



Research article

Antifungal and anti-mycotoxigenic activity of selected South African medicinal plants species

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ABSTRACT

Antifungal and anti-mycotoxigenic activity of 25 acetone leaf extracts of South African medicinal plants with potent antioxidant activity were investigated against three phytopathogenic fungal strains. The extracts exhibited varying degrees of *in vitro* anti-mycotoxigenic effect against *Fusarium verticillioides*, *Aspergillus flavus* and *Aspergillus ochraceus*. *Markhamia obtusifolia* (Baker) Sprague exhibited the lowest minimum inhibitory concentration (MIC) values as low as 0.08 mg/ml against *Aspergillus flavus* and *Fusarium verticillioides* at both 24 and 48 hr incubation period, while *Curtisia dentata* exhibited similar MIC value against *Aspergillus ochraceus*. *Curtisia dentata* further yielded the highest total activity of 1583 ml/g against *Aspergillus ochraceus* at 24 and 48 hr incubation period. In the mycelial growth inhibition (MGI) evaluation, *Fusarium verticillioides* was more sensitive to plants extracts, while *Kirkia wilmsii* exhibited highest MGI of 50.08% against *Fusarium verticillioides* on the 6th day of incubation. Five acetone extracts from *Acokanthera oppositifolia*, *Bauhinia galpinii*, *Combretum caffrum*, *Ricinus communis* and *Solanum aculeastrum* exhibited lowest IC₅₀ value of 0.01 mg/ml against (2,2'-azinobis-3-ethylbenzthiazoline-6-sulphonic acid (ABTS). *Curtisia dentata* and *Markhamia obtusifolia* extracts were further subjected to gas chromatography mass-spectrophotometry (GC-MS) analysis. *Curtisia dentata* revealed the presence of triterpenoid compounds, β -amyrin (53.30%) and α -amyrin (6.42%), while *Markhamia obtusifolia* yielded the presence of neophytadiene (4.38%) and palmitic acid (3.61%). The results suggest that natural products from plants may well be used as possible substitutes for synthetic fungicides. Given the antifungal and antioxidant potential of the selected plants, they may have potential as possible leads for the development of biofungicides that may well prevent oxidation related food spoilage.

1. Introduction

Agriculture and food industries are suffering from major losses as a result of fungal infections and subsequent mycotoxin contamination of crops and feeds by microorganisms during pre and post-harvest (Mdee et al., 2009). These toxins are mostly produced by ubiquitous, saprophytic fungal strains belonging to the genus *Aspergillus*, *Fusarium*, *Aternaria*, *Claviceps* and *Penicillium*. Mycotoxin food contamination is a worldwide food safety problem with an estimate of about twenty-five percent of world's crop annually affected (Garcia et al., 2012). Crop infection by these fungi does not only result into mycotoxin contamination but it also leads to a reduced crop yield and quality that hinders the

trade markets of many countries. Consumption of food contaminated with mycotoxins does not only affect plants, but also poses a huge risk to both human and animal health. Occurrence of mycotoxin levels in food systems are mostly regulated by legislations and limits in developed countries. Although some developing and underdeveloped countries adopted these regulations, the enforcement remains less due to socio-economic impacts and unacceptable agricultural practises (Wagacha and Muthomi, 2008).

Exposure to mycotoxins occurs mainly through direct consumption of contaminated crops and plant-derived food or mycotoxin metabolites carried over in animal products (Thompson and Henke, 2000). Studies also show that mycotoxins are associated with several health

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effects such as carcinogenesis, hepatotoxicity, nephrotoxicity, teratogenicity and mutagenicity (Reddy et al., 2010). Most researches are focused on mycotoxins of agricultural and health importance including among others fumonisins, zearalones, deoxynivalenol, T-2, aflatoxins and ochratoxins which mostly infect maize, wheat and rice (Alshannaq and Yu, 2017). Significant exposure to fumonisin B₁ is suspected to increase the risk of oesophageal cancer, while fumonisins are associated with neural tube defects (Marasas et al., 2004). According to van Rensburg et al. (2015), commercially produced maize in South Africa is mainly contaminated with *Fusarium verticilloides* during pre-harvest. With maize being a staple diet to most of African countries, there is an increased risk of fumonisin consumption (Chelule et al., 2001).

Medicinal plants have been the basis for medicinal treatment through much of human history, and such traditional medicines are still widely practiced today where many people across the world are resorting to these products for treatment of various health challenges in different healthcare settings (Begaa and Messaoudi, 2019). Plants offer excellent perspectives for the discovery of new therapeutic products, some of which can be developed as biofungicides (for prevention of contamination) and nutraceuticals for prevention of mycotoxicosis. These plant-based biofungicides may have low to no toxic effects. Plants are considered to be rich source of secondary metabolites which are pharmacologically active compounds recognised by pharmacologists to have reactions towards sickness. The enormous chemical diversity of plant secondary metabolites presents a valuable resource for possible development of new pharmaceuticals. Several strategies to control mycotoxin contamination both pre-harvest and post-harvest have been applied which include among others the use of atoxigenic fungi/bacteria to control toxigenic fungal strains. Genetically modified crops and synthetic fungicides are also used to control crop contamination by phyto-pathogenic fungi; however, these are expensive to most resource poor farmers (Salako, 2002).

Synthetic fungicides application appears to be the most favored method, but due to negative environmental hazards, long degradation periods and toxigenic fungal strains developing resistance to most synthetic fungicides, there is a need for a search of an alternative less expensive and eco-friendly method to control fungal contamination both pre and post-harvest (Daferera et al., 2000). Medicinal plants harbor valuable secondary metabolites, which have tremendous biological activities against variety of pathogens and may well prevent oxidation in crops (Prakash et al., 2012). The aim of this study is to explore the *in vitro* antifungal and antioxidant activity of selected South African medicinal plants, with the aim of developing a less expensive, easily biodegradable and eco-friendly bio-fungicide that can be used as a possible substitute to synthetic fungicides. The plants used in the current study were selected based on their availability in the National Botanical Garden (NBI) as the identity of the plant is already confirmed. Only leaves were used due to plant conservation consideration.

2. Materials and methods

2.1. Plant collection

Leaves of twenty-five plant species without symptoms of fungal or bacterial infections were collected from tress growing in the Lowveld Botanical Garden (30°57'58.16" E 25°26'42.61" S Long 30.96800 Lat -25.44669) in the Mpumalanga province, South Africa, in March 2016. Mr Willem Froneman (South African National Biodiversity Institute) confirmed the identity of the plants and matched those in the herbarium with voucher specimen numbers reported (Table 1). The leaves were shade dried at room temperature in a well-ventilated room, pulverised into fine powder and stored in glass bottles in a cool dry place.

Table 1

Plant species used and their specimen voucher numbers.

Plant species	Family	Voucher No.
<i>Acokanthera oppositifolia</i> (Lam.) Codd.	Apocynaceae	Glow 179/1986
<i>Apodytes dimidiata</i> E.Mey ex Arn.	Metteniusaceae	Glow 63/1986
<i>Artemisia afra</i> Jacq. ex Willd.	Asteraceae	Glow 27/2010
<i>Brachylaena discolor</i> DC.	Asteraceae	Glow 47/1993
<i>Bauhinia galpini</i> N.E.Br.	Fabaceae	Glow 27/1986
<i>Milletia grandis</i> (E.Mey.) Skeels	Fabaceae	Glow 264/1981
<i>Breonadia salicina</i> (Vahl) Hepper & J.R.I. Wood	Rubiaceae	Glow 137/1985
<i>Capparis tamentosa</i> Lam.	Capparaceae	Glow 22/1982
<i>Combretum caffrum</i> (Eckl. & Zeyh.) Kuntze	Combretaceae	Glow 92/1997
<i>Curtisia dentata</i> (Burm.f.) C.A.Sm.	Cornaceae	Glow 96/1998
<i>Dracaena mannii</i> Bakker	Asparagaceae	Glow 30/1971
<i>Ficus natalensis</i> Hochst.	Moraceae	Glow 87/1975
<i>Harpephyllum caffrum</i> Bernh.	Anacardiaceae	Glow 164/1974
<i>Heteromorpha arborens</i> (Spreng.) Charm & Schldl.	Apiaceae	Glow 88/1972
<i>Kirkia wilmsii</i> Engl.	Kirkiaceae	Glow 82/1972
<i>Markhamia obtusifolia</i> (Baker) Sprague	Bignoniaceae	Glow 16/1994
<i>Maytenus undata</i> (Thunb.) Blakelock	Celastraceae	Glow 157/1986
<i>Mystrocydon aethiopicum</i> (Thunb.) Loes.	Celastraceae	Glow 51/1983
<i>Ricinus communis</i> L.	Euphorbiaceae	Glow 322/1988
<i>Solanum aculeastrum</i> Dunal.	Solanaceae	Glow 229/1987
<i>Spirostachys africana</i> Sond.	Euphorbiaceae	Glow 146/1972
<i>Strychnos mitis</i> S.Moore	Loganiaceae	Glow 243/1993
<i>Warburgia salutaris</i> (G.Bertol) Chiov.	Canellaceae	Glow 167/1988
<i>Xylotheca kraussiana</i> Hochst.	Achariaceae	Glow 228/1990
<i>Zanthoxylum capense</i> (Thunb) Harv.	Rutaceae	Glow 117/1985

2.2. Extraction

The powdered leaf material (3g) of each plant species was added to 30 ml acetone-AR grade (1:10 w/v) in glass bottles. The bottles were vigorously shaken in a laboratory shaker overnight (Already Enterprise Inc., Taiwan, Model LM-600 RD) at 120 rpm. The resulting supernatant was filtered through Whatman no.1 filter paper and concentrated to dryness using rotary evaporator. Percentage extraction yield of each plant extract was determined by dividing the total extracted mass by dried plant mass used for extraction. Plant extracts of 10 mg/ml concentration dissolved in 30% acetone were prepared for subsequent bioassays.

2.3. Antifungal activity

2.3.1. Selected fungal pathogens

Freshly prepared fungal cultures of *Aspergillus flavus* (PPRI: 14636), *Aspergillus ochraceus* (PPRI: 6816) and *Fusarium verticilloides* (PPRI: 10148) were purchased from Agricultural Research Council-Plant Protection Research Institute (ARC-PPRI). Dr Adriaana Venter-Jacobs (Agricultural Research Council-Plant Protection Research Institute) confirmed their identities. The fungal cultures were sub cultured from potato dextrose agar (PDA) slants into the plates of freshly prepared Potato dextrose broth (PDB) growth medium.

2.3.2. Inoculum preparation

Sterile distilled water and 0.1% tween-80 spore suspensions were prepared by gently scrubbing the conidia from periphery of actively growing 4 to 5 days old cultures of *Fusarium* and *Aspergillus* respectively.

Tween-80 assists with coating *Aspergillus* spores and prevents them from floating in liquid broth media. A full loop of each spore suspension was transferred into 50 ml freshly prepared potato dextrose broth (PDB) and incubated for 3 to 4 days at 30 °C until slight turbidity was observed. A turbid inoculum was gently agitated using a probe sonicator to release the conidia from the hyphae and filtered through sterile autoclaved non-absorbent piece of cheesecloth. The final inoculum was adjusted to approximately 1×10^6 conidia ml^{-1} using haemocytometer (Petrikou et al., 2001).

2.3.3. Determination of minimum inhibitory concentrations (MIC) and total activity of plant extracts

Microplate dilution method developed by Eloff (1998) with slight modifications was used to determine minimum inhibitory concentrations of 10 mg/ml plant extracts against selected phyto-pathogenic fungi. Briefly, 100 μl of plant extracts were transferred into the first row of wells in a flat bottom sterile 96 well plate (Merck, RSA) laid with 100 μl autoclaved distilled water. The contents in the wells were serially diluted two fold from wells A to H. Aliquot of 100 μl of quantified inoculum of each fungal strain was added into all the wells. Amphotericin B and 30 % acetone were prepared as positive and negative controls respectively. All the wells were then loaded with 40 μl of 2 mg/ml *p*-Iodonitroterazolium (INT) chloride (Sigma-Aldrich, Germany). The plates were sealed with parafilm and incubated at 30 °C for 24 and 48 hours under 100% humidity. Minimum inhibitory concentration was determined as the lowest concentration of plant extract that inhibits microbial growth after 24 and 48 hours of incubation. The extracts were tested in triplicates. Total activity was calculated by dividing quantity extracted in milligrams from 1g of plant material by MIC value (mg/ml), to determine total activity level of antifungal compounds of each extract (Masoko et al., 2005).

2.3.4. Mycelial growth inhibition (MGI) study

The effect of plant extracts on inhibition of fungal mycelia growth was evaluated by using a method previously applied by Shukla et al. (2012) with slight modifications. Briefly, three biological replicates of varying concentrations of plant extracts and Amphotericin B were added into Potato Dextrose Agar petri dishes during preparation to yield the final concentrations of MIC values (i.e MIC, $\frac{1}{2}$ MIC and $\frac{1}{4}$ MIC concentrations). The PDA plates without amendments were used as controls for each tested concentration. The discs of 4mm diameter cut from periphery of fresh and actively growing 4–5 days old fungal strains were aseptically inoculated upside down at the centre of all treatments and control set and a drop (10 μl) spore suspensions of all fungal species was pipetted at the centre of the plates. The plates were then incubated for nine days at 30 °C and the mycelial growth diameter was measured after every three days. Percentage MGI was determined from growth diameter using the formula;

$$\% \text{MGI} = (\Delta \text{Dc} - \Delta \text{Dt}) / \Delta \text{Dc} \times 100$$

Where ΔDc = Average mycelial growth diameter measured at three points on control sets and ΔDt = Average mycelial growth diameter measured at three points in the treatment sets.

2.4. Antioxidant activity

2.4.1. 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity

The DPPH radical scavenging activity of acetone plant extracts was determined using the method previously described by (Mongalo et al., 2012) with slight modifications. Briefly, aliquots of plant extracts were serially diluted two-fold to yield concentrations (1000, 500, 250, 125, 63, 31, 16, 8, 4 $\mu\text{g}/\text{ml}$) in 100 μl of methanol inside 96 well microplate, followed by addition of 100 μl methanol solution of 0.02 mg/ml DPPH (w/v) into all wells. Ascorbic acid (0.5 mg/ml) and methanol were used as positive and negative controls respectively. The plates were then incubated in dark for 1 hr prior to absorbance reading at 517.

2.4.2. The 2,2'-azinobis(3-ethylbenzthiazoline-6-sulphonic acid (ABTS) radical scavenging activity

ABTS solution was prepared by dissolving 7mM ABTS and 2mg of potassium persulphate in 3 ml distilled water. The solution was incubated in dark for 16–17 hours. The solution was then diluted 1:60 (v/v) with pure grade methanol. In a sterile 96 well microplate laid with 100 μl methanol, plant extracts were serially diluted two fold to yield different concentrations (500, 250, 125, 63, 31, 16, 8, 4 $\mu\text{g}/\text{ml}$) followed by addition of 100 μl methanol solution of ABTS into all the wells. The plates were then incubated in dark for 5 minutes. Ascorbic acid (0.5 mg/ml) and methanol were used as positive and negative controls respectively. The absorbance was measured at 734 nm (Re et al., 1999).

In both assays (DPPH and ABTS), the percentage of inhibition (IC_{50}) were calculated using the formula:

$$\% \text{ inhibition} = 1 - (A_t / A_0) \times 100$$

Where, A_t equals absorbance of treated sample, A_0 equals absorbance of negative control. IC_{50} values were determined using Prism pad programme from three experimental replicates.

3. Results and discussion

The investigated plants varied in their percentage leaf extracts yield. The highest percentage yield of the plant leaf extracts was obtained from *Xylothea kraussiana* (35.0%), while the lowest was obtained from both *Acokanthera oppositifolia* and *Ficus natalensis* yielding 3.33% (Table 2). The percentage yields in the crude extracts of these plant species may be proportionately associated with biological compounds responsible for various biological activities such as antimicrobial activity, antioxidant activity and antiproliferative effect (Mongalo et al., 2019).

Minimum inhibitory concentration (MIC) is the lowest concentration of plant extract that inhibits the fungal growth. In this study, the tested plants showed potent to weak antifungal activities, indicated by varying degrees of colour density in the wells where fungal proliferation is indicated by reduction of yellowish *p*-Iodonitroterazolium (INT) chloride to a reddish coloured formazan. The MIC values ranged from as low as 0.08 to 2.5 mg/ml against tested phyto-pathogenic strains of *A. flavus*, *A. ochraceus* and *F. verticilloides* were observed (Table 2). As there is no validated endpoint criteria for *in vitro* testing of plant extracts; a proposed classification by Souza et al., 2007 was adopted in determining the antifungal activity of the extracts. That is, extracts with $\text{MIC} < 0.5$ mg/ml were considered strong inhibitors while MIC between 0.5 and 1.6 mg/ml were considered moderate inhibitors and extracts with $\text{MIC} > 1.6$ mg/ml were considered weak inhibitors. *Aspergillus flavus* had moderate sensitivity to most plant extracts, namely *Acokanthera oppositifolia*, *Apodytes dimidiata*, *Artemisia affra*, *Bauhinia galpinii*, *Brachylaena discolor*, *Breynia salicina*, *Combretum caffrum*, *Kirkia wilmsi*, *Maytenus undata*, *Milletia grandis*, *Myroxylon aethiopicum*, *Solanum aculeastrum* and *Spirostachys africana* with MIC value of 0.16 mg/ml after 24 hr incubation period. *Curtisia dendata*, *Markhamia obtusifolia* and *Maytenus undata* extracts had good antifungal activity against *Aspergillus ochraceus* with MIC values ranging between 0.08 and 0.16 mg/ml.

Similarly, the MIC values of 0.08 and 0.16 mg/ml were recorded for *M. obtusifolia* and *C. dendata* respectively against *Fusarium verticilloides*. A noteworthy activity was observed for *M. obtusifolia* extract, with strong antifungal activity against all tested myco-toxicogenic fungal strains. The varying sensitivity levels of the tested fungi to varying concentrations of plants extracts can be attributed to the type of the conidia formed as a protective mechanism to the toxicity of plant extracts. External factors such as stress due to application of fungicides, nitrogen fertilizers and environmental factors like temperature, pH and moisture are known to influence the fungal conidia formation (Leandro et al., 2003; Paterson, 2007; Mabuza et al., 2018). *Ricinus communis*, *Zanthoxylum capense*, *C. dendata*, *M. obtusifolia*, *M. undata* and *M. aethiopicum* had good antifungal activity, with the lowest MIC value of 0.08 mg/ml comparable to the

Table 2

Minimum inhibitory concentrations (mg/ml) of twenty-five acetone plant extracts against fungi recorded after 24 and 48 hours incubation periods.

Medicinal plants	Extraction Yield	<i>A. flavus</i>		<i>A. ochraceous</i>		<i>F. verticilloides</i>	
		24 hrs	48 hrs	24 hrs	48 hrs	24 hrs	48 hrs
<i>Acokanthera oppositifolia</i>	3.33	0.16	0.63	1.25	2.5	2.5	2.5
<i>Apodytes dimidiata</i>	17.67	0.16	0.16	1.25	2.5	2.5	1.25
<i>Artemisia afra</i>	16.00	0.16	0.78	0.31	1.25	1.25	2.5
<i>Bauhinia galpini</i>	6.33	0.16	0.16	0.10	1.25	0.20	0.20
<i>Brachylaena discolor</i>	4.33	0.16	0.16	2.5	2.5	2.5	2.5
<i>Breonadia salicina</i>	13.67	0.16	0.16	1.25	2.5	2.5	2.5
<i>Capparis tamentosa</i>	8.67	0.31	1.25	0.31	2.5	2.5	2.5
<i>Combretum caffrum</i>	8.33	0.16	0.16	0.16	0.63	0.63	0.31
<i>Curtisia dentata</i>	12.67	0.63	1.25	0.08	0.08	0.16	0.16
<i>Dracaena mannii</i>	7.00	0.31	0.31	0.63	0.63	2.5	2.5
<i>Ficus natalensis</i>	3.33	2.5	2.5	0.31	0.63	0.31	0.63
<i>Harpephyllum caffrum</i>	12.33	0.63	1.25	0.78	1.56	0.78	0.78
<i>Heteromorpha arborescens</i>	14.00	0.63	2.5	0.31	1.25	1.25	1.25
<i>Kirkia wilmsii</i>	16.67	0.16	0.16	2.5	2.5	2.5	2.5
<i>Markhamia obtusifolia</i>	7.67	0.08	0.08	0.16	0.16	0.08	0.08
<i>Maytenus undata</i>	8.00	0.16	0.16	0.08	0.16	0.31	0.63
<i>Milletia grandis</i>	8.00	0.16	0.16	0.20	0.39	0.39	0.78
<i>Mystroxydon aethiopicum</i>	8.67	0.16	0.16	0.31	0.31	0.08	0.16
<i>Ricinus communis</i>	8.00	0.08	0.16	0.20	0.39	0.39	0.39
<i>Solanum aculeastrum</i>	7.00	0.16	0.16	0.78	0.39	0.26	0.33
<i>Spirostachys africana</i>	18.67	0.16	0.16	0.16	0.31	0.31	0.31
<i>Strychnos mitis</i>	16.33	1.25	1.25	0.31	2.5	0.63	2.5
<i>Warburgia salutaris</i>	12.00	0.78	0.78	0.13	0.39	0.10	0.20
<i>Xylothea kraussiana</i>	35.00	0.78	0.78	2.5	2.5	0.63	0.63
<i>Zanthoxylum capense</i>	8.00	0.78	0.78	0.16	2.5	0.08	0.63
Amphotericin B (mg/ml)		0.16	1.56	0.16	1.25	1.56	1.56

Bold value indicates noteworthy activity.

antifungal activity of the antibiotic drug, Amphotericin B, that had the lowest MIC value of 0.16 mg/ml. These results are corroborating those of other South African medicinal plant species, which exhibited a similar MIC value against other mycotoxigenic fungal strains (Masoko et al., 2005), while other plant extracts yielded MIC values as low as 0.02 mg/ml (Mahlo et al., 2010; Manganyi et al., 2015; Eloff et al., 2017). However, it is important to note that the consensus on antimicrobial agents from medicinal plants is that an extract with MIC value of 0.1 mg/ml in an in vitro assay is noteworthy and may be further explored for possible antimicrobial compounds (Mongalo et al., 2015).

Total activity of the selected medicinal plants are presented in Table 3. Extracts of *C. dentata* exhibited the highest total activity yielding 1583 ml/g against *Aspergillus ochraceous*, while *Apodytes dimidiata* exhibited total activity of 1104 ml/g against *Aspergillus flavus*. These results suggest that 1 g of the acetone extract from *C. dentata* can still inhibit the growth of *A. ochraceous* even when diluted with 1583 ml of distilled water (Fadipe et al., 2015). It is important to note that the total activity is dependent on the solubility of the plant materials in a specific solvent and the activity of such extract on the selected microorganisms.

The antifungal stability of plant extracts against fungal strains of *F. verticilloides*, *A. flavus* and *A. ochraceous* was also evaluated by

Table 3

Total activity in (ml/g) of selected medicinal plants after 24 and 48 hours incubations.

Plant species	<i>A. flavus</i>		<i>A. ochraceous</i>		<i>F. verticilloides</i>	
	24 hrs	48 hrs	24 hrs	48 hrs	24 hrs	48 hrs
<i>A. oppositifolia</i>	208	53	27	13	13	13
<i>A. dimidiata</i>	1104	1104	141	71	71	141
<i>A. affra</i>	205	1000	516	128	128	64
<i>B. galpinii</i>	396	396	633	51	317	317
<i>B. discolor</i>	271	271	17	17	17	17
<i>B. salicina</i>	854	854	109	55	55	55
<i>C. tamentosa</i>	280	69	280	35	35	35
<i>C. caffrum</i>	521	521	521	132	132	269
<i>C. dentata</i>	201	101	1583	1583	792	792
<i>D. mannii</i>	226	226	111	111	28	28
<i>F. natalensis</i>	13	13	108	53	108	53
<i>H. caffrum</i>	196	99	158	79	158	158
<i>H. arborescens</i>	222	56	452	112	112	112
<i>K. wilmsii</i>	1042	1042	67	67	67	67
<i>M. obtusifolia</i>	958	958	479	479	958	958
<i>M. undata</i>	500	500	1000	500	258	127
<i>M. grandis</i>	500	500	400	205	205	103
<i>M. aethiopicum</i>	542	542	280	280	1083	542
<i>R. communis</i>	103	500	400	205	205	205
<i>S. aculeastrum</i>	438	438	90	179	269	212
<i>S. africana</i>	1167	1167	1167	602	602	602
<i>S. mitis</i>	131	131	527	65	259	65
<i>W. salutaris</i>	154	154	923	308	1200	600
<i>X. kraussiana</i>	449	449	140	140	556	556
<i>Z. capense</i>	103	103	500	32	1000	127

Bold value indicates noteworthy activity.

amending plant extracts into PDA growth media. The extracts varied in the extent of degree of fungal inhibition, inhibiting mycelial growth in a concentration dependent manner (Tables 4, 5, and 6). Extracts of *A. affra*, *C. dentata*, *K. wilmsii*, *M. aethiopicum* and *S. aculeastrum* exhibited good inhibition of mycelial growth of *Fusarium verticilloides* with percentage inhibition values of 46.78, 37.29, 47.17, 41.58, and 38.11 respectively at the highest concentration tested after three days incubation period. Both *A. dimidiata* and *M. aethiopicum* extracts exhibited potent MGI (% inhibition >10 in all three tested concentrations) against *Aspergillus flavus*, while extracts from *S. aculeastrum* and *B. salicina* yielded potent inhibition of mycelial growth of *Fusarium verticilloides* and *Aspergillus ochraceous* respectively.

The extracts of *B. salicina*, *D. mannii*, *S. africana*, and *W. Salutaris* slightly inhibited the growth of *A. ochraceous* with percentage values 26.33, 27.99, 31.04 and 28.59 respectively. Mycelial growth inhibition results show that *F. verticilloides* is more sensitive to plants extracts with hyphal inhibition percentages ranging between the highs of 37.29%–47.17% compared to both *Aspergillus flavus* (29.73–38.15%) and *Aspergillus ochraceous* (26.33–31.04%). Similar findings were previously reported, where mycotoxigenic members of *Fusarium spp.* were found to be more sensitive to crude acetone plant extracts than mycotoxigenic members of *Aspergillus spp.* (Thembo et al., 2010).

Free radical-scavenging activity of DPPH and ABTS results are summarised in (Table 7), presented as inhibitory concentration leading to 50% reduction of free radicals (IC₅₀). The results of the tested plant extracts ranged from the lowest IC₅₀ values of 0.03 and 0.01 mg/ml and the highest IC₅₀ values of 0.87 and 0.33 mg/ml against free radicals of DPPH and ABTS respectively. The extracts of *A. oppositifolia*, *A. affra*, *C. caffrum*, *R. communis* and *S. aculeastrum* had good antioxidant activities against ABTS with IC₅₀ values of 0.01 mg/ml, while *B. salicina*, *C. caffrum*, *C. dentata*, *F. natalensis* and *H. caffrum* showed potent antioxidant activity against free radicals of DPPH.

Medicinal plants extracts from some species which includes *B. salicina*, *C. dentata*, *C. caffrum*, *R. communis*, *S. aculeastrum* exhibited reasonable antifungal and antioxidant activity. Prakash et al. (2015) found the relationship between the antifungal and antioxidant activities of isolated essential oils of *Curcuma longa* and *Zingiber officinale* which

Table 4Percentage mycelia growth inhibition (%MGI) of varying concentrations of acetone extracts on *Fusarium verticilloides*.

Plant name	3days			6days			9 days		
	MIC	0.5MIC	0.25MIC	MIC	0.5MIC	0.25MIC	MIC	0.5MIC	0.25MIC
<i>A. oppsitifolia</i>	19.77 ± 0.69	18.50 ± 0.68	12.29 ± 0.20	30.84 ± 0.63	26.66 ± 0.00	4.99 ± 0.51	20.56 ± 0.33	11.08 ± 0.00	2.76 ± 0.53
<i>A. affra</i>	46.78 ± 0.53	38.25 ± 0.05	29.22 ± 0.82	45.77 ± 0.84	36.90 ± 0.84	27.38 ± 0.00	32.29 ± 0.00	20.60 ± 0.12	7.13 ± 0.00
<i>A. dimidiata</i>	16.51 ± 0.16	10.05 ± 0.00	5.29 ± 0.13	18.20 ± 0.61	8.87 ± 0.01	1.21 ± 0.09	2.99 ± 0.34	0.25 ± 0.27	0.38 ± 0.06
<i>B. discolor</i>	25.30 ± 0.17	21.29 ± 0.15	8.32 ± 0.16	28.21 ± 0.82	23.71 ± 0.31	10.12 ± 0.40	22.62 ± 0.70	17.17 ± 0.21	6.27 ± 0.00
<i>B. galpinii</i>	5.14 ± 0.50	3.08 ± 0.00	2.65 ± 0.00	3.02 ± 0.57	3.14 ± 0.00	1.06 ± 0.27	0.46 ± 0.14	0.77 ± 0.38	0.83 ± 0.49
<i>B. salcina</i>	19.62 ± 0.25	6.87 ± 0.21	2.15 ± 0.13	19.53 ± 0.22	5.51 ± 0.99	5.02 ± 0.00	15.05 ± 0.00	14.25 ± 0.50	6.58 ± 0.07
<i>C. cafferum</i>	6.66 ± 0.00	2.57 ± 0.56	3.82 ± 0.27	6.88 ± 0.00	11.37 ± 0.68	0.38 ± 0.63	3.33 ± 0.00	1.26 ± 0.60	0.27 ± 0.84
<i>C. dendata</i>	37.29 ± 0.89	30.40 ± 0.00	19.19 ± 1.00	24.61 ± 0.00	25.28 ± 0.23	11.10 ± 0.58	18.61 ± 0.00	8.83 ± 0.23	7.96 ± 0.58
<i>C. tamentosa</i>	19.02 ± 0.57	13.85 ± 0.85	20.19 ± 0.00	22.06 ± 0.46	21.61 ± 0.08	19.24 ± 0.24	15.83 ± 0.31	9.13 ± 0.69	12.40 ± 0.77
<i>D. mannii</i>	4.12 ± 0.82	3.91 ± 0.00	4.37 ± 0.39	5.94 ± 0.00	3.74 ± 0.63	1.74 ± 0.00	0.40 ± 0.95	0.16 ± 0.95	0.35 ± 0.95
<i>F. natalensis</i>	16.20 ± 0.01	14.23 ± 0.74	11.66 ± 0.54	5.65 ± 0.66	8.62 ± 0.30	5.62 ± 0.63	0.47 ± 0.00	0.16 ± 0.54	0.93 ± 0.74
<i>H. arbroscences</i>	22.61 ± 0.60	19.08 ± 0.64	17.17 ± 0.96	14.59 ± 0.92	15.87 ± 0.63	12.97 ± 0.58	9.56 ± 0.86	7.87 ± 0.00	10.77 ± 0.84
<i>H. cafferum</i>	17.73 ± 0.39	3.44 ± 0.54	6.86 ± 0.19	7.36 ± 0.29	1.27 ± 0.92	4.30 ± 0.24	1.32 ± 0.41	0.77 ± 0.42	2.08 ± 0.27
<i>K. wilmsii</i>	47.17 ± 0.17	36.28 ± 0.00	23.89 ± 0.65	50.08 ± 0.53	33.36 ± 0.17	23.03 ± 0.64	13.73 ± 0.00	6.75 ± 0.41	8.53 ± 0.30
<i>M. aethopicum</i>	41.58 ± 0.10	20.66 ± 0.61	5.92 ± 0.52	35.78 ± 0.04	14.94 ± 1.00	2.21 ± 0.21	35.90 ± 0.56	26.40 ± 0.00	17.35 ± 0.00
<i>M. grandis</i>	22.51 ± 0.00	19.14 ± 0.50	21.33 ± 0.97	12.68 ± 1.00	8.61 ± 0.59	13.43 ± .00	8.45 ± 0.00	5.17 ± 0.51	9.12 ± 0.69
<i>M. obstufolia</i>	21.82 ± 0.12	15.79 ± 0.00	10.47 ± 0.10	6.04 ± 0.88	9.72 ± 0.00	0.50 ± 0.20	4.34 ± 0.54	0.79 ± 0.94	0.31 ± 0.85
<i>M. undata</i>	27.65 ± 0.58	16.62 ± 0.25	12.66 ± 0.17	17.22 ± 0.76	10.76 ± 0.64	6.59 ± 0.65	4.73 ± 0.54	0.37 ± 0.76	0.27 ± 0.17
<i>R. communis</i>	19.06 ± 0.29	7.62 ± 0.26	2.70 ± 0.50	18.84 ± 0.47	3.93 ± .30	2.58 ± 0.33	18.12 ± 0.53	4.10 ± 0.48	0.79 ± 0.00
<i>S. aculeastrum</i>	38.11 ± 0.17	31.97 ± 0.61	17.01 ± 0.90	34.32 ± 0.16	33.71 ± 0.20	24.55 ± 0.80	29.16 ± 0.49	28.87 ± 0.61	25.79 ± 0.69
<i>S. africana</i>	8.14 ± 1.00	5.82 ± 0.00	6.10 ± 0.69	6.57 ± 0.67	3.87 ± 0.77	4.55 ± 0.67	0.87 ± 0.45	1.73 ± 1.00	1.48 ± 0.56
<i>S. mitis</i>	9.46 ± 0.57	4.07 ± 0.94	3.65 ± 0.82	7.69 ± 0.55	3.38 ± 0.79	2.00 ± 0.38	1.63 ± 0.00	0.27 ± 0.13	0.56 ± 0.58
<i>W. salutaris</i>	35.65 ± 0.00	15.05 ± 0.04	4.93 ± 0.46	14.61 ± 0.00	10.57 ± 0.73	10.27 ± 0.09	5.27 ± 0.51	7.10 ± 0.00	2.62 ± 0.43
<i>X. kraussiana</i>	20.01 ± 0.82	6.87 ± 0.85	5.73 ± 0.38	24.20 ± 0.29	13.62 ± 0.60	5.05 ± 0.51	4.49 ± 0.62	0.99 ± 0.75	0.53 ± 0.51
<i>Z. capense</i>	10.67 ± 0.17	9.36 ± 0.63	3.36 ± 0.00	6.90 ± 0.51	4.29 ± 0.50	2.86 ± 0.50	0.25 ± 0.48	1.98 ± 0.54	1.36 ± 0.6
Amphotericin B	66.39 ± 0.68	60.92 ± 0.84	56.65 ± 0.62	55.93 ± 0.04	49.01 ± 0.50	44.88 ± 0.71	49.38 ± 0.56	43.43 ± 0.47	39.13 ± 0.24

Bold value indicates noteworthy activity.

Data represents average radial mycelia growth inhibition ± standard deviation, n = 3.

also inhibited the secretion of aflatoxin from *Aspergillus flavus*. The findings in this study also proved Tian et al. (2011) hypothesis stating that the inhibition of mycotoxins by mycotoxigenic fungi is attributed to reduced fungal growth and inhibition of key enzymes during mycotoxin production.

Studies show that phytochemicals with antioxidant activity may be used as food preservatives and prevent lipid oxidation which ultimately leads to food spoilage (Prakash et al., 2015). A lower IC₅₀ value indicates higher antioxidant activity and better protection against oxidation. Antioxidant capacity is widely used as parameter to measure bioactive

Table 5Percentage mycelia growth inhibition (%MGI) of varying concentrations of acetone extracts on *Aspergillus flavus*.

Plant Name	3days			6days			9 days		
	MIC	0.5MIC	0.25MIC	MIC	0.5MIC	0.25MIC	MIC	0.5MIC	0.25MIC
<i>A. oppsitifolia</i>	12.29 ± 0.17	10.91 ± 0.21	10.55 ± 0.00	6.37 ± 0.68	3.34 ± 0.33	3.42 ± 0.08	2.63 ± 0.21	0.67 ± 0.79	1.23 ± 0.54
<i>A. affra</i>	13.62 ± 0.04	9.57 ± 0.10	13.28 ± 0.16	11.55 ± 0.28	12.33 ± 0.24	9.50 ± 0.28	4.66 ± 0.54	4.85 ± 0.33	2.50 ± 0.66
<i>A. dimidiata</i>	30.79 ± 0.00	26.37 ± 0.51	22.70 ± 0.34	25.51 ± 0.56	23.95 ± 0.31	22.36 ± 0.00	17.06 ± 0.00	15.33 ± 0.59	12.99 ± 0.00
<i>B. discolor</i>	8.64 ± 0.16	10.67 ± 0.05	3.75 ± 0.74	0.13 ± 0.41	1.50 ± 0.27	1.47 ± 0.51	4.12 ± 0.15	4.30 ± 0.00	6.50 ± 0.00
<i>B. galpinii</i>	3.00 ± 0.49	0.08 ± 0.15	3.55 ± 0.78	0.40 ± 0.20	0.57 ± 0.00	0.94 ± 0.00	2.21 ± 0.47	2.23 ± 0.33	7.71 ± 8.71
<i>B. salcina</i>	5.75 ± 0.07	1.70 ± 0.97	2.62 ± 0.19	0.49 ± 0.25	0.14 ± 1.00	1.80 ± 0.61	3.56 ± 1.06	0.96 ± 0.56	0.12 ± 0.48
<i>C. cafferum</i>	13.64 ± 0.20	15.23 ± 0.07	11.61 ± 0.61	7.34 ± 0.01	2.99 ± 0.86	5.05 ± 0.18	2.80 ± 0.39	1.95 ± 0.18	2.73 ± 0.24
<i>C. dendata</i>	8.35 ± 0.40	1.72 ± 0.18	0.72 ± 0.06	9.13 ± 0.33	4.87 ± 0.42	4.19 ± 0.95	4.72 ± 0.99	0.41 ± 0.18	0.26 ± 0.08
<i>C. tamentosa</i>	7.97 ± 0.71	6.37 ± 0.32	3.52 ± 0.73	2.03 ± 0.00	1.69 ± 0.00	0.02 ± 0.02	0.13 ± 0.30	0.40 ± 0.43	0.14 ± 0.29
<i>D. mannii</i>	21.04 ± 0.00	15.35 ± 0.77	14.79 ± 0.25	12.41 ± 0.95	6.99 ± 0.79	2.38 ± 0.54	2.97 ± 0.72	1.92 ± 0.12	0.86 ± 0.52
<i>F. natalensis</i>	1.86 ± 0.35	6.55 ± 0.04	6.24 ± 0.49	5.14 ± 0.72	4.43 ± 0.53	1.06 ± 0.06	7.24 ± 0.00	3.36 ± 0.56	1.78 ± 0.05
<i>H. arbroscences</i>	32.13 ± 0.65	28.96 ± 0.93	19.90 ± 0.40	13.69 ± 0.74	10.31 ± 0.17	10.29 ± 0.08	2.28 ± 0.00	4.11 ± 0.38	1.45 ± 0.38
<i>H. cafferum</i>	4.73 ± 0.35	6.19 ± 0.10	5.49 ± 0.35	0.91 ± 0.00	2.21 ± 0.55	3.18 ± 0.81	4.97 ± 0.00	1.08 ± 0.37	2.43 ± 0.92
<i>K. wilmsii</i>	12.09 ± 0.07	5.91 ± 0.07	1.89 ± 0.61	19.35 ± 0.79	2.23 ± 0.09	0.11 ± 0.39	4.00 ± 0.56	4.14 ± 0.38	0.76 ± 0.09
<i>M. aethopicum</i>	38.15 ± 0.26	31.64 ± 0.21	25.46 ± 0.33	35.52 ± 0.13	23.18 ± 0.56	16.58 ± 0.00	14.73 ± 0.74	11.99 ± 0.00	15.04 ± 0.68
<i>M. grandis</i>	8.26 ± 0.00	9.17 ± 0.00	5.94 ± 0.27	3.47 ± 0.00	0.63 ± 0.17	0.51 ± 0.59	2.04 ± 0.23	1.62 ± 0.63	3.35 ± 0.29
<i>M. obstufolia</i>	18.38 ± 0.19	5.53 ± 0.26	7.84 ± 0.38	8.11 ± 0.28	0.42 ± 0.00	0.02 ± 0.33	7.26 ± 0.24	6.78 ± 0.57	7.06 ± 0.07
<i>M. undata</i>	8.35 ± 0.40	1.72 ± 0.18	0.72 ± 0.06	3.55 ± 0.29	2.56 ± 0.69	0.79 ± 0.00	6.51 ± 4.69	3.73 ± 2.78	2.68 ± 4.22
<i>R. communis</i>	9.38 ± 0.04	5.13 ± 0.10	9.02 ± 0.16	4.02 ± 0.49	2.03 ± 0.34	2.64 ± 0.46	0.81 ± 0.29	0.67 ± 0.74	4.16 ± 0.61
<i>S. aculeastrum</i>	11.40 ± 0.28	3.77 ± 0.41	5.59 ± 0.25	7.02 ± 0.38	4.00 ± 0.48	0.58 ± 0.04	3.63 ± 0.60	4.49 ± 0.66	1.28 ± 0.00
<i>S. africana</i>	19.17 ± 0.07	13.72 ± 0.10	10.94 ± 0.44	6.57 ± 0.00	2.14 ± 0.35	1.18 ± 0.93	4.48 ± 0.39	3.93 ± 0.00	2.14 ± 0.00
<i>S. mitis</i>	11.35 ± 0.11	7.47 ± 0.80	1.30 ± 0.84	7.66 ± 0.15	1.74 ± 0.49	1.03 ± 0.66	1.36 ± 0.51	0.65 ± 0.37	0.66 ± 0.17
<i>W. salutaris</i>	29.73 ± 0.58	25.71 ± 0.18	16.64 ± 0.40	11.12 ± 0.00	11.88 ± 0.17	8.40 ± 0.59	3.07 ± 0.00	3.78 ± 0.74	7.16 ± 0.61
<i>X. kraussiana</i>	5.52 ± 0.27	2.88 ± 0.19	0.77 ± 0.70	5.54 ± 0.00	2.21 ± 0.35	0.42 ± 0.41	5.82 ± 0.31	3.16 ± 0.36	1.55 ± 0.93
<i>Z. capense</i>	12.54 ± 0.00	7.66 ± 0.96	2.57 ± 0.03	7.25 ± 1.54	1.63 ± 0.24	0.22 ± 0.43	6.15 ± 0.54	3.10 ± 0.46	1.70 ± 0.75
Amphotericin B	59.55 ± 0.31	55.40 ± 0.02	50.29 ± 0.53	55.15 ± 0.48	52.34 ± 0.37	50.74 ± 0.13	42.68 ± 0.29	41.72 ± 0.64	40.22 ± 0.34

Bold value indicates noteworthy activity.

Data represents average radial mycelia growth inhibition ± standard deviation, n = 3.

Table 6Percentage mycelia growth inhibition (%MGI) of varying concentrations of acetone extracts on *Aspergillus ochraceus*.

Plant Name	3days			6days			9 days		
	MIC	0.5MIC	0.25MIC	MIC	0.5MIC	0.25MIC	MIC	0.5MIC	0.25MIC
<i>A. oppositifolia</i>	1.32 ± 0.00	6.02 ± 0.32	0.47 ± 0.23	2.19 ± 0.00	1.91 ± 0.00	0.72 ± 0.22	0.09 ± 0.84	1.09 ± 0.70	0.92 ± 0.00
<i>A. affra</i>	18.57 ± 0.41	16.19 ± 0.13	10.33 ± 0.15	18.69 ± 0.68	9.77 ± 0.05	1.71 ± 0.39	18.89 ± 0.34	11.69 ± 0.09	2.72 ± 0.51
<i>A. dimidiata</i>	4.61 ± 0.39	3.83 ± 0.00	0.99 ± 0.32	2.54 ± 0.00	3.09 ± 0.14	1.62 ± 0.033	1.21 ± 0.42	1.50 ± 0.22	1.18 ± 0.12
<i>B. discolor</i>	13.47 ± 0.00	8.28 ± 0.00	5.94 ± 0.45	7.25 ± 0.51	3.89 ± 0.00	1.03 ± 0.58	10.94 ± 0.42	6.59 ± 0.00	4.10 ± 0.00
<i>B. galpinii</i>	3.38 ± 1.57	1.23 ± 1.72	4.04 ± 0.06	3.41 ± 1.84	2.79 ± 2.05	1.33 ± 0.13	2.41 ± 1.36	0.85 ± 1.02	0.24 ± 0.23
<i>B. salicina</i>	26.33 ± 0.76	27.19 ± 0.17	23.81 ± 0.23	25.52 ± 0.60	23.50 ± 0.00	21.41 ± 0.40	16.50 ± 0.96	14.53 ± 0.34	12.47 ± 0.31
<i>C. cafferum</i>	5.00 ± 0.97	3.12 ± 0.51	6.12 ± 0.50	10.85 ± 0.12	7.31 ± 0.38	8.06 ± 0.00	3.12 ± 0.00	0.49 ± 0.30	5.21 ± 0.64
<i>C. dendata</i>	14.20 ± 0.51	10.79 ± 0.00	14.14 ± 0.14	5.58 ± 0.62	5.57 ± 0.31	6.67 ± 0.84	4.29 ± 1.38	6.44 ± 0.47	6.20 ± 1.38
<i>C. tamentosa</i>	9.04 ± 0.66	6.12 ± 0.59	2.29 ± 0.00	3.63 ± 0.00	1.51 ± 0.56	1.36 ± 0.14	1.51 ± 0.00	0.54 ± 0.40	0.02 ± 0.35
<i>D. mannii</i>	27.99 ± 0.22	28.00 ± 0.57	25.37 ± 0.70	11.42 ± 0.00	13.94 ± 0.00	12.50 ± 0.77	11.30 ± 0.98	11.44 ± 0.64	7.92 ± 0.67
<i>F. natalensis</i>	11.54 ± 0.36	11.28 ± 0.51	9.39 ± 0.09	9.91 ± 0.00	0.47 ± 0.00	0.05 ± 0.07	1.84 ± 0.11	0.84 ± 0.00	0.63 ± 0.55
<i>H. arborescences</i>	16.11 ± 0.62	11.95 ± 0.61	6.86 ± 0.67	14.92 ± 0.51	12.83 ± 0.00	5.20 ± 0.89	13.28 ± 0.43	11.54 ± 0.72	3.41 ± 0.29
<i>H. cafferum</i>	16.29 ± 0.90	9.53 ± 0.00	1.76 ± 0.13	9.96 ± 0.25	7.15 ± 0.00	4.49 ± 0.00	2.63 ± 0.00	1.33 ± 0.87	0.43 ± 0.57
<i>K. wilmsii</i>	7.34 ± 0.15	5.63 ± 0.73	2.21 ± 0.59	4.06 ± 0.31	5.36 ± 0.22	1.40 ± 0.20	1.68 ± 0.62	0.36 ± 0.52	0.18 ± 0.26
<i>M. aethiopicum</i>	11.28 ± 0.03	10.76 ± 0.03	3.71 ± 0.74	0.24 ± 0.22	2.18 ± 0.18	2.35 ± 0.17	3.77 ± 0.00	3.80 ± 0.00	2.84 ± 3.90
<i>M. grandis</i>	14.58 ± 0.49	11.05 ± 0.00	12.48 ± 0.86	5.05 ± 0.64	6.10 ± 0.67	6.70 ± 0.86	5.84 ± 0.00	6.02 ± 1.38	4.94 ± 0.38
<i>M. obtusifolia</i>	20.40 ± 0.50	21.40 ± 0.23	26.92 ± 0.02	18.05 ± 0.85	10.66 ± 0.85	10.06 ± 0.54	6.51 ± 0.00	3.73 ± 0.00	2.68 ± 0.00
<i>M. undata</i>	10.35 ± 0.10	11.43 ± 0.37	2.21 ± 0.09	7.04 ± 0.00	6.61 ± 0.09	2.07 ± 0.19	2.63 ± 0.35	1.83 ± 0.35	1.39 ± 0.40
<i>R. communis</i>	3.65 ± 0.42	3.50 ± 0.28	0.33 ± 0.85	3.54 ± 0.29	3.15 ± 0.59	1.11 ± 0.01	4.19 ± 0.00	1.33 ± 0.05	1.25 ± 0.89
<i>S. aculeastrum</i>	15.36 ± 0.71	10.46 ± 0.16	5.27 ± 0.45	6.79 ± 0.82	6.59 ± 0.08	1.76 ± 0.08	5.24 ± 0.00	4.78 ± 0.00	1.07 ± 0.81
<i>S. africana</i>	31.04 ± 0.02	24.88 ± 0.50	25.82 ± 0.23	18.05 ± 0.85	10.66 ± 0.85	10.06 ± 0.54	6.51 ± 0.00	3.73 ± 0.00	2.68 ± 0.00
<i>S. mitis</i>	24.69 ± 0.26	21.41 ± 0.00	19.46 ± 0.38	14.88 ± 1.31	2.47 ± 0.82	1.00 ± 0.64	2.07 ± 0.29	1.89 ± 0.56	0.75 ± 0.50
<i>W. salutaris</i>	28.59 ± 0.27	20.28 ± 0.32	13.37 ± 0.84	35.75 ± 0.29	22.62 ± 0.69	16.49 ± 0.00	14.73 ± 0.74	3.69 ± 0.62	0.24 ± 0.00
<i>X. kraussiana</i>	7.34 ± 0.32	5.63 ± 0.07	2.21 ± 0.12	10.07 ± 0.25	6.91 ± 0.11	7.44 ± 0.27	9.16 ± 0.13	8.76 ± 0.34	6.17 ± 2.57
<i>Z. capense</i>	8.00 ± 0.38	6.51 ± 0.97	0.51 ± 0.51	7.01 ± 0.62	6.01 ± 0.93	1.50 ± 0.82	4.40 ± 0.00	2.50 ± 0.52	0.85 ± 0.34
Amphotericin B	61.94 ± 0.31	57.67 ± 0.02	52.30 ± 0.47	54.94 ± 0.73	53.12 ± 0.15	51.10 ± 0.69	42.67 ± 0.62	42.66 ± 0.31	43.33 ± 0.84

Bold value indicates noteworthy activity.

Data represents average radial mycelia growth inhibition ± standard deviation, n = 3.

and functional constituents in food commodities (Sowndhararajan and Kang, 2013).

This suggests that the extracts contain phyto-compounds with some antioxidant activity capable of scavenging imbalanced reactive oxygen species (ROS), which are considered major cause of many diseases such

Table 7

The 2,2-Diphenyl-1-picrylhydrazyl and 2,2'-azinobis (3-ethylbenzthiazoline-6-sulphonic acid) radical scavenging activity assay results.

Plant species	DPPH	ABTS
<i>Acokanthera oppositifolia</i>	0.64 ± 0.54	0.01 ± 0.00
<i>Apodytes dimidiata</i>	0.23 ± 0.07	0.03 ± 0.00
<i>Artemesia affra</i>	0.30 ± 0.12	0.01 ± 0.00
<i>Bauhinia galpini</i>	0.43 ± 0.02	0.05 ± 0.03
<i>Brachylaena discolor</i>	0.17 ± 0.06	0.03 ± 0.01
<i>Breonadia salicina</i>	0.07 ± 0.02	0.33 ± 0.24
<i>Capparis tamentosa</i>	0.39 ± 0.16	0.08 ± 0.02
<i>Combretum cafferum</i>	0.07 ± 0.01	0.01 ± 0.00
<i>Curtisia dentata</i>	0.03 ± 0.01	0.03 ± 0.09
<i>Dracaena mannii</i>	0.16 ± 0.02	0.03 ± 0.00
<i>Ficus natalensis</i>	0.06 ± 0.03	0.05 ± 0.03
<i>Harpephyllum cafferum</i>	0.06 ± 0.04	0.04 ± 0.01
<i>Heteromorpha arborescens</i>	0.65 ± 0.45	0.10 ± 0.00
<i>Kirkia wilmsii</i>	0.13 ± 0.01	0.04 ± 0.01
<i>Markhamia obtusifolia</i>	0.20 ± 0.00	0.02 ± 0.01
<i>Maytenus undata</i>	0.35 ± 0.02	0.05 ± 0.00
<i>Milletia grandis</i>	0.87 ± 0.05	0.06 ± 0.04
<i>Mystroxydon aethiopicum</i>	0.16 ± 0.05	0.02 ± 0.01
<i>Ricinus communis</i>	0.35 ± 0.09	0.01 ± 0.00
<i>Solanum aculeastrum</i>	0.17 ± 0.08	0.01 ± 0.00
<i>Spirostachys africana</i>	0.23 ± 0.12	0.10 ± 0.03
<i>Strychnos mitis</i>	0.46 ± 0.01	0.05 ± 0.00
<i>Warburgia salutaris</i>	0.38 ± 0.09	0.06 ± 0.00
<i>Xylotheca kraussiana</i>	0.15 ± 0.01	0.03 ± 0.00
<i>Zanthoxylum capense</i>	0.53 ± 0.35	0.10 ± 0.01
L-Ascorbic acid	0.001 ± 0.00	0.003 ± 0.00

Bold value indicates noteworthy activity.

Data presented as IC₅₀ in mg/ml ± standard error, n = 3.

as cancer, cardiovascular diseases, neurodegenerative diseases, hypertension and AIDS. The antioxidant compounds can also be used to prevent lipid peroxidation in food commodities (Amorati and Valgimigli, 2015; Nunes et al., 2012). Lipid peroxidation is a complex aerobic cellular process where oxygen interacts with polyunsaturated fatty acids resulting in reduction of food shelf life (Prakash et al., 2015). Plant based extracts are reported to contain phytochemicals such as phenolic compounds that may be for responsible for antioxidant activity (Moyo et al., 2010). Extracts of *A. oppositifolia*, *A. affra*, *C. cafferum*, *R. communis* and *S. aculeastrum* had good antioxidant activities against ABTS with IC₅₀ values of 0.01 mg/ml, While *B. saligna*, *C. cafferum*, *C. dendata*, *F. natalensis*, *H. cafferum* showed potent antioxidant activity against free radicals of DPPH.

Earlier, the antioxidant activity of methanol extracts from *C. dentata*, 1:1 methanol: dichloromethane extracts from *Solanum aculeastrum*, *Bauhinia galpinii*, *Ricinus communis* and *Warburgia salutaris* against DPPH have been reported (Fadipe et al., 2015; Mongalo et al., 2018). It is not easy to compare these results due to differences in terms of solvents used, collection site, geographical areas and collection times. However, these plants extracts are promising inhibitors of free radicals that may well be used to curb oxidation in various agricultural products.

GC-MS analysis of most active antifungal and potentially active medicinal plants is reported in Tables 8 and 9. *Curtisia dentata* revealed the presence of triterpenoids such as β-amyryn (53.30%), α-amyryn (6.42%), β-sitosterol (2.47%) and vitamin E (4.99%), while *Markhamia obtusifolia* yielded the presence of neophytadiene (4.38%), palmitic acid (3.61%) and 4-(1E)-3-Hydroxy-1-propenyl)-2-methoxyphenol (2.04%). Triterpenes are well known to possess potential antifungal activity against both human and crop infecting fungal strains (Liu et al., 2015; Kongue et al., 2013; Hu et al., 2018).

Some of the detected compounds from *Curtisia dentata* extracts are reported to possess potential antimicrobial activities. For instance, α-amyryn and β-sitosterol are well known for their antifungal activity against *Aspergillus flavus* and *Aspergillus niger* (Singh and Singh, 2003). In addition, β-amyryn and its derivatives exhibited potential antifungal

Table 8Phytochemical profiling of acetone extract of *Curtisia dentata* using GC-MS.

No.	RT	Compound name	Classification	Peak area (%)
1	6.79	1,2,3-Benzenetriol	Benzenetriol	0.50
2	8.09	1,6-Anhydro-beta-D-glucopyranose (Levogluconan)	Carbohydrate	1.86
3	8.25	Myristicin	Phenylpropene	0.19
4	9.02	4-Propylpyrocatechol	Benzene derivative	1.08
5	10.81	Myristic acid (Tetradecanoic acid)	Fatty acid	0.36
6	11.85	Neophytadiene	Terperenoid	1.34
7	11.93	6,10,14-Trimethyl-2-pentadecanone	Essential oil	0.35
8	12.46	Phytol	Diterpene	0.38
9	13.63	Hexadecanoic acid (Palmitic acid)	Fatty acid	1.60
10	16.41	2-sec-Butylcyclopentanone	Hydrocarbon	0.32
11	19.67	4,8,12,16-Tetramethylheptadecan-4-olide	Diterpene	0.59
12	25.56	1-Docosene	Alkene	1.29
13	27.39	Pentacosane	Akane	0.86
14	28.20	β -Tocopherol (Vitamin E isomer)	Vitamin	0.34
15	28.32	γ -Tocopherol (Vitamin E)	Vitamin	1.63
16	28.46	4-Methylcholesta-8,14,24-trien-3-ol	Cholesterol	1.07
17	28.60	β -Sitosterol acetate	Triterpenoid	1.42
18	28.67	1-Eicosanol	Arachidyl alcohol	0.55
19	28.83	Vitamin E	Vitamin	4.99
20	29.63	1,30-Triacontanediol	Fatty alcohol	0.64
21	29.75	Stigmastan-4-one, 3-methoxy-, (3 β .,24S)-	Triterpenoid	0.59
22	29.94	7-Oxcholestan-3-yl acetate	Acetate derivative	0.29
23	30.25	γ -Sitosterol	Triterpenoid	2.47
24	30.40	Ursa-9(11),12-dien-3-ol	Triterpenoid	0.90
25	30.66	α-Amyrin	Triterpenoid	6.42
26	30.79	1,2-dihydroxybenzene (catechol)	benzenediol	1.07
27	31.15	β-Amyrin	Triterpenoid	53.30
28	32.23	Betulin	Triterpenoid	0.73
29	32.64	Hopenone b	Lipid	0.60
30	32.88	2-Azafluoranthene	Polycyclic nitrogen fraction	0.33
31	33.08	24-Methyl-9,19-cyclolanost-25-en-3-ol	Hydroxy steroid	2.56

Bold value indicates noteworthy activity.

activity against various *Candida* species through the inhibition of adhesion of the fungus to the cell (Johann et al., 2007). Furthermore, such triterpenes may well possess anticancer, anti-inflammatory, anti-oxidative and anti-viral activities (Chudzik et al., 2015). Earlier in our studies, β -sitosterol and betulinic acid from methanol extract of the leaves of *C. dentata* were reported to possess potent anti-mycobacterial activity and no cytotoxicity to both HepG2 and HEK293 cell lines (Mongalo et al., 2016; Fadipe et al., 2015). Furthermore, betulinic acid and lupeol yielded a potent anti-parasitic effect (Opoku et al., 2016). The antifungal activity of *Curtisia dentata* as reported in the current paper may well be attributed to the triterpenes as identified abundantly in the GC-MS chemical spectra. The mode of action of the compounds may be through mycelial growth inhibition. However, other studies suggested that the compounds may act through cell membrane damage, compromising the integrity and permeability of fungal cells (Haraguchi et al., 1999; Leite et al., 2014) and possible leakage of cations from the cytoplasm (Wong et al., 2019).

The acetone extract from *Markhamia obtusifolia* leaves revealed the presence of ursolic acid [3 β -hydroxyurs-12-en-28-oic acid], pomolic acid [3 β ,19 α -dihydroxy-urs-12-en-28-oic acid], 2-epi-tormentonic acid [2 β ,3 β ,19 α -trihydroxy-urs-12-en-28-oic acid] (Eloff et al., 2008; Nchu et al., 2010). Contrarily, the plant species in the current study revealed the presence of palmitic acid and neophytadiene as major constituents of the plant species. The bioactivity of the plant species may well be

Table 9Phytochemical profiling of acetone extract of *Markhamia obtusifolia*.

No.	RT	Compound name	Type of compound	Peak area (%)
1	7.43	2,1,3-Benzothiadiazole (Piazthiole)	Benzene derivative	1.05
2	7.57	n-Tridecane	Alkane	0.80
3	8.05	2,4-di-tert-butylphenol	Phenol	1.13
4	10.67	4-((1E)-3-Hydroxy-1-propenyl)-2-methoxyphenol (Coniferol)	Phenylpropanoid	2.04
5	10.83	Myristic acid	Fatty acid	0.85
6	11.17	Loliolide	Lactone	0.48
7	11.86	Neophytadiene	sesquiterpenoids	4.38
8	11.94	6,10,14-Trimethyl-2-pentadecanone	Sesquiterpenoids	0.83
9	13.05	4-Ethylcyclohexanone	Saturated hydrocarbon	0.49
10	13.68	Palmitic acid	Fatty acid	3.61
11	14.13	1-(1-Hydroxybutyl)-2,5-dimethoxybenzene	Phenethylamine	0.75
12	15.79	γ -Decalactone	Lactone/aroma compound	0.23
13	15.86	2,2'-methylenebis-cyclopentanone,	Methane derivative	0.36
14	15.93	Phytol	Diterpene	0.24
15	16.36	Oleic acid	Fatty acid	0.23
16	16.42	4,4,6-Trimethyl-2-cyclohexen-1-ol	Terpene	0.71
17	16.74	Stearic acid	Fatty acid	0.40
18	17.12	6-Formyl-3-methyl-2-oxo-4-hexenoic acid	Fatty acid	0.18
19	18.27	Hahnfett	N/A	0.48
20	19.68	4,8,12,16-Tetramethylheptadecan-4-olide	Diterpene	0.34
21	19.87	Oleoamide	Fatty acid amide	0.27
22	24.82	3-phenyl-Benzo[f][1,7]naphthyridin-5(6H)-one	Alkaloid	0.24
23	25.40	exo-7-methylbicyclo[4.1.0]hept-2-ene-endo-7-	Cyclopentone	0.16
24	25.55	1-Nonadecene	Alkene	0.47
25	25.60	Heneicosane	Alkane	0.13
26	26.64	PELTGERIN	N/A	0.49
27	26.93	Dimethyl 4,6-dioxo-5,6-dihydro-4H-pyrido[3,2,1-jk]carbazole-5-spirocyclohexane-1,3-dicarboxylate	Ether derivative	0.52
29	27.01	Endomethylamino-2-endobornanol-3	Amino acid derivative	0.24
31	27.39	Docosane	Alkane	1.20
32	27.90	2,6,8-trimethyl-Pyrido[3,4-d]pyrimidin-4(3H)-one	Pyrimidine derivative	0.93
33	28.02	N-methyl-N-[2-chloro-3-(1,2-epoxy-4-acetoxy-5-methylcyclohex-6-ON-YL)prop-2-enyl]-N-[4,4-dimethoxybutanoic acid]amide	N/A	1.49
35	28.21	β -Tocopherol	Vitamin	1.38

attributed to the two compounds individually or synergistically. Palmitic acid and neophytadiene from other plant species has been reported to be effective against *Aspergillus* species (Altieri et al., 2007; Felicio et al., 2010). Furthermore, these compounds possess potential antimicrobial and anti-inflammatory activity (Ivanova et al., 2017).

Although the selected medicinal plants exhibited a noteworthy anti-fungal and antioxidant activity, the cytotoxicity studies of these plants is of paramount importance and still needs to be explored. According to Zotter et al. (2019), the organic compounds may have a devastating effect on human and animal health. These may include the deterioration of functionality on essential organs that may include heart, liver, lungs and the entire central nervous system (Shankar et al., 2018). It is important to note that African Traditional Medicine use mostly water as a solvent for extracting the active compounds responsible for management of relevant

infections.

4. Conclusions

The results obtained in this study suggests that plant extracts and phyto-compounds have the potential for the development of alternative bio-fungicides and may be used as substitutes for synthetic fungicides. Given the fungicidal, fungi-toxic and antioxidant activity of the investigated plants, the plant selected medicinal plants species may have potential to be used as possible leads for the development of bio-fungicides that can also prevent oxidation related to food spoilage. However, toxicity-profiling studies of the active plants species still needs to be explored. Furthermore, *in vivo* studies, using crops as study model, are also necessary and still needs to be explored.

Declarations

Author contribution statement

P.M. Dikhoba: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data.

N.I. Mongalo: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

E.E. Elgorashi: Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

T.J. Makhafole: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data.

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Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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