the minimum concentration of the test substances that yielded 99%-100% visible growth inhibition on sub-culture of 100 µl of serial dilutions and incubation at 35°C for more than 48hours. Any dilution showing 90% growth inhibition was recorded as MIC₉₀ [10].

2.6 Determination of the Effects of Plant Extracts on Viable Colony Forming Units (CFU) of the Fungi Using Time-Kill (Inhibition) Assay

The effects of 50% methanol extracts of *Euphorbia abyssinica* were evaluated by a time-kill assay using the macrobroth dilution technique. The extracts were reconstituted in 20% Dimethyl Sulfoxide (DMSO) and appropriately diluted to the required concentrations. The inoculums size was determined according to the type of fungus, (e.g. 1 x10⁶ for *Candida albicans*; and1 x10⁵ for dermatophytes). About 1.00ml of the extract was added to 9ml of Saboraud dextrose broth, seeded with the appropriate concentrations of the test fungus to achieve concentrations equivalent to 0.5 x MIC, 1 x MIC, 2 x MIC, or 4 x MICs values. Two

sets of control tubes were included for each experiment. One set was seeded with the organism in broth without extract, and the other set contained broth without organism and extract. The control drug voriconazole was similarly diluted. All the fungal cultures were incubated at 25-35°C for ≥48 hours. Immediately after inoculation of the tubes, aliquots of 100 µl of the negative control tubes contents were taken, serially diluted in saline and seeded on nutrient agar plates to determine the zero hour counts. The same was done for the tubes which contained the test fungi after 0 hour, 3 hours, 6 hours, 24 hours and 48 hours, respectively. After incubation, the emergent colonies were counted and the mean count (CFU) of each test organism was determined and expressed as log₁₀. The Minimum Lethal Concentrations (MLCs) of the extract were the lowest concentrations that gave 99.9% to 100% killing [9].

2.7 Statistical Analysis

The Randomised Complete Block Design (Two-way analysis of variance) was used to analyze the data obtained in the study. The means that were significantly different were separated using Duncan's New Multiple Range Test.

3 RESULTS

3.1 Antifungal Activities of *Euphorbia abyssinica* Extracts

The results of the antifungal activities of 100mg of *E. abyssinica* stem- bark showed that the extract inhibited the growth of all the fungi tested. However, the 50% methanol extract showed better activity (14days or more) than the absolute methanol and aqueous extract (12days or more) (Table 1).

Euphorbia abyssinica latex extracts, also showed similar activity on the fungi tested like the stem bark but inhibition of growth was for lesser number of days. The 50% methanol extract inhibited the growth of all the dermatophytes for 12days or more (Table 1). The control anti mycotic agent (voriconazole) inhibited the growth of all the fungal isolates tested for over 14days, including *Aspergillus*, *Mucor*, *Rhizopus* and *Geotricum species* (Table 1).

3.2 The MICs and MIC₉₀s (mg/ml) of 50% Methanol Extract of Plants on the Fungi

The MICs and MIC₉₀s were evaluated using the 50% methanol extracts of *E. abyssinica* stem- bark. was more active on the yeasts, with *Candida albicans* showing MIC value of 0.39mg/ml and MIC₉₀ of 1.56mg/ml, followed by *Rhodotorulla* species (MIC 0.49mg/ml, MIC₉₀ 3.13mg/m) and *B. haptosporus* (MIC 0.59mg/ml, MIC₉₀ 3.13mg/ml). *E. floccosum* was the most susceptible of the dermatophytes with MIC of 0.98mg/ml and MIC₉₀ of 1.56mg/ml. This was closely followed by *Trichophyton rubrum* (MIC 0.1.95 mg/ml, MIC₉₀ 3.13mg/ml), *M. canis* (MIC 3.9mg/ml, MIC₉₀ 3.13mg/ml) and *T. mentagrophytes* (MIC 0.39mg/ml, MIC₉₀ 6.25mg/ml) respectively. The least susceptible of the fungi were *Microsporum gypseum* (MIC 25mg/ml, MIC₉₀ 25mg/ml) and *T. tonsurans* (MIC 12mg/ml, MIC₉₀ 25mg/ml) respectively (Table 2). Extracts of *E. abyssinica* latex showed its lowest MIC (1.95mg/ml) on, *B. haptosporus* and *E. floccosum* respectively and least MIC₉₀ (3.13mg/ml) on *E. floccosum* and *C. albicans* respectively).The least susceptible of the fungi tested to this extract was *Microsporum gypseum* (MIC /MIC₉₀ of 25mg/ml) (Table 2). For the control drug (voriconazole), *C. albicans* (MIC 0.313 µg/ml, MIC₉₀ of 0.5µg/ml), and *T. rubrum* (MIC 0.375 µg/ml, MIC₉₀ of 0.5µg/ml), were the most susceptible fungi while the least susceptible of the fungi was *B. haptosporus* with MIC of 1.25µg/ml, and MIC₉₀ of 4µg/ml (Table 2).

3.3. Antimicrobial Effect of 50% Methanol Extract of *E. abyssinica* Stem- Bark on Viable Cell Count of Fungi Using Time-Kill (Inhibition) Assay.

The inhibition of the fungal cells was estimated as the reduction of viable cell count following exposure to a given concentration of plant extract or control drug (voriconazole) over a period of time (hours). *E. abyssinica* 50% Methanol extract, even at its multiples of MICs did not inhibit *C. albicans* completely in 48hours but the cell count was reduced to very significant levels by the 4MIC and 2MIC (1.56mg and 0.78mg/ml,) respectively (Fig. 1).The control drug (voriconazole 0.5 µg/ml) killed *C. albicans* in 48hours. The, 4MIC (15mg/ml,) concentration inhibited *T. mentagrophytes* in 3hours, while the 2MIC (7.8mg/ml) inhibited it in 6hours. The 1MIC (3.9 mg/ml) reduced the viable cell count to 2.1 log₁₀ in 48 hours but 0.5MIC (1.95mg/ml) concentration had no effect on *T. mentagrophytes* (Fig. 2). It was inhibited by the control drug (1µg /ml) in 48hours.*M. gypseum*was inhibited by 4MIC and 2MIC (100 mg/ml and 50 mg/ml) concentrations in 3hours respectively but 1MIC (25mg/ml) concentration as well as the sub inhibitory concentration (12.5mg/ml) did not totally inhibit *M. gypseum* cells in 48hours (fig.3). The *M. gypseum* cells were inhibited by the control drug (Voriconazol 2 µg /ml) in 48hours.

223 The exposure of *E. floccosum* to 4MIC (3.9mg/ml) concentration of *E. abyssinica* 50% methanol extract
224 reduced the viable cell count to undetectable levels of growth in 3 hours, and 2 MIC (1.96mg/ml) inhibited
225 it to about 99% in 48 hours respectively (fig. 4). But the cells exposed to 1MIC (0.98mg/ml) and 0.5 MIC
226 (0.49mg/ml) were inhibited to greater than $2\log_{10}$ in 48hours. These concentrations could do better if aloud
227 to have contact with the cells for much longer periods of time. *E. floccosum* cells were also inhibited by
228 the control drug (voriconazole 1 μ g/ml) in 48hours (fig.4).

229 **Table 1: Periods of inhibition (in days) of the growth of some pathogenic fungi by plant extract**

230

Stem-bark	<i>B. h</i>	<i>R. s</i>	<i>C. a</i>	<i>T. m</i>	<i>T. r</i>	<i>T. t</i>	<i>T. v</i>	<i>M. g</i>	<i>M. c</i>	<i>E. f</i>	<i>A. fl</i>	<i>A. fu</i>	<i>M. s</i>	<i>A. n</i>	<i>G. s</i>	<i>R. n</i>
A.methanol	>12	>12	>12	>12	>12	>12	>12	>12	>12	>14	>14	>12	>12	12	7	12
Aqueous	12	12	12	12	12	12	12	12	12	>12	12	12	12	12	7	12
50% Methanol	>14	>14	>14	>14	>14	>14	>14	>14	>14	>14	>12	>12	>12	>12	> 7	> 12
Latex																
A.methanol	12	12	>12	>12	12	12	12	12	12	>12	12	12	12	12	7	12
Aqueous	12	12	12	12	12	12	12	12	12	12	12	12	12	12	7	12
50% Methanol	>14	>14	>14	>14	14	14	14	>14	14	>14	14	1 2	12	12	7	>12
Voriconazole (16µg/ml)	>14	>14	>14	>14	>14	>14	>14	>14	>14	>14	>14	>14	>14	>14	>14	>14

231 *B. h*=*Basidiobolus haptosporus*, *R. s*= *Rhodotorulla species*, *C.a*= *Candida albicans*, *T. m*= *Trichophyton mentagrophytes*, *T. r*=*Trichophyton rubrum*,

232 *T.t*=*Trichophyton tonsurans*, *T.v*=*Trichophyton violaceum*, *M.g*, *Microsporum gypseum*, *M.c*= *Microsporum canis*, *E.f*= *Epidermophyton floccosum*, *A.fl*.

233 *Aspergillus flavus*, *A.fu*=*Aspergillus fumigatus*, *M.s*= *Mucor species*, *G.S*=*Geotricum species*, *R.n*=*Rhizopus nigricans*,>=more than *A. n*= *Aspergillus niger*.

234 A.methanol=Absolute methanol

235

236

237 **Table 2: MICs and MIC₉₀s of plant extracts (mg/ml) on some pathogenic fungi**

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Fungal Species	<i>E. abyssinica</i> Stem-bark Extract		<i>E. abyssinica</i> Latex Extract		Voriconazole (µg/ml)	
	MIC±SD (mg/ml)	MIC ₉₀ (mg/ml)	MIC±SD (mg/ml)	MIC ₉₀ (mg/ml)	MIC±SD (µg/ml)	MIC ₉₀ (µg/ml)
<i>C. albicans</i>	0.39±0.00	1.56	2.73±0.78	3.13	0.313±0.13	0.50
<i>Rhodotorulla species</i>	0.49±0.19	3.13	2.34±0.90	6.25	0.75±0.29	2.00
<i>B. haptosporus</i>	0.59±0.23	3.13	1.95±0.78	6.25	1.25±0.50	4.00
<i>E. floccosum</i>	0.98±0.39	3.13	1.95±0.78	3.13	0.313±0.13	1.00
<i>T. mentagrophytes</i>	3.9±1.6 0	6.25	6.25±0.00	12.5	0.189±0.07	1.00
<i>T. rubrum</i>	1.95±1.36	3.13	3.91±1.56	12.5	0.375±0.14	0.50
<i>T. violaceum</i>	3.91±1.56	6.25	5.476±1.56	12.5	0.50±0.00	0.50
<i>M. canis</i>	3.91±2.70	3.13	2.73±2.3	6.25	0.25±0.00	0.50
<i>T. tonsurans</i>	12.5±0.00	6.25	18.7±57.20	25	0.25±0.00	1.00
<i>M. gypseum</i>	25.0±0.00	25	25.0±0.00	25	0.75±0.29	2.00
<i>Aspergillus favus</i>	12.5±0.00	25	15.63±6.30	25	0.25±0.00	0.50
<i>Aspergillus Niger</i>	6.25±0.78	25	12.5±0.00	25	0.25±0.00	1.00
<i>Aspergillus fumigatus</i>	6.25±0.59	25	12.5±0.00	25	0.25±0.00	1.00
<i>Rhyzopus nigrcans</i>	12.5±0.78	25	25.0±0.39	50	0.189±0.07	1.00
<i>Mucor species</i>	25.0±0.39	50	25.0±0.39	50	0.375±0.14	1.00
<i>Geotricum species</i>	50.0±0.195	100	50.0±0.195	100	0.75±0.29	2.00

239 *Post Hoc Tests Duncan*^{a,b}

240 *a Uses Harmonic Mean Sample Size = 16.000*

241 *b. Alpha = .05*

242 *C. albicans significant mean .896, most susceptible yeast tested*

243 *Epidermophyton floccosum, significant mean.1.053, the most susceptible of the dermatophytes tested*

244 *M. gypseum, significant mean 14.641, least susceptible of the dermatophytes tested*

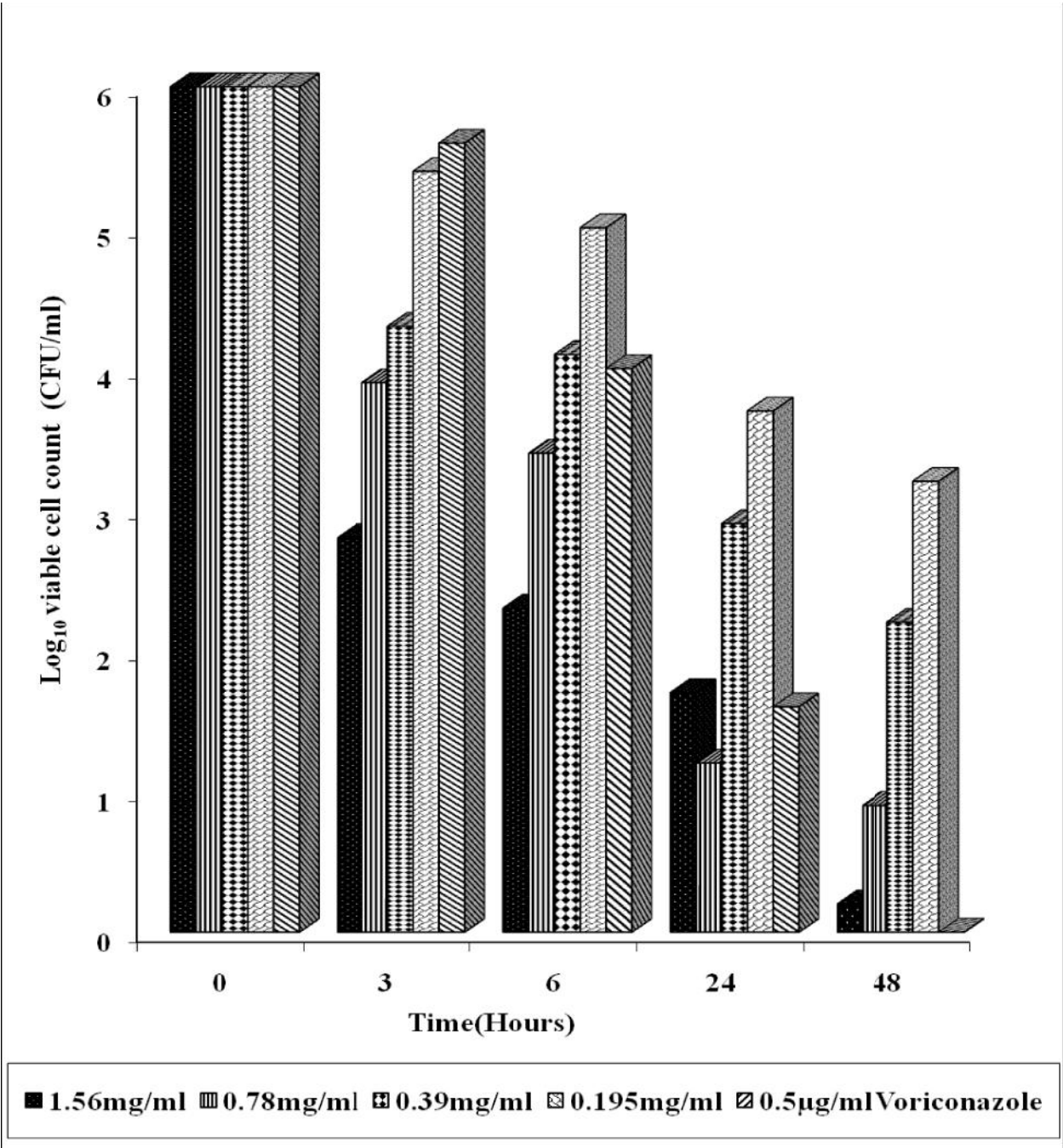
245 *Voriconazole significant mean .4938*

246 *Euphorbia abyssinica stem-bark 5.3608*

247 *Euphorbia abyssinica latex 7.1086*

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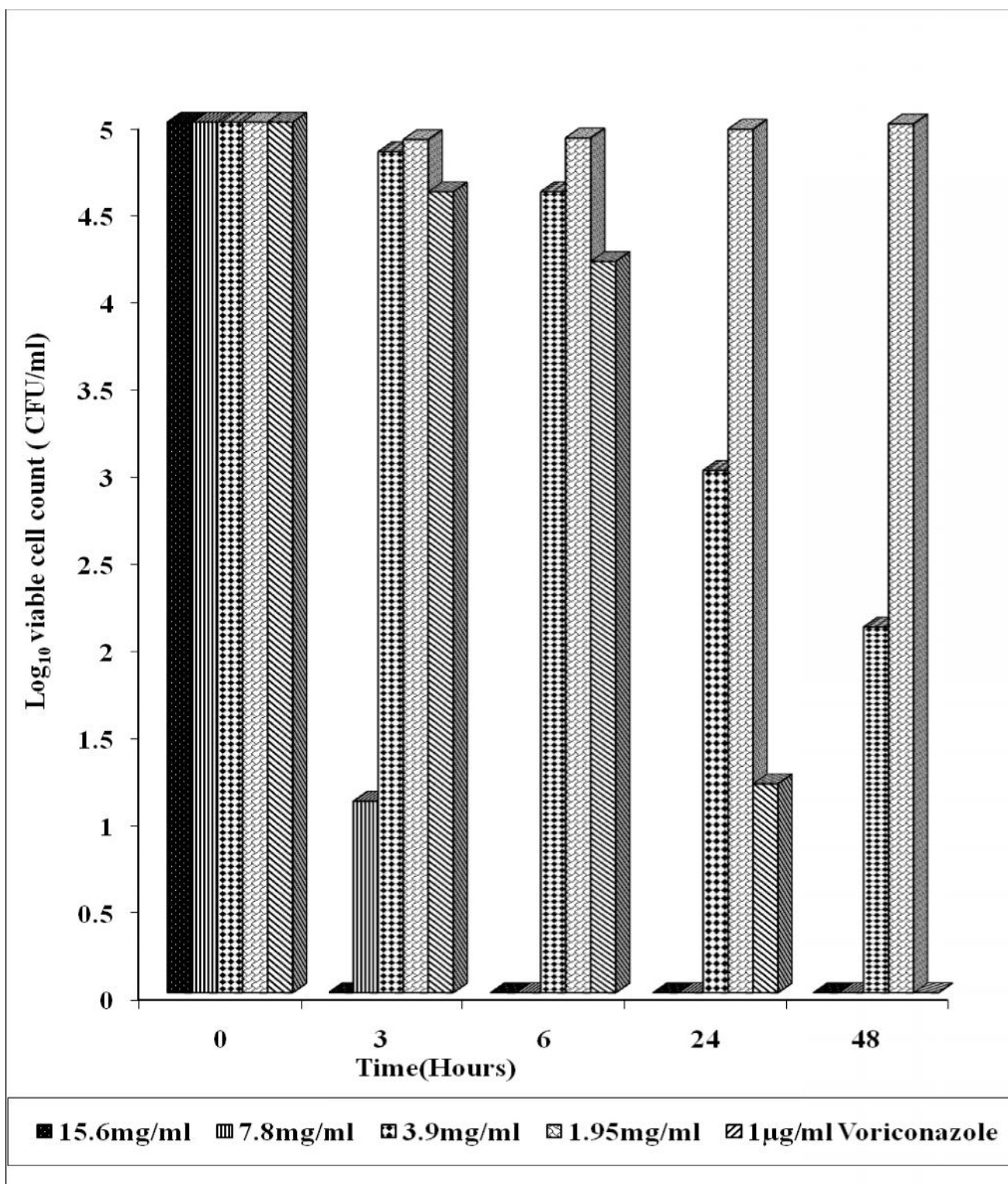
249
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252
253 Figure 1: Effect of 50% Methanol Extract of *Euphorbia abyssinica* Stem-Bark on Viable
254 Cell count of *Candida albicans*.

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258 Figure 2: Effect of 50% Methanol Extract of *Euphorbia abyssinica* Stem-Bark on Viable259 Cell Count of *Trichophyton mentagrophytes*

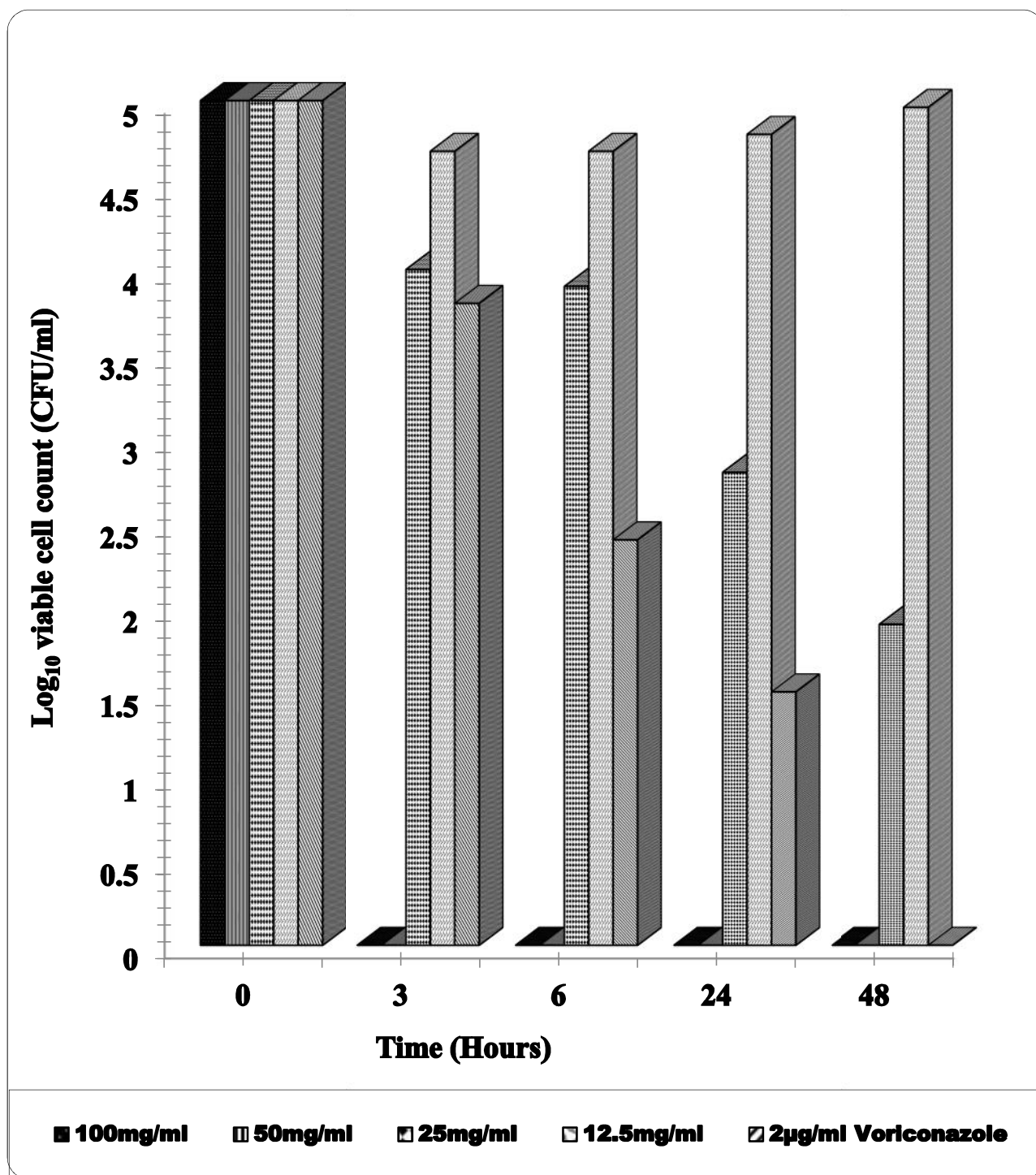


Figure 3: Effect of 50% Methanol Extract of *Euphorbia abyssinica* Stem-Bark on Viable Cell Count of *Microsporium gypseum*

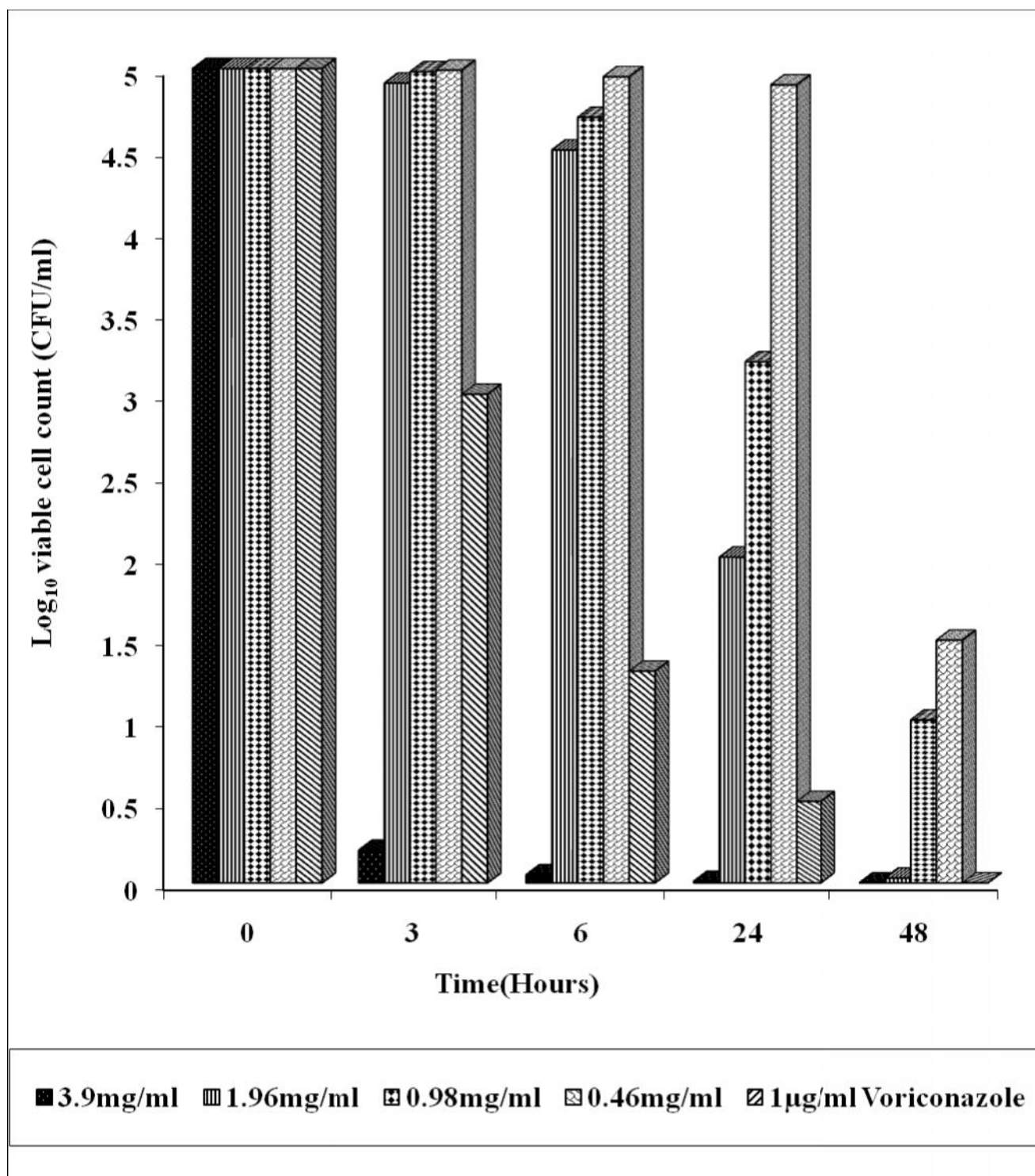


Figure 4: Effect of 50% Methanol Extract of *Euphorbia abyssinica* Stem- Bark on Viable Cell Count of *Epidermophyton floccosum*.

4 DISCUSSION

The study of antifungal activity of crude extracts (absolute methanol, water and 50% methanol) of *Euphorbia abyssinica* by agar dilution and macrobroth dilution method showed that the extracts inhibited the growth of the test organisms. In the agar dilution, there was no significant difference between the activity of the stem-bark and that of the latex extracts. Also there was no significant difference between the activity of the absolute methanol and the aqueous extracts even though the methanol extract were more active when compared with the aqueous extract. The slight difference in the activity might have been probably due to the solubility of the chemical and bioactive constituents in the extracting solvents (methanol and water). [1] reported the significant antifungal activity of methanol and aqueous extracts of *Euphorbia tirucalli* with mean inhibition zone of 15.33 ± 0.88 mm and 17.33 ± 0.33 mm respectively for *C. albicans* (ATCC9002) and *A. niger* 14.67 ± 0.67 and 16.33 ± 0.33 for methanol and aqueous respectively. The antifungal activity of methanol and aqueous extracts of other *Euphorbia* species have also been reported by other researchers [11,12, 13,14, 15, 16,17].

The 50% methanol extracts of the two plant parts (stem-bark and latex) showed significantly higher antifungal activity than the absolute methanol and aqueous plant extracts. In the macrobroth dilution method the results showed that at a significant value of $\alpha = .01$ the 50% methanol extract of the stem-bark was more active than the 50% methanol extract of the latex. They were significant at mean values of 5.361 for stem-bark extract and 7.109 for the latex extract.

This activity was dose dependent as greater inhibition of growth was observed at high concentrations than at lower concentrations in the Time kill assay. Similar observations as these, have also been reported with other *Euphorbia* species extracts by other researchers [18 19,15].

It has been observed that organic solvents extract better than water [15]. However a mixture of the two solvents yielded better results in this study.

Both yeasts and moulds, dematophytes and opportunistic pathogens were inhibited by extracts of the two plant parts studied. *Epidermophyton floccosum* and *Candida albicans* were the most susceptible of the moulds and yeasts tested respectively. Though, the differences between the means was highly significant. The more susceptibility pattern of the yeasts showed that they were more susceptible than the moulds at $P = .05$ and significant mean values of .896 for *Candida albicans* and 1.053 for *Epidermophyton*

flocossum. [4] also in their research on *E. szovitsii* stated that the latex extracts of the plant showed inhibitory effects at different ratios against yeasts and dermatophytes with the lowest MIC shown by *Epidermophyton* sp. In a similar study by [2], *Euphorbia kamerunica* inhibited the growth of *Microsporum audouinii* for 9 days. He also reported that the growth of *Aspergillus niger* was inhibited by aqueous and methanol extracts of this plant while *Microsporum canis* var. *distortum* was also inhibited by ethanol extract at 2.5 mg/ml and 5 mg/ml in the same experiment. The extracts from the two plant parts therefore showed some anti-fungal activity against the test organisms.

4.1 Conclusion

From this experiment crude extracts of *Euphorbia abyssinica* plant were efficacious against some dermatophytes, yeasts and opportunistic pathogens which supports its ethno medicinal uses as a broad spectrum herbal remedy. Further research on the fractions of these plant extracts to reveal the actual bioactive compounds will be of great value. This plant might thereafter be used as treatment for infections caused by these fungi.

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