https://doi.org/10.48047/AFJBS.6.15.2024.10478-10508

African Journal of Biological Sciences

Journal homepage: http://www.afjbs.com

Research Paper Open Access

PHYTOCHEMICAL ANALYSIS AND ANTIFUNGAL ACTIVITY OF *PARKIA BIGLOBOSA* AND *EUCALYPTUS CAMALDULENSIS* (STEU.)

Boubacar Abdou Soumana [1, 4]* **Abdoulkarim Issa Ibrahim**[2], ***Ifeanyi Famous Ossamulu***[3], [3]***Muhammad Muhammad Wun***[4], ***Helen Shnada Auta*** [1], ***Hadiza Lami Muhammad***[1], ***Hussein Anthony Makun***[1]

[1]Africa Centre of excellence for mycotoxin and food safety ACEMFS/Fut Minna
[2]Department of Agronomy and Food Technology, University Boubakar Ba of Tillaberi, Niger republic
[3]Department of Biochemistry, Federal University of Technology Minna, Nigeria;
[4]Department of Microbiology, Federal University of Technology, P. M.B. 65, Minna, Niger state, Nigeria

*__Corresponding author__: soumana_bas@yahoo.fr

Volume 6, Issue 15, Sep 2024

Received: 15 July 2024

Accepted: 25 Aug 2024

Published: 25 Sep 2024

doi: 10.48047/AFJBS.6.15.2024.10478-10508

ABSTRACT

Plants offer a promising source of bioactive compounds for developing antimicrobial agents, amid growing concerns about harmful chemical fungicides, driving a shift towards organic and cost-effective solutions. This study assessed the potential of Parkia biglobosa and Eucalyptus camaldulensis leaf extracts as antifungal agents against sorghum fungal contaminants. Fresh leaves of P. biglobosa and E. camaldulensis were collected, authenticated and extracted using the cold maceration method. Following the determination of the extracts' phytochemical profiles by conventional procedures, compounds were identified and quantified using gas chromatography- mass spectrometry (GC-MS) analysis. The agar well diffusion method was used to assess the extracts' antifungal efficacy against fungus species isolated from sorghum grains. A wide variety of bioactive chemicals were found in the ethanolic leaf extracts during phytochemical screening in both extracts. E. camaldulensis displayed higher concentrations of phenols (21.32mg/g), alkaloids (7.29mg/g), saponins (13.71mg/g), tannins (12.88mg/g), carbohydrates (18.87mg/g), and reducing sugars (8.77mg/g) while P. biglobosa extracts were richer in flavonoids (13.01mg/g), terpenoids (8.61mg/g), and glycosides (5.92mg/g). GC-MS analysis identified a various compound in the extracts, with 32 detected in E. camaldulensis and 27 in P. biglobosa. Notably, E. camaldulensis contained compounds like eugenol, 4-(3-hydroxybutyl) phenol, and 1,2-dehydroviridiflorol, while P. biglobosa contained 1,2,3-benzenetriol, 2-naphthalenol, and oleic acid. The antifungal activity was concentration dependent as inhibition zones increases with increasing concentration of the extract. E. camaldulensis displayed higher activity, inhibiting the growth of P. glandicola (13 mm), F. solani (16 mm), A. glaucus (13 mm), Curvularia sp. (10 mm), and A. niger (10 mm) at 100 mg/ml. P. biglobosa extract exhibited some inhibitory effects against specific fungi, including P. glandicola (14 mm), A. glaucus (15 mm), and A. flavus (8 mm) at the same concentration. The study showed the promising antifungal potential of P. biglobosa and E. camaldulensis leaf extracts against sorghum fungal contaminants. The results of the GC-MS analysis and phytochemical screening identified a number of bioactive chemicals, indicating that the plant extracts could be used as a sustainable, affordable, and environmentally friendly substitute for synthetic fungicides in the management of fungal growth in sorghum grains.

Keywords: Parkia biglobosa, Eucalyptus camaldulensis, Phytochemical analysis, Antifungal activity, GC-MS.

1.0 INTRODUCTION

Globally, fungal contamination of stored food grains causes significant economic and health challenges (Mohapatra et al., 2017). While chemical fungicides have been traditionally used to control fungal and mycotoxin contamination of grains, concerns about their negative effect on the environmental and health risks prompts the need for finding more safer and eco-friendly alternatives (Nesci et al., 2016; Lagogianni and Tsitsigiannis, 2018; Ons et al., 2020). Plants as rich sources of organic bioactive compounds with antimicrobial properties (Dar et al., 2023; Pacyga et al., 2024), offer promising opportunity, as such this study focused on investigating the antifungal properties and bioactive compounds leaf extracts from Eucalyptus camaldulensis and Parkia biglobosa as eco-friendly alternatives to synthetic fungicides for food grain storage.

The African locust bean, or Parkia biglobosa, is a perennial deciduous tree that belongs to the Fabaceae family (Sut et al., 2024). It is a widely distributed savannah tree in Africa (Adejumo et al., 2013; Komolafe and Oyelade, 2015; Yaméogo and Yanogo, 2023). Due to its nutritional, antimicrobial, and antioxidant properties, almost all of its parts are used for a variety of purposes, from foods to medicinal agents (Chhikara et al., 2018; Arinola et al., 2019). As such, it is an important economic resource. Its seeds, bark, and leaves have been utilised in traditional medicine to treat a range of illnesses and have demonstrated promise as a source of chemotherapeutic chemicals (Abioye et al., 2013; Saleh et al., 2021). River red gum, or Eucalyptus camaldulensis, is a tree of the Myrtaceae family (Sabo and Knezevic, 2019). The oil found in the leaves of this tropical and subtropical tree is well known for its medicinal qualities. It has been reported to have multiple biological and pharmacological activities, such as antioxidant, cytotoxic, antimicrobial, larvicidal, pesticidal, and anti-dermatophytes (Sani et al., 2014; Ghareeb et al., 2018). The tree is found throughout the world.

Numerous biologically active substances, such as steroids, tannins, polyphenolics, glycosides, terpenes, alkaloids, flavonoids, saponins, and many more, have been linked to both Parkia biglobosa and Eucalyptus camaldulensis (Ghalem and Mohamed, 2014; Sani et al., 2014;

Komolafe and Oyelade, 2015; Anigboro et al., 2020). The different actions of the plants are caused by these biodegradable, non-toxic, and ecologically friendly substances, which provide an answer to the problems created by synthetic fungicides when it comes to managing fungal infestation in stored food grains.

2.0 MATERIALS AND METHODS

2.1 Chemicals: This investigation used analytical-grade chemicals from BDH Chemicals Ltd. in Poole, England.

2.2 Samples collection and Preparation: Gidan Kwano, Minna, Niger State, Nigeria is where fresh leaves of Eucalyptus camaldulensis (Steu.) and Parkia biglobosa were gathered. To assure precise identification, a botanist from the Department of Plant Biology at Federal University of Technology, Minna, Niger State, Nigeria, taxonomically authenticated the plant specimens.

2.3 Extraction of the crude extracts

Freshly picked leaves of Parkia biglobosa and Eucalyptus camaldulensis were shade-dried for a week at room temperature, in accordance with the procedure outlined by AOAC (2010). A mechanical grinder was then used to grind the dry leaves into a coarse powder. Each powdered sample was then weighed at 100 grammes and macerated for 72 hours in 500 millilitres of ethanol (1:5 w/v ratios). Whatman No. 1 filter paper was used to filter the resultant suspensions, and the filtrates were concentrated at 40°C at reduced pressure (204 mbar) until they were entirely dry.

2.4 Phytochemical Screening: The crude ethanolic extracts of samples of Eucalyptus camaldulensis and Parkia biglobosa were subjected to qualitative and quantitative phytochemical tests using standard procedures as outlined by Gupta et al. (2013), Das et al. (2014), Ejikeme et al. (2014), Ezeonu and Ejikeme (2016), Madhu et al. (2016), Saptarini et al. (2016), and Malik (2017).

2.4.1 Qualitative Phytochemical Screening

2.4.1.1 Phenols Test

The extract was pipetted into a sterile beaker in two millilitres. The extract was then mixed with a few drops of newly made neutral ferric chloride solution. The presence of phenols was indicated by the solution taking on a vivid blue-green hue.

2.4.1.2 Flavonoids Test (also known as Shindo's Test)

In a water bath kept at 40–50°C, a solution containing 1.3 mL of the extract and 0.5 g of magnesium was brought to a boil for five minutes. The solution began to turn orange to red, which suggested the presence of flavonoids**.**

2.4.1.3 Terpenoids Test

The Salkowski test was used to look into the presence of terpenoids. Five millilitres of the ethanolic extract and two millilitres of chloroform were combined in a test tube. The blend was given a light shaking. After that, a thin layer was carefully created along the test tube's side by adding 3 mL of concentrated sulfuric acid. The presence of terpenoids was shown by the creation of a reddish-brown coloration at the interface between the two layers.

2.4.1.4 Alkaloid Test

After pipetting one millilitre of the extract into a test tube, 0.5 millilitre of diluted hydrochloric acid and a few drops of Mayer's reagent were added. An alkaloid test resulted in a positive result when a white precipitate formed.

2.4.1.5 Saponins Test

The foam test was used to determine whether saponins were present. A few drops of distilled water were added to 2.5 millilitres of the filtrate. For a minute, the mixture was given a thorough shake. Saponins were present when a stable, long-lasting foam layer formed that lasted for at least a minute.

2.4.1.6 Tannins Test

The ferric chloride test was used to determine whether tannins were present. The 1:1 ethanolic extract was diluted with two to three millilitres and a few drops of 10% alcoholic ferric chloride solution. The solution developed a dark blue coloration, which suggested the presence of tannins.

2.4.1.7 Steroids Test

The Liebermann-Burchard test was used to look into the presence of steroids. The extract was poured into a test tube in one millilitre. Next, to create separate layers, 2 mL of concentrated sulfuric acid and 2 mL of acetic anhydride were carefully placed along the test tube's side. Steroids were present when the hue at the interface between the layers changed from blue or green to a reddish-brown colour.

2.4.1.8 Cardiac glycosides (Keller-Killani test)

The existence of cardenolides was investigated using the Keller-Killiani test. Two millilitres of glacial acetic acid with one drop of ferric chloride solution (FeCl3) was combined with five millilitres of each extract. Subsequently, different layers were formed by carefully adding 1 mL of concentrated sulfuric acid down the test tube's side. The presence of cardenolides would be indicated by the creation of a brown ring at the contact between the layers.

2.4.1.9 ***Carbohydrates Test***

Molisch's test was used to look into the presence of carbohydrates. In a test tube, two millilitres of Molisch's reagent (a solution of α-naphthol in ethanol) was combined with three millilitres of the extract. To ensure adequate mixing, the mixture was gently shaken. The test tube was then carefully filled with two millilitres of concentrated sulfuric acid to create two separate layers. A positive test result for the presence of carbohydrates was shown by the development of a red or violet coloration at the layer-to-layer contact.

2.4.1.10 Reducing Sugar Test (Benedict test)

A full mixture of 3 mL of distilled water and fifty milligrammes (0.05g) of the extract were weighed and combined. To get a clear filtrate, the mixture was filtered again. To 3 mL of Benedict's reagent in a test tube, three drops of the filtrate were applied. A bath of boiling water was used to heat the mixture for five minutes. After heating, the plant extract contains reducing sugars as evidenced by the production of a brick-red precipitate.

2.4.1.2 Quantitative phytochemical Screening

2.4.2.1 Total Phenol Determination

A Soxhlet apparatus was used to defatten the extract (2g) during a 2-hour period using 100ml of diethyl ether. After that, the fat-free sample was extracted for 15 minutes using 50 millilitres of petroleum ether to remove the phenolic component. A 50 ml flask was filled with a 5 ml aliquot of the extract, 10 ml of distilled water, 2 ml of ammonium hydroxide solution, and 5 ml of concentrated amyl alcohol. The liquid was diluted to the appropriate amount and left to react for thirty minutes in order to produce colour. A spectrophotometer was used to measure the absorbance at 505 nm. Tannic acid was used as the standard to create a calibration curve.

2.4.2.2 Total Flavonoid Determination

The aluminium chloride colorimetric technique was utilised to ascertain the total flavonoid content. With quercetin serving as the reference, a calibration curve was created. A 1.5 ml methanol test tube was filled with an aliquot of the diluted sample (0.5 ml), and then 0.1 ml of

10% AlCl3 solution and 0.1 ml of sodium acetate (NaCH3COO-) were added. Next, 2.8 cc of distilled water was added to the reaction mixture to dilute it. A spectrophotometer was used to measure the absorbance at 415 nm following a 30-minute incubation period at room temperature. The 10% AlCl3 solution was replaced with an equivalent volume of distilled water to create a blank.

2.4.2.3 Terpenoids Determination

100 mg sample of the dried plant extract (WI) was filtered after being steeped in 9 mL of ethanol for a whole day. Using a separating funnel, 10 mL of petroleum ether was added to the extract after it had been filtered. Once in glass vials that had been previously weighed, the ether extract was moved and given time to dry entirely (WF). The following formula was used to determine the yield (%) of total terpenoids content:

$$\frac{WI - WF}{WI} X\ 100$$

Where WI and WF represent the initial and final weights, respectively.

2.4.2.4 Total Alkaloids Determination

We dissolved 0.5g of the material in a 1:1 solution of 20% H2SO4 and 96% ethanol. After adding 1 millilitre of the filtrate to 5 millilitres of 60% tetraoxosulphate (VI), the mixture was let to stand for five minutes. After adding 5 millilitres of 0.5% formaldehyde, the mixture was allowed to sit for three hours. At 565 nm, the absorbance was measured. The alkaloid concentration was estimated using the extinction coefficient (E296, ethanol {ETOH} = 15136 M-1cm-1) of vincristine, which was utilised as the reference alkaloid.

2.4.2.5 Saponins Determination

Following a 4-hour boil in 20 millilitres of 1N HCl, 0.5 grammes of the extract were cooled, filtered, and extracted using 50 millilitres of petroleum ether. After the ether layer had evaporated completely, the residue was dissolved in five millilitres of acetone-ethanol. Six millilitres of ferrous sulphate reagent and two millilitres of concentrated H2SO4 were added to three aliquots of 0.4 millilitres each that were placed in different test tubes. Following ten minutes, the mixture was well combined, the absorbance was measured at 490 nm, and a calibration curve was created using standard saponin.

2.4.2.6 Tannin Determination

A precise 50ml beaker was filled with 0.2g of the extract. The mixture was covered with parafilm and incubated in a water bath at 77–80°C for one hour after 25 millilitres of 50% methanol was added. To guarantee homogeneity, the mixture was vigorously shook. A double layer of Whatman No. 41 filter paper was used to quantitatively filter the extract before it was transferred into a 100 ml volumetric flask. After adding 25 ml of water, 2.5 ml of Folin-Denis reagent, and 10 ml of Na2CO3, the mixture was thoroughly mixed. After thoroughly mixing the solution and diluting it with water to the appropriate level, it was let to stand for 20 minutes in order to turn bluish-green. A UV spectrophotometer (Model 752) was used to measure the absorbances of the tannic acid standard solutions and samples at 760 nm following colour development. A calibration curve was created by plotting absorbance against tannic acid concentration.

2.4.2.7 Steroids Determination

A volumetric flask with a capacity of 10ml was filled with an aliquot (1ml) of the extract. Next, 0.5 ml of a 0.5% w/v potassium hexacyanoferrate (III) solution was added, after which 2 ml of 4N sulfuric acid and 2 ml of 0.5% w/v iron (III) chloride were added. After 30 minutes of constant shaking heating in a water bath at 70±2°C, the mixture was diluted to the appropriate amount using distilled water. Using a reagent blank as a reference, the absorbance was measured at 780 nm.

2.5 Gas Chromatography-Mass spectrometry (GC-MS) analysis

A PerkinElmer Clarus 500 Model running Turbomass version 5.2 software was used to conduct the GC-MS analysis. A 30 m x 250 µm, 5% phenol, and 95% dimethylpolysiloxane Elite-5 MS column was utilised. The programme for oven temperature was configured as follows: 50ºC was the starting temperature; it raised by 8ºC/min to 220ºC after 5 minutes, and then by 7ºC/min to 280ºC for 15 minutes. The carrier gas, helium gas (99.999%), was used at a steady flow rate of 1 millilitre per minute. A 2µl portion of the material was introduced into the column at 280ºC using a 10:1 split ratio. Electron ionisation was employed, and the ionising energy was 70eV. The range of mass was 40–600 amu. 150ºC was the source temperature and 200ºC was the inlet line temperature. The GC ran for fifty minutes in total.

2.5.1 Compound identification: The compounds were identified by mass spectra comparison with the NIST 2005 database. There are more than 62,000 patterns in the database. The components of the test substance were identified by name, molecular weight, molecular formula,

and structure (Olivia et al., 2021).

2.6 Fungal Isolates Used in the Study

In this study, pure cultures of various fungal isolates, namely *Aspergillus niger, Trichoderma* sp., *Aspergillus fumigatus, Mucor* sp., *Aspergillus flavus, Penicillium glandicola, Aspergillus glaucus, Fusarium solani, Chrysosporium* sp., and *Curvularia* sp., were used for antifungal studies. These fungal isolates were previously isolated from sorghum grains collected from stores and markets across the four zones of the Niger Republic, as part of a separate study.

2.7 Antifungal activity of ethanolic leaf extracts of *Parkia biglobosa* and *Eucalyptus camaldulensis*

The agar well diffusion method was used to assess the resultant extracts' antifungal efficacy. Normal saline was used to prepare fungal isolate suspensions until the turbidity reached the 2.0 McFarland standard (Debalke et al., 2018; Gaziano et al., 2018). Using a sterile cork borer, 8mm diameter aseptic agar wells were made. Potato dextrose agar plates were consistently swabbed with the standardised test organisms. Following the introduction of each extract solution (50, 75, and 100 mg/mL) into each well, the plates were incubated for 24 hours at 27°C (Selvamohan et al., 2012). The diameter of the inhibitory zone surrounding each well served as a proxy for the antifungal activity (Mighri et al., 2010).

3.0 RESULTS

3.1 Qualitative assessment of phytochemical components

A qualitative phytochemical examination of the ethanolic leaf extracts of Eucalyptus camaldulensis and Parkia biglobosa showed the presence of several metabolites. Phenols, saponins, tannins, and carbohydrates were substantially present in the Eucalyptus camaldulensis leaf extract, according to research. Glycosides, flavonoids, and steroids were not found, however terpenoids, alkaloids, and reducing sugars were found in smaller concentrations. Glycosides, flavonoids, and phenols were, however, significantly present in the Parkia biglobosa extract. There was a decreased concentration of terpenoids, saponins, steroids, carbs, and reducing sugars; alkaloids and tannins were absent. Parkia biglobosa and Eucalyptus camaldulensis ethanolic leaf extracts include the compounds that have been qualitatively characterised (Table 1).

Table 1: Qualitative analysis of the phytochemical components in leaf extracts from Eucalyptus camaldulensis and Parkia biglobosa

Constituents	Observation	
	Eucalyptus camaldulensis	***Parkia biglobosa***
Phenols	+++	++
Flavonoids	-	++
Terpenoids	+	+
Alkaloids	+	-
Saponins	++	+
Tannins	++	-
Steroids	-	+
Glycosides	-	++
Carbohydrate	++	+
Reducing Sugar	+	+

Keys; +++=Very high; ++=moderate; + = present; - = absent

3.2 Quantitative determination of phytochemical constituents

Table 2 shows the quantitative analysis of phytochemicals in *Parkia biglobosa* and *Eucalyptus camaldulensis* ethanolic leaf extracts. The number of phytochemicals in each extract varies significantly, according to the results, with *Eucalyptus camaldulensis* containing higher amounts of Phenols (21.32mg/g), Alkaloids (7.29mg/g), Saponins (13.71mg/g), Tannins (12.88mg/g), Carbohydrates (18.87mg/g), and Reducing Sugar (8.77mg/g). In contrast, *Parkia biglobosa* extracts contained higher amounts of Flavonoids (13.01mg/g), Terpenoids (8.61mg/g), and Glycosides (5.92mg/g) while Steroids were not detected in either of the extract.

Table 2: Quantitative determination of the phytochemical constituent of *Eucalyptus camaldulensis* and Parkia *biglobosa* leaf extracts

Phytochemical (mg/g)	***Eucalyptus camaldulensis***	***Parkia biglobosa***	P value

Phenols	21.32±0.00[b]	12.92±0.01[a]	0.00
Flavonoids	0.00±0.00[a]	13.01±0.00[b]	0.00
Terpenoids	6.89±0.00[a]	8.61±0.00[b]	0.00
Alkaloids	7.29±0.01[b]	0.00±0.00[a]	0.00
Saponins	13.71±0.01[b]	6.66±0.00[a]	0.00
Tannins	12.88±0.00[b]	5.97±0.01[a]	0.00
Steroids	0.00±0.00[a]	0.00±0.00[a]	0.00
Glycosides	0.00±0.00[a]	5.92±0.00[b]	0.00
Carbohydrates	18.87±0.01[b]	5.96±0.00[a]	0.00
Reducing sugar	8.77±0.01[b]	6.30±0.00[a]	0.00

The values are shown as mean ± standard error of mean (SEM). At $p < 0.05$, values exhibiting distinct superscripts within a row indicate significant differences.

3.3 GC-MS Profiling of Ethanolic Leaf Extracts of *Eucalyptus camaldulensis* and *Parkia biglobosa*

Eucalyptus camaldulensis and Parkia biglobosa were found to contain 32 and 27 chemicals, respectively, according to GC-MS analysis of the ethanolic leaf extracts. Tables 3 and 4 list the chemical components in the extracts together with their retention times (RT), molecular formulas, molecular weights (MW), and concentrations (%). Several bioactive compounds were found in both extracts, according to the analysis: acetic acid, hexanoic acid, benzoene, (ethenyloxy)-, ethanol, 2-(2-ethoxyethoxy)-, eugenol, 2-Propenoic acid, 3-phenyl-, methyl ester, (E)-, 4-(3-Hydroxybutyl)phenol, naphthalene, decahydro-1,5-dimethyl-, naphthalene, 1,2,3,4,4a,5,6,8a-octahydro-4a,8-dimethyl-2-(1-methylethenyl)-, [2R-(2α,4aα,8aβ)]1,2-Dehydroviridiflorol, Diethyl Phthalate, n-Hexadecanoic acid, 9,12-Octadecadienoic acid (Z,Z)-, 1-E-11,Z-13-Octadecatriene, 2-Pentadecanol, 1,8,11-Heptadecatriene, (Z,Z)-, cis,cis,cis-7,10,13-Hexadecatrienal, methyl ester of pentadecanoic acid, ethyl ester of hexadecanoic acid, methyl

ester of 7,10-octadecadienoic acid, 9, 12, and 15-Octadecatrienoic acid, methyl ester, ethyl oleate, (Z, Z, Z)- ethyl ester of 9,12-octadecadienoic acid Phytol, 9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester, butyl octyl ester, 1,1-dimethoxy-, (Z)-, 1,2-Benzenedicarboxylic acid, Eucalyptus camaldulensis with its Squalene and Stigmasterol 2,3-Dihydro-4H-pyran, 2,3-Butanediol, Benzenetriol 1, 2, 3, 3-hydroxy-3-methyl-2-butanone, Acetate, cis-Z-α-Bisabolene epoxide, 1,methyl-2-(2-propenyl)-, 2-Naphthalenol, benzene, and 9,9-Dimethoxybicyclo[3.3.1]nona-2,4-dione, methyl, cis-9-Hexadecenal, 9-t-Butyltricyclo[4.2.1.1(2,5)], phenol, 2,6-bis(1,1-dimethylethyl)-, α-D-Glucopyranoside, and methyldecane-9,10-diol, methyl ester, 7-Hexadecenoic acid, (Z)-, n-Hexadecanoic acid, Phytol, Oleic Acid, n-Hexadecanoic Acid, Hexadecanoic Acid, ethyl ester, Octadecanoic Acid, 9-Heptadecanone, 9,12-Octadecadienoic acid (Z,Z)-, Vitamin E, (Z)-, 9-octadecenamide, Parkia biglobosa: 1,1,6-Trimethyl-3-methylene-2-(3,6,9,13-tetramethyl-6-ethenye-10,14-dimethylene-pentadec-4-enyl)cyclohexane and Bis(tridecyl) phthalate. The matching chromatograms are shown in Figures 1 and 2.

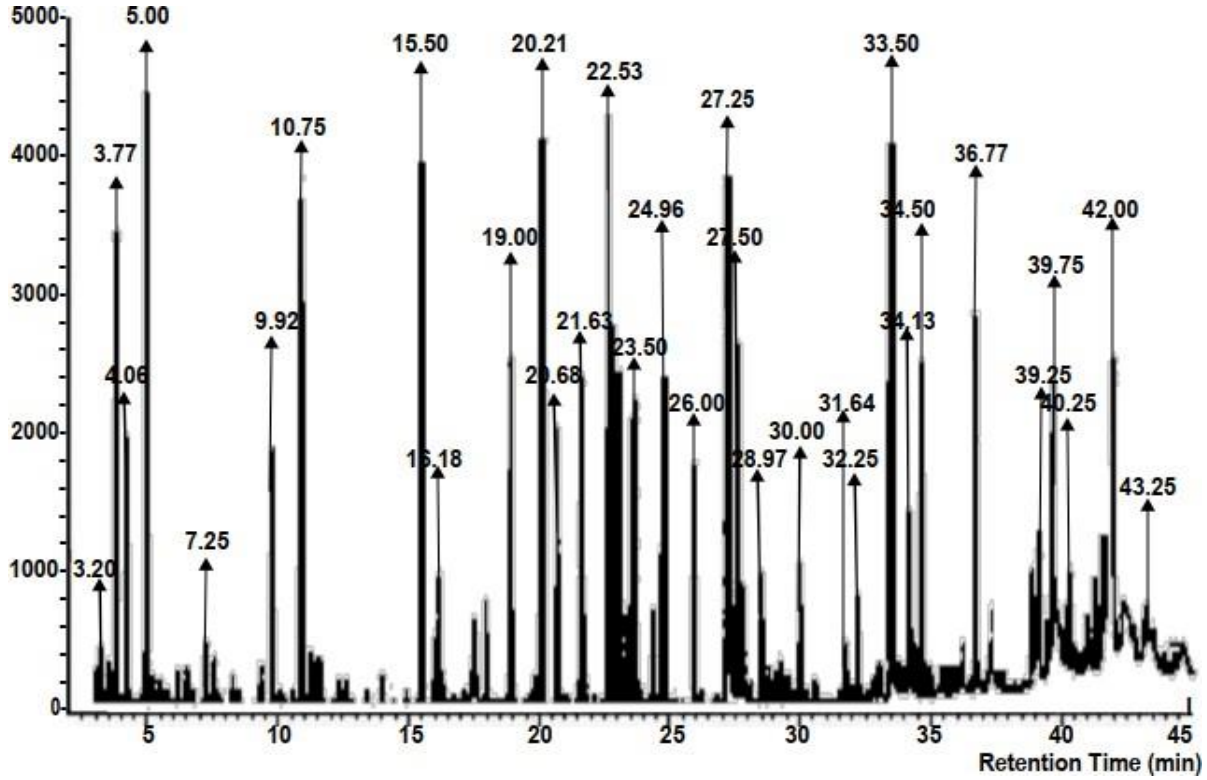


Figure 1: GC-MS chromatogram of ethanolic leaf extract of *Eucalyptus camaldulensis*

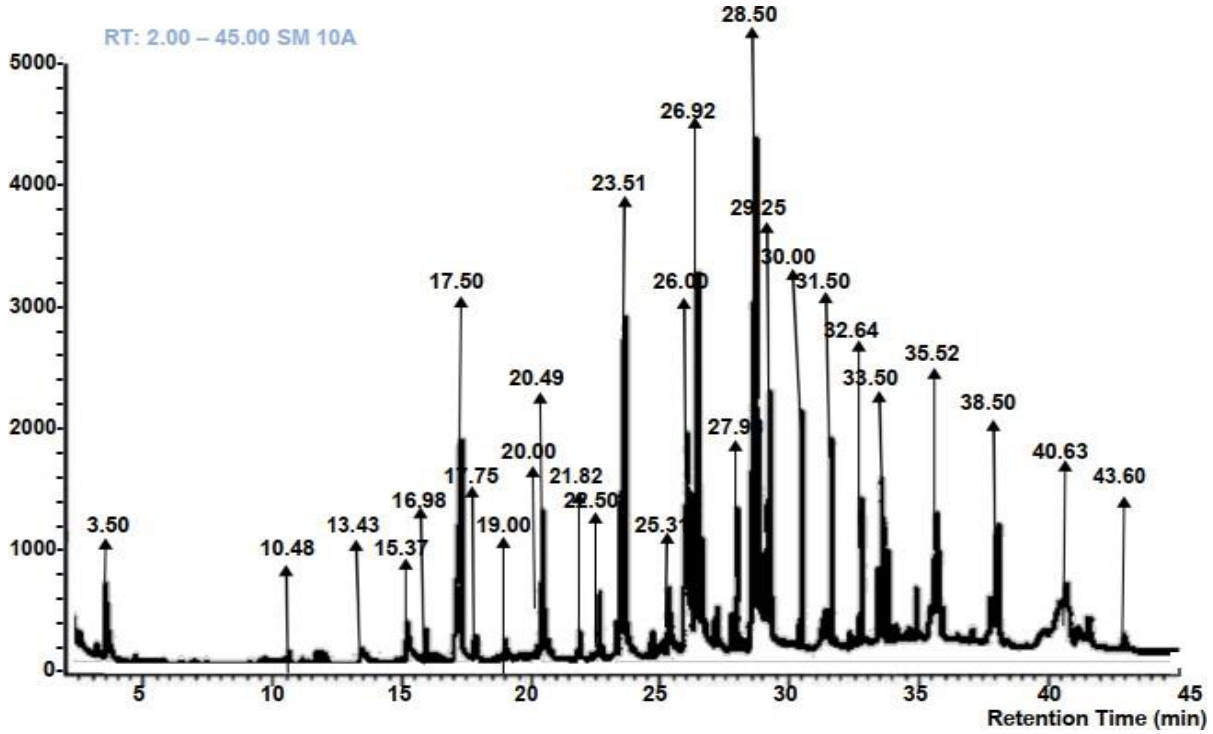
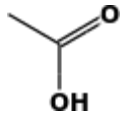
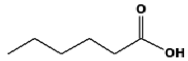
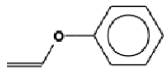
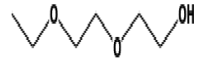
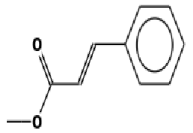
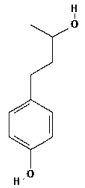
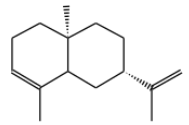
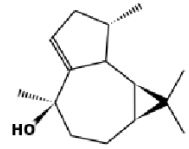
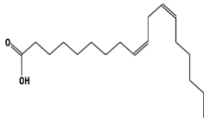
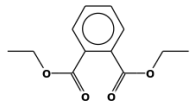
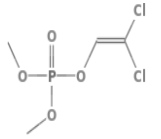
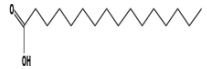
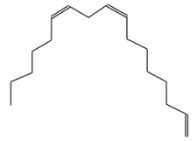
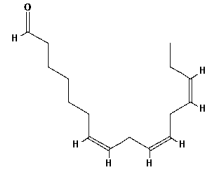
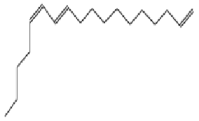


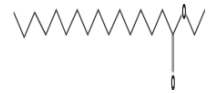
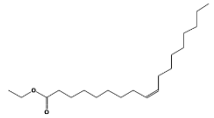
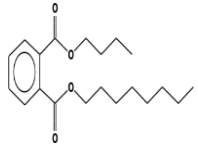
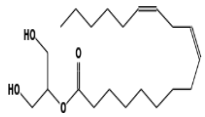
Figure 2: GC-MS chromatogram of ethanolic leaf extract of *Parkia biglobosa*

Table 3: Bioactive compounds present in the ethanolic leaf extract of *Eucalyptus camaldulensis*

Peak	RT	Compound Detected	Mol. Formula	MW	Peak Area (%)	Comp (wt%)	m/z	Structures
1	3.20	Acetic acid	$C_2H_4O_2$	60	0.72	1.31	41, 55, 140	
2	3.77	Hexanoic acid	$C_6H_{12}O_2$	116	4.91	5.37	43, 45, 60	
3	4.0	Benzene, (ethenyloxy)-	C_8H_8O	120	4.17	5.04	41, 60, 116	
4	5.00	Ethanol, 2-(2-ethoxyethoxy)-	$C_6H_{14}O_3$	134	6.49	7.31	51, 91, 120	
5	7.25	Eugenol	$C_{10}H_{12}O_2$	164	0.64	1.01	45, 59, 134	
6	9.92	2-Propenoic acid, 3-phenyl-, methyl ester, (E)-	$C_{10}H_{10}O_2$	162	1.15	1.43	51, 131, 162	
7	10.72	4-(3-Hydroxybutyl)phenol	$C_{10}H_{14}O_2$	166	5.18	5.35	43, 107, 166	

8	15.50	Naphthalene, decahydro-1,5-dimethyl-	$C_{12}H_{22}$	166	5.65	0.01	41, 95, 166	
9	16.18	Naphthalene, 1,2,3,4,4a,5,6,8a-octahydro-4a,8-dimethyl-2-(1-methylethenyl)-, [2R-(2α,4aα,8aβ)]-	$C_{15}H_{24}$	204	1.32	1.72	41, 93, 204	
10	19.00	1,2-Dehydroviridiflorol	$C_{15}H_{24}O$	220	3.94	1.97	41, 73, 220	
11	20.21	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	280	5.93	7.49	41, 57, 280	
12	20.68	Diethyl Phthalate	$C_{12}H_{14}O_4$	222	2.87	3.26	50, 140, 222	
13	21.63	Diethyl Phthalate	$C_{12}H_{14}O_4$	222	3.43	0.01	74, 109, 220	
14	22.53	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	5.44	7.15	43, 73, 256	

15	23.50	2-Pentadecanol	$C_{15}H_{32}O$	228	3.15	1.76	43, 45, 228	
16	24.96	1,8,11-Heptadecatriene, (Z,Z)-	$C_{17}H_{30}$	234	3.43	1.88	43, 93, 234	
17	26.00	Z,Z-2,13-Octadecadien-1-ol	$C_{18}H_{34}O$	266	4.03	2.83	41, 55, 266	
18	27.25	8-Heptadecenoic acid	$C_{17}H_{32}O_2$	268	5.47	5.68	41, 55, 268	
19	27.50	cis,cis,cis-7,10,13-Hexadecatrienal	$C_{16}H_{26}O$	234	3.73	1.47	41, 79, 234	
20	28.97	1,E-11,Z-13-Octadecatriene	$C_{18}H_{32}$	248	1.48	1.53	41, 67, 248	
21	30.00	Pentadecanoic acid, methyl ester	$C_{16}H_{32}O_2$	256	0.90	1.35	43, 74, 256	
22	31.64	7,10-Octadecadienoic acid, methyl ester	$C_{19}H_{34}O_2$	294	0.57	1.02	41, 67, 294	

23	32.25	Hexadecanoic acid, ethyl ester	$C_{18}H_{36}O_2$	284	0.84	2.42	43, 88, 284	
24	35.50	9,12,15-Octadecatrienoic acid, methyl ester, (Z,Z,Z)-	$C_{19}H_{32}O_2$	292	5.33	6.39	41, 79, 292	
25	34.13	Ethyl Oleate	$C_{20}H_{38}O_2$	310	1.81	1.42	43, 55, 310	
26	34.50	9,12-Octadecadienoic acid, ethyl ester	$C_{20}H_{36}O_2$	308	3.44	4.46	41, 67, 308	
27	36.77	Phytol	$C_{20}H_{40}O$	296	4.01	5.21	43, 71, 296	
28	39.25	9-Octadecene, 1,1-dimethoxy-, (Z)-	$C_{20}H_{40}O_2$	312	1.67	2.13	41, 71, 312	
29	39.75	1,2-Benzenedicarboxylic acid, butyl octyl ester	$C_{20}H_{30}O_4$	334	2.84	2.97	41, 149, 334	
30	40.25	9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester	$C_{21}H_{38}O_4$	354	1.06	1.68	41, 65, 354	
31	42.00	Squalene	$C_{30}H_{50}$	410	3.62	4.80	41, 69, 410	

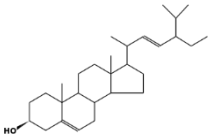
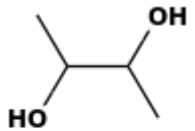
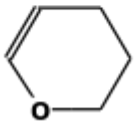
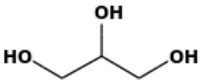
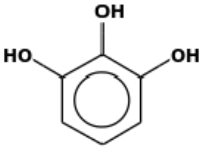
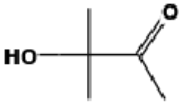
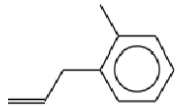
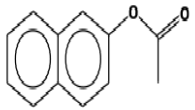
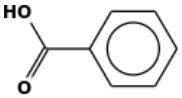
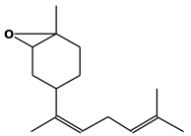
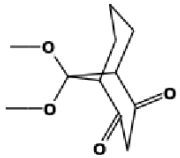
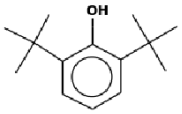
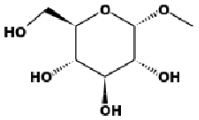
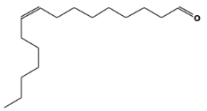
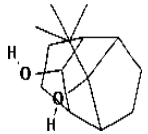
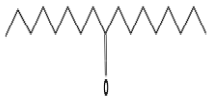
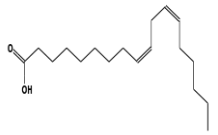
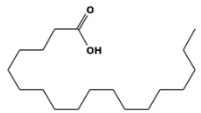
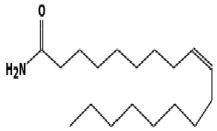
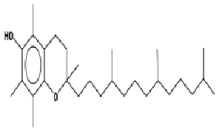
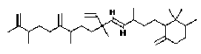
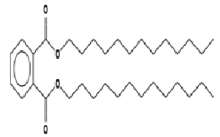
32	43.25	Stigmasterol	$C_{29}H_{48}O$	412	0.77	2.57	43, 55, 412	

Table 4: Bioactive compounds present in the ethanolic leaf extract of *Parkia biglobosa*

Peak	RT	Compound Detected	Mol. Formula	MW	Peak Area (%)	Comp (wt%)	m/z	Structures
1	3.50	2,3-Butanediol	$C_4H_{10}O_2$	90	2.09	2.65	45, 57, 90	
2	10.48	2,3-Dihydro-4H-pyran	C_5H_8O	84	0.29	0.54	41, 55, 84	
3	13.43	Glycerin	$C_3H_8O_3$	92	0.31	1.61	43, 61, 92	
4	15.37	1,2,3-Benzenetriol	$C_6H_6O_3$	126	1.13	1.75	52, 80, 126	
5	16.98	3-Hydroxy-3-methyl-2-butanone	$C_5H_{10}O_2$	102	1.07	1.46	43, 59, 102	

6	17.50	Benzene, 1-methyl-2-(2-propenyl)-	$C_{10}H_{12}$	132	5.69	3.15	51, 117, 132	
7	17.75	2-Naphthalenol, acetate	$C_{12}H_{10}O_2$	186	0.62	1.55	43, 115, 186	
8	19.00	Benzoic acid	$C_7H_6O_2$	122	0.55	1.31	51, 105, 122	
9	20.00	cis-Z-α-Bisabolene epoxide	$C_{15}H_{24}O$	220	1.76	1.79	41, 43, 220	
10	20.49	9,9-Dimethoxybicyclo[3.3.1]nona-2,4-dione	$C_{11}H_{16}O_4$	212	3.57	3.21	43, 81, 212	
11	21.82	Phenol, 2,6-bis(1,1-dimethylethyl)-	$C_{14}H_{22}O$	206	0.89	1.73	41, 57, 206	
12	22.50	α-D-Glucopyranoside, methyl	$C_7H_{14}O_6$	194	1.90	1.94	43, 60, 194	
13	23.51	cis-9-Hexadecenal	$C_{16}H_{30}O$	238	8.74	7.11	41, 55, 238	

14	25.31	9-t-Butyltricyclo[4.2.1.1(2,5)]decane-9,10-diol	$C_{14}H_{24}O_2$	224	1.76	0.51	67, 149,224	
15	26.00	7-Hexadecenoic acid, methyl ester, (Z)-	$C_{17}H_{32}O_2$	268	5.64	4.98	43, 73, 268	
16	26.92	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	9.66	9.83	43, 73, 256	
17	27.98	9-Heptadecanone	$C_{17}H_{34}O$	254	3.72	3.10	43, 71, 254	
18	28.50	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	280	13.05	14.63	41, 81, 280	
19	29.25	Phytol	$C_{20}H_{40}O$	296	5.77	5.83	43, 71, 296	
20	30.00	Oleic Acid	$C_{18}H_{34}O_2$	282	5.54	4.98	41, 55, 282	
21	31.50	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	4.45	4.73	43, 73, 256	

22	32.64	Hexadecanoic acid, ethyl ester	$C_{18}H_{36}O_2$	284	3.18	3.26	88, 101, 284	
23	33.50	Octadecanoic acid	$C_{18}H_{36}O_2$	284	8.46	9.33	43, 73, 284	
24	35.52	9-Octadecenamide, (Z)-	$C_{18}H_{35}NO$	281	4.65	2.69	41, 59, 281	
25	38.50	Vitamin E	$C_{29}H_{50}O_2$	430	2.58	3.02	43, 165, 430	
26	40.63	1,1,6-Trimethyl-3-methylene-2-(3,6,9,13-tetramethyl-6-ethenye-10,14-dimethylene-pentadec-4-enyl)cyclohexane	$C_{33}H_{56}$	452	1.96	1.71	55, 95, 452	
27	43.60	Bis(tridecyl) phthalate	$C_{34}H_{58}O_4$	530	0.96	1.59	43, 149, 530	

3.4 Antifungal Activity of *Parkia biglobosa* and *Eucalyptus camaldulensis* Ethanolic Leaf Extracts

Parkia biglobosa and Eucalyptus camaldulensis both have antifungal action, as seen in Table 5. Because the inhibitory zones grow as the extract concentration increases, the antifungal activity was concentration-dependent which *Eucalyptus camaldulensis* exhibited higher antifungal activity against *Penicillium glandicola* (13 mm), *Fusarium solani* (16 mm), *Aspergillus glaucus* (13 mm), *Curvularia* sp. (10 mm) and *Aspergillus niger* (10 mm) at 100mg/ml concentration. While showing weaker antifungal activity compared to *Eucalyptus camaldulensis*, *Parkia biglobosa* extract did exhibit some inhibition against some fungal species. *Parkia biglobosa* showed antifungal activity against *Penicillium glandicola* (14 mm), *Aspergillus glaucus* (15 mm), and *Aspergillus flavus* (8 mm) at the 100 mg/ml concentration. Neither extract showed antifungal activity against *Trichoderma* sp. and Mucor sp..

Table 5: Antifungal Activity of *Eucalyptus camaldulensis* and *Parkia biglobosa* ethanolic leaf extracts

Isolate	Conc (mg/ml)	Zones of inhibition (mm)		
		Eucalyptus camaldulensis	***Parkia biglobosa***	**Control (Nystatin 1000UI/ml)**
Aspergillus niger	50	8	-	15
	75	10	-	
	100	10	8	
Trichoderma sp.	50	-	-	11
	75	-	-	
	100	-	-	
Aspergillus fumigatus	50	-	-	15
	75	8	8	
	100	8	10	
Mucor sp	50	-	-	12
	75	-	-	
	100	-	-	
Aspergillus flavus	50	-	-	10

	75	-	8	
	100	8	8	
	50	10	9	
Penicillium glandicola	75	13	11	17
	100	13	14	
	50	8	10	
Aspergillus glaucus	75	10	14	19
	100	10	15	
	50	11	-	
Fusarium solani	75	13	-	21
	100	16	-	
	50	-	-	
Chrysosporium sp.	75	-	7	10
	100	-	8	
	50	7	-	
Curvularia sp.	75	10	8	15
	100	10	8	

4.0 DISCUSSION

In Africa, fungal infestations in vital crops result in significant economic losses annually, threatening food security and the livelihoods of millions of people (Kumar and Kalita, 2017; Udomkun et al., 2017; Gbashi et al., 2018). During the past few decades, research has switched towards organic control approaches due to growing concerns about the detrimental side effects of chemical fungicides. The potential of Eucalyptus camaldulensis and Parkia biglobosa leaf extracts as antifungal agents against sorghum fungal contamination was assessed in the current study. Their potential richness in bioactive compounds was shown by the qualitative phytochemical examination of leaf extracts from Eucalyptus camaldulensis and Parkia biglobosa. This analysis identified a variety of secondary metabolites that have been reported to have antioxidant, antimicrobial, and other actions. The results of the examination showed the

existence of several metabolites, such as alkaloids, phenols, sugars that reduce, flavonoids, tannins, carbohydrates, and glycosides. Though flavonoids, steroids, and glycosides were not detected in *Eucalyptus camaldulensis* extracts while alkaloids and tannins were absent in *Parkia biglobosa*. Quantitative analysis revealed significant difference in the quantity of phytochemicals between the extracts, with *Eucalyptus camaldulensis* containing higher amounts of Phenols (21.32mg/g), Alkaloids (7.29mg/g), Saponins (13.71mg/g), Tannins (12.88mg/g), Carbohydrates (18.87mg/g), and Reducing Sugar (8.77mg/g) while *Parkia biglobosa* extracts contained higher amounts of Flavonoids (13.01mg/g), Terpenoids (8.61mg/g), and Glycosides (5.92mg/g and Steroids were not detected in the extracts.

The identified phytochemicals, including phenols, saponins, tannins, and terpenoids, are known to possess antifungal properties (Rao et al., 2010; Upadhyay, 2011; Rohini and Rajesh, 2020; Simonetti et al., 2020; Okla et al., 2021). Rao et al. (2010) noted that these phytochemicals responsible for a plant's defence against insect infestations and diseases often exhibit potent antifungal activity against a wide range of fungal pathogens. This aligns with findings from various other studies. Sani et al. (2014) identified tannins, saponins, alkaloids, and steroids in *Eucalyptus camaldulensis* leaf extracts, while flavonoids, glycosides, and terpenoids were absent. Conversely, Komolafe and Oyelade (2015) reported the presence of saponins, tannins, glycosides, and flavonoids in *Parkia biglobosa* leaf extracts, but alkaloids were not detected. The presence of these secondary metabolites suggests that both plant species possess antifungal and other numerous properties.

The GC-MS analysis identified numerous compounds in the extracts, with 32 detected in *Eucalyptus camaldulensis* and 27 in *Parkia biglobosa*. In *Eucalyptus camaldulensis* the most notable compounds were eugenol, 4-(3-hydroxybutyl)phenol, 1,2-dehydroviridiflorol, phytol, squalene and stigmasterol. Eugenol and phytol have been reported to exhibits a wide range of bioactivities including antifungal and antimicrobial activities (Pejin et al., 2014; Ghaneian et al., 2015; Sakthivel et al., 2018; Lima et al., 2020; Didehdar et al., 2022; Abidin et al., 2023).

Eugenol is an allylphenol, a type of phenolic compound where an allyl group is attached to a guaiacol molecule (Anjum et al., 2020). It falls within the broader category of allylbenzene compounds. Ulanowska and Olas (2021) report that the phenolic compound eugenol is a primary constituent of many essential oils. Various studies including Wang et al. (2010), Rana et al.

(2011), Abd-Elsalam and Khokhlov, (2015), Xie et al. (2015), Didehdar et al. (2022) and Abidin et al. (2023) have reported the eugenol have antifungal activity against a wide range of fungi species including *Candida* sp., *Botrytis cinerea, Trichophyton mentagrophytes* and *Microsporum canis*, *Zygosaccharomyces rouxii*, *Fusarium oxysporum.* Phytol is classified as a diterpene alcohol and is a natural product formed when chlorophyll is metabolized by plants (de Moraes et al., 2014; Olofsson et al., 2014). Like eugenol, phytol has also been shown to exhibit antifungal activity against various fungal strains. Studies by Passos et al. (2012), Pejin et al. (2014), Devi et al. (2015), and Ghaneian et al. (2015) demonstrate this potential. However, the precise mechanism by which phytol exerts its antifungal effect remains unclear (Lima et al., 2020). Our findings on the presence of eugenol in *Eucalyptus* leaf extracts align with previous reports by Ghaffar et al. (2015) and Sharma et al. (2022). Both studies identified eugenol, alongside other compounds, in essential oils extracted from *Eucalyptus* species.

The compounds 1,2,3-benzenetriol, 2-naphthalenol, and oleic acid were the most notable in *Parkia biglobosa* ethanolic leaf extract. 1,2,3-benzenetriol also known as Pyrogallol or pyrogallic acid is a hydroxylated organic compound in the phenol family (Shin et al., 2019), with proven antimicrobial action (Akshaya et al., 2021; Yao et al., 2021), Its mode of action is due to oxidised chemicals inhibiting enzymes (Lima et al., 2016). Several studies have reported the presence of similar compounds in *Parkia biglobosa*. Ibrahim et al. (2013) identified them in stem bark extracts, while Sangodare et al. (2017) found them in fruit hull extracts. While Rohini and Rajesh, (2020) and Fernandes et al. (2021) reported these compounds in leaf and seed extracts, from other *Parkia* species.

Analysis of *Eucalyptus camaldulensis* and *Parkia biglobosa* ethanolic leaf extracts revealed the presence of various secondary metabolites with potential antifungal activity. Subsequent antifungal studies demonstrated varying degrees of activity against different sorghum fungal contaminants. The extracts inhibited the growth of *Penicillium glandicola*, *Fusarium solani*, *Aspergillus glaucus*, *Curvularia* sp., *Aspergillus niger*, and *Aspergillus flavus*. This antifungal activity was concentration-dependent, with larger inhibition zones observed at higher extract concentrations. Notably, *Eucalyptus camaldulensis* extracts exhibited higher antifungal activity compared to *Parkia biglobosa* while neither extract inhibited the growth of *Trichoderma* sp. and *Mucor* sp.

The observed antifungal activity of these plant extracts against various sorghum fungal contaminants suggests their potential as viable, cost-effective and eco-friendly alternatives to chemical fungicides for sorghum grain protection. This result aligns with previous studies by Bukar et al. (2010), Gakuubi et al. (2017), Sabo and Knezevic (2019), Yahaya et al. (2019), Al-Hadid et al. (2022), and Entonu et al. (2023), which collectively demonstrated the antifungal effects of *Eucalyptus camaldulensis* and *Parkia biglobosa* extracts against various fungal species, albeit with some variations in the degree of inhibition and effective concentrations.

5.0 CONCLUSION

The study showed the potential of *Parkia biglobosa* and *Eucalyptus camaldulensis* leaf extracts as antifungal agents against sorghum fungal contaminants. The phytochemical screening and GC-MS analysis revealed presence of various bioactive compounds, which may contribute to the antifungal activity. Thus, suggesting that these plant extracts could be used as a cost-effective and eco-friendly alternative to chemical fungicides for controlling fungal growth in sorghum grains.

REFERENCES

Abd-Elsalam, K. A., & Khokhlov, A. R. (2015). Eugenol oil nanoemulsion: antifungal activity against Fusarium oxysporum f. sp. vasinfectum and phytotoxicity on cottonseeds. *Applied Nanoscience, 5*, 255-265. https://doi.org/10.1007/s13204-014-0398-y

Abidin, Z. Z., Salleh, N. M., Himratul-Aznita, W. H., Ahmad, S. F., Lim, G. S., Mohd, N. R., & Dziaruddin, N. (2023). Antifungal effects of eugenol on Candida albicans adherence to denture polymers. *PeerJ, 11*, e15750. https://doi.org/10.7717/peerj.15750

Abioye, E. O., Akinpelu, D. A., Aiyegoro, O. A., Adegboye, M. F., Oni, M. O., & Okoh, A. I. (2013). Preliminary phytochemical screening and antibacterial properties of crude stem bark extracts and fractions of Parkia biglobosa (Jacq.). *Molecules*, *18*(7), 8485-8499.

Adejumo, A. A., Azeez, I. O., Geply, J. J., & Oboite, F. O. (2013). Processing, utilization and challenges of African locust bean (Parkia biglobosa, Jacque benth) in Arigidi Akoko, Ondo State, Nigeria. *Journal of Agriculture and Social Research (JASR)*, *13*(1), 39-49.

Akshaya, S. B., Krishnamoorthy, A. S., Sangeetha, C., Nakkeeran, S., & Thiribhuvanamala, G. (2021). Investigation on antifungal metabolites of Chinese caterpillar fungus Ophiocordyceps sinensis (Berk.) against wilt causing pathogen, *Fusarium* spp. *Annals. Phytomed*, *10*(1), 195-201. https://doi.org/10.21276/ap.2021.10.1.20

Al-Hadid, K. J., Al-Karablieh, N., Abu-Irmaileh, B., Sharab, A., & Al-Jaafreh, A. M. (2022). Antibacterial, antifungal, antioxidant, and anti-proliferative effects of Eucalyptus camaldulensis

and Pistacia atlantica ethanol extracts. *Tropical Journal of Natural Product Research (TJNPR)*, *6*(2), 207–212. https://doi.org/10.26538/tjnpr/v6i2.7.

Anigboro, A. A., Avwioroko, O. J., & Cholu, C. O. (2020). Phytochemical constituents, antimalarial efficacy, and protective effect of Eucalyptus camaldulensis aqueous leaf extract in plasmodium berghei-infected mice. *Preventive nutrition and food science*, *25*(1), 58. https://doi.org/10.3746%2Fpnf.2020.25.1.58

Anjum, N. F., Purohit, M. N., Yogish Kumar, H., Ramya, K., Javid, S., Salahuddin, M. D., & Prashantha Kumar, B. R. (2020). Semisynthetic derivatives of eugenol and their biological properties: A fleeting look at the promising molecules. *Journal of Biologically Active Products from Nature*, *10*(5), 379-404. https://doi.org/10.1080/22311866.2020.1837674

Arinola, S. O., Oje, O. J., & Omowaye-Taiwo, O. A. (2019). Evaluation of physicochemical properties and phytochemical composition of African locust bean (Parkia biglobosa) pulp. *Applied Tropical Agriculture*, *24*(1), 64-69.

Association of Official Analytical Chemists, AOAC (2010). *Official methods of analysis. Association of Official Analytical Chemists,* 15th Edition Washington D.C., USA. 184pp

Bukar, A., Uba, A., & Oyeyi, T. I. (2010). Phytochemical analysis and antimicrobial activity of Parkia biglobosa (Jacq.) Benth. extracts against some food-borne microorganisms. *Advances in Environmental Biology*, 74-80.

Chhikara, N., Devi, H. R., Jaglan, S., Sharma, P., Gupta, P., & Panghal, A. (2018). Bioactive compounds, food applications and health benefits of Parkia speciosa (stinky beans): a review. *Agriculture & food security*, *7*(1), 1-9. https://doi.org/10.1186/s40066-018-0197-x

Dar, R. A., Shahnawaz, M., Ahanger, M. A., & Majid, I. (2023). Exploring the diverse bioactive compounds from medicinal plants: a review. *J. Phytopharm*, *12*, 189-195.

Das, B. K., Al-Amin, M. M., Russel, S. M., Kabir, S., Bhattacherjee, R., & Hannan, J. M. A. (2014). Phytochemical screening and evaluation of analgesic activity of Oroxylum indicum. *Indian journal of pharmaceutical sciences*, *76*(6), 571.

de Moraes, J., de Oliveira, R. N., Costa, J. P., Junior, A. L., de Sousa, D. P., Freitas, R. M., ... & Pinto, P. L. (2014). Phytol, a diterpene alcohol from chlorophyll, as a drug against neglected tropical disease Schistosomiasis mansoni. *PLoS neglected tropical diseases*, *8*(1), e2617. https://doi.org/10.1371/journal.pntd.0002617

Debalke, D., Birhan, M., Kinubeh, A., & Yayeh, M. (2018). Assessments of antibacterial effects of aqueous-ethanolic extracts of Sida rhombifolia's aerial part. *The Scientific World Journal*, *2018*. https://doi.org/10.1155/2018/8429809

Devi, O., Rao, K., Bidalia, A., Wangkheirakpam, R., & Singh, O. (2015). GC-MS analysis of phytocomponents and antifungal activities of Zanthoxylum acanthopodium DC. collected from Manipur, India. *European Journal of Medicinal Plants*, *10*(1), 1-9. https://doi.org/10.9734/ejmp/2015/19353

Didehdar, M., Chegini, Z., & Shariati, A. (2022). Eugenol: A novel therapeutic agent for the inhibition of Candida species infection. *Frontiers in Pharmacology*, *13*, 872127. https://doi.org/10.3389%2Ffphar.2022.872127

Ejikeme, C., Ezeonu, C. S., & Eboatu, A. N. (2014). Determination of Physical and Phytochemical Constituents of some Tropical Timbers Indigenous to niger delta area of nigeria. *European Scientific Journal*, *10*(18), 247-270.

Entonu, E. E., Ngene, A. C., Odu, C. E., Ogaji, A. O., Nguhemen, H. W., Ochiheche, E. G., ... & Egbere, J. O. (2023). Evaluation of antifungal effect of Parkia biglobosa and Vitellaria paradoxa against selected pathogenic fungi. *MYCOPATH*, *21*(1).

Ezeonu, C. S., & Ejikeme, C. M. (2016). Qualitative and quantitative determination of phytochemical contents of indigenous Nigerian softwoods. *New journal of Science*, *2016*(1), 5601327. https://doi.org/10.1155/2016/5601327

Fernandes, R. D. M. N., Rodrigues, M. A. M., Panontin, J. F., Alves, D. R., Morais, S. M., Soares, I. M., & Scapin, E. (2021). Chemical investigation, toxic potential and acetylcholinesterase inhibitory effect of *Parkia platycephala* leaf and seed extracts. *Journal of Medicinal Plants Research, 15*(9), 401-412. https://doi.org/10.5897/jmpr2021.7158

Gakuubi, M. M., Maina, A. W., & Wagacha, J. M. (2017). Antifungal activity of essential oil of Eucalyptus camaldulensis Dehnh. against selected *Fusarium* spp. *International journal of microbiology*, *2017*(1), 8761610. https://doi.org/10.1155/2017/8761610

Gaziano, R., Campione, E., Iacovelli, F., Marino, D., Pica, F., Di Francesco, P. & Bianchi, L. (2018). Antifungal activity of Cardiospermum halicacabum L. (Sapindaceae) against Trichophyton rubrum occurs through molecular interaction with fungal Hsp90. *Drug design, development and therapy*, 2185-2193. https://doi.org/10.2147/dddt.s155610

Gbashi, S., Madala, N. E., De Saeger, S., De Boevre, M., Adekoya, I., Adebo, O. A., & Njobeh, P. B. (2018). The socio-economic impact of mycotoxin contamination in Africa. *Mycotoxins-Impact and Management Strategies*.

Ghaffar, A., Yameen, M., Kiran, S., Kamal, S., Jalal, F., Munir, B., ... & Jabbar, A. (2015). Chemical composition and in-vitro evaluation of the antimicrobial and antioxidant activities of essential oils extracted from seven Eucalyptus species. *Molecules*, *20*(11), 20487-20498. https://doi.org/10.3390%2Fmolecules201119706

Ghalem, B. R., & Mohamed, B. (2014). Antibacterial activity of essential oil of north west Algerian Eucalyptus camaldulensis against Escherichia coli and Staphylococcus aureus. *Journal of Coastal Life Medicine, 2*(10), 799-804. https://doi.org/10.12980/jclm.2.201414b57

Ghaneian, M. T., Ehrampoush, M. H., Jebali, A., Hekmatimoghaddam, S., & Mahmoudi, M. (2015). Antimicrobial activity, toxicity and stability of phytol as a novel surface disinfectant. *Environmental Health Engineering and Management Journal*, *2*(1), 13-16.

Ghareeb, M. A., Habib, M. R., Mossalem, H. S., & Abdel-Aziz, M. S. (2018). Phytochemical analysis of Eucalyptus camaldulensis leaves extracts and testing its antimicrobial and schistosomicidal activities. *Bulletin of the National Research Centre*, *42*, 1-9. https://doi.org/10.1186/s42269-018-0017-2

Gupta, M., Thakur, S., Sharma, A., & Gupta, S. (2013). Qualitative and quantitative analysis of phytochemicals and pharmacological value of some dye yielding medicinal plants. *Orient Journal of Chemistry*, *29*(2), 475-81. https://doi.org/10.13005/ojc/290211

Ibrahim, M. A., Koorbanally, N. A., & Islam, M. S. (2013). In vitro anti-oxidative activities of the various parts of *Parkia biglobosa* and GC-MS analysis of extracts with high activity. *African Journal of Traditional, Complementary and Alternative Medicines*, *10*(5), 283-291. https://doi.org/10.4314/ajtcam.v10i5.10

Komolafe, K., & Oyelade, W. A. (2015). Phytochemical screening and in vitro antioxidant activity of *Parkia biglobosa* extract. *Journal of Biology, Agriculture and Healthcare*, *5*(9), 91-5. https://doi.org/10.7176/jbah/9-16-04

Komolafe, K., & Oyelade, W. A. (2015). Phytochemical screening and in vitro antioxidant activity of Parkia biglobosa extract. *Journal of Biology, Agriculture and Healthcare, 5*(9), 91-5.

Kumar, D., & Kalita, P. (2017). Reducing postharvest losses during storage of grain crops to strengthen food security in developing countries. *Foods*, *6*(1), 8. https://doi.org/10.3390/foods6010008

Lagogianni, C. S., & Tsitsigiannis, D. I. (2018). Effective chemical management for prevention of aflatoxins in maize. *Phytopathologia Mediterranea*, *57*(1), 186-198.

Lima, T. L., Souza, L. B., Tavares-Pessoa, L. C., Santos-Silva, A. M. D., Cavalcante, R. S., Araújo-Júnior, R. F. D., ... & Silva-Júnior, A. A. D. (2020). Phytol-loaded solid lipid nanoparticles as a novel anticandidal nanobiotechnological approach. *Pharmaceutics*, *12*(9), 871. https://doi.org/10.3390/pharmaceutics12090871

Lima, V. N., Oliveira, C. D., Santos, E. S., Morais, L. P., Tintino, S. R., Freitas, T. S., ... & Coutinho, H. D. (2016). Gallic acid, Caffeic acid and Pyrogallol. *Microbial pathogenesis.* doi:10.1016/j.micpath.2016.08.004.

Madhu, M., Sailaja, V., Satyadev, T. N., & Satyanarayana, M. V. (2016). Quantitative phytochemical analysis of selected medicinal plant species by using various organic solvents. *Journal of Pharmacognosy and Phytochemistry*, *5*(2), 25-29.

Malik, S. K. (2017). Qualitative and quantitative estimation of terpenoid contents in some important plants of Punjab, Pakistan. *Pakistan Journal of Science*, *69*(2). https://doi.org/10.57041/pjs.v69i2.364

Mighri, H., Hajlaoui, H., Akrout, A., Najjaa, H., & Neffati, M. (2010). Antimicrobial and antioxidant activities of Artemisia herba-alba essential oil cultivated in Tunisian arid zone. *Comptes Rendus Chimie*, *13*(3), 380-386. https://doi.org/10.1016/j.crci.2009.09.008

Mohapatra, D., Kumar, S., Kotwaliwale, N., & Singh, K. K. (2017). Critical factors responsible for fungi growth in stored food grains and non-Chemical approaches for their control. *Industrial Crops and Products*, *108*, 162-182.

Nesci, A., Passone, M. A., Barra, P., Girardi, N., García, D., & Etcheverry, M. (2016). Prevention of aflatoxin contamination in stored grains using chemical strategies. *Current Opinion in Food Science*, *11*, 56-60.

Okla, M. K., Alatar, A. A., Al-Amri, S. S., Soufan, W. H., Ahmad, A., & Abdel-Maksoud, M. A. (2021). Antibacterial and antifungal activity of the extracts of different parts of *Avicennia marina* (Forssk.) Vierh. *Plants*, *10*(2), 252. https://doi.org/10.3390/plants10020252

Olivia, N. U., Goodness, U. C., & Obinna, O. M. (2021). Phytochemical profiling and GC-MS analysis of aqueous methanol fraction of Hibiscus asper leaves. *Future Journal of Pharmaceutical Sciences*, *7*, 1-5. https://doi.org/10.1186/s43094-021-00208-4

Olofsson, P., Hultqvist, M., Hellgren, L. I., & Holmdahl, R. (2014). Phytol: A chlorophyll component with anti-inflammatory and metabolic properties. *Recent Advances in Redox Active Plant and Microbial Products: From Basic Chemistry to Widespread Applications in Medicine and Agriculture*, 345-359. https://doi.org/10.1007/978-94-017-8953-0_13

Ons, L., Bylemans, D., Thevissen, K., & Cammue, B. P. (2020). Combining biocontrol agents with chemical fungicides for integrated plant fungal disease control. *Microorganisms*, *8*(12), 1930.

Pacyga, K., Pacyga, P., Topola, E., Viscardi, S., & Duda-Madej, A. (2024). Bioactive Compounds from Plant Origin as Natural Antimicrobial Agents for the Treatment of Wound Infections. *International Journal of Molecular Sciences*, *25*(4), 2100.

Passos, J. L., Barbosa, L. C. A., Demuner, A. J., Alvarenga, E. S., da Silva, C. M., & Barreto, R. W. (2012). Chemical characterization of volatile compounds of Lantana camara L. and L. radula Sw. and their antifungal activity. *Molecules*, *17*(10), 11447-11455. https://doi.org/10.3390/molecules171011447

Pejin, B., Savic, A., Sokovic, M., Glamoclija, J., Ciric, A., Nikolic, M.,.....& Mojovic, M. (2014). Further in vitro evaluation of antiradical and antimicrobial activities of phytol. *Natural Product Research*, *28*(6), 372-376.

Rana, I. S., Rana, A. S., & Rajak, R. C. (2011). Evaluation of antifungal activity in essential oil of the Syzygium aromaticum (L.) by extraction, purification and analysis of its main component eugenol. *Brazilian Journal of Microbiology*, *42*, 1269-1277. https://doi.org/10.1590%2FS1517-83822011000400004

Rao, A., Zhang, Y., Muend, S., & Rao, R. (2010). Mechanism of antifungal activity of terpenoid phenols resembles calcium stress and inhibition of the TOR pathway. *Antimicrobial agents and chemotherapy*, *54*(12), 5062-5069. https://doi.org/10.1128/aac.01050-10

Rohini, C. K., & Rajesh, Y. C. (2020). A Review on Phytopharmacological Profile of Traditionally used medicinal plant *Parkia biglandulosa* (Mimosaceae). *Asian Journal of Pharmaceutical Research*, *10*(1), 34-38. https://doi.org/10.5958/2231-5691.2020.00008.8

Sabo, V. A., & Knezevic, P. (2019). Antimicrobial activity of Eucalyptus camaldulensis Dehn. plant extracts and essential oils: A review. *Industrial crops and products*, *132*, 413-429. https://doi.org/10.1016/j.indcrop.2019.02.051

Sakthivel, R., Malar, D. S., & Devi, K. P. (2018). Phytol shows anti-angiogenic activity and induces apoptosis in A549 cells by depolarizing the mitochondrial membrane potential. *Biomedicine & Pharmacotherapy*, *105*, 742-752. https://doi.org/10.1016/j.biopha.2018.06.035

Saleh, M. S., Jalil, J., Zainalabidin, S., Asmadi, A. Y., Mustafa, N. H., & Kamisah, Y. (2021). Genus Parkia: Phytochemical, medicinal uses, and pharmacological properties. *International Journal of Molecular Sciences*, *22*(2), 618.

Sangodare, R. S. A., Okibe, P. O., Mohammed, M., Jajere, M. U., Abubakar, A., Aribido, O. S., ... & Kolo, M. T. (2017). Chemical investigation of *Parkia Biglobosa* fruit hull using GS-MS. *American Journal of Research Communication*, *5*(2), 59-65.

Sani, I., Abdulhamid, A., & Bello, F. (2014). Eucalyptus camaldulensis: Phytochemical composition of ethanolic and aqueous extracts of the leaves, stem-bark, root, fruits and seeds. *Journal of scientific and innovative Research*, *3*(5), 523-526. http://dx.doi.org/10.31254/jsir.2014.3510

Saptarini, N. M., Herawati, I. E., & Permatasari, U. Y. (2016). Total flavonoids content in acidified extract of flowers and leaves of gardenia (Gardenia jasminoides Ellis). *Asian Journal of Pharmaceutical and Clinical Research*, *9*(1), 213-215.

Selvamohan, T., Ramadas, V., & Kishore, S. S. S. (2012). Antimicrobial activity of selected medicinal plants against some selected human pathogenic bacteria. *Advances in Applied Science Research*, *3*(5), 3374-3381.

Sharma, A. D., Agnish, S., & Inderjeet, K. (2022). Chemical composition analysis and biological activities of essential oil from Eucalyptus polybractea (L.), growing in plains of Punjab, Northern India, by two different hydro-distillation methods. *Notulae Scientia Biologicae*, *14*(1), 11191-11191. https://doi.org/10.15835/nsb14111191

Shin, M., Park, E., & Lee, H. (2019). Plant- inspired pyrogallol- containing functional materials. *Advanced functional materials*, *29*(43), 1903022. https://doi.org/10.1002/adfm.201903022

Simonetti, G., Brasili, E., & Pasqua, G. (2020). Antifungal activity of phenolic and polyphenolic compounds from different matrices of Vitis vinifera L. against human pathogens. *Molecules*, *25*(16), 3748. https://doi.org/10.3390%2Fmolecules25163748

Sut, S., Dall'Acqua, S., Sinan, K. I., Zengin, G., Uba, A. I., Etienne, O. K., & Mahomoodally, M. F. (2024). Bioactive agents from Parkia biglobosa (Jacq.) R. Br. ex G. Don bark extracts for health promotion and nutraceutical uses. *Journal of the Science of Food and Agriculture*, *104*(5), 2820-2831.

Udomkun, P., Wiredu, A. N., Nagle, M., Bandyopadhyay, R., Müller, J., & Vanlauwe, B. (2017). Mycotoxins in Sub-Saharan Africa: Present situation, socio-economic impact, awareness, and outlook. *Food Control*, *72*, 110-122. https://doi.org/10.1016/j.foodcont.2016.07.039

Ulanowska, M., & Olas, B. (2021). Biological properties and prospects for the application of eugenol—a review. *International journal of molecular sciences*, *22*(7), 3671. http://dx.doi.org/10.3390/ijms22073671

Upadhyay, R. K. (2011). Plant natural products: Their pharmaceutical potential against disease and drug resistant microbial pathogens. *Journal Pharmaceutical Research*, *4*(4), 1179-1185.

Wang, C., Zhang, J., Chen, H., Fan, Y., & Shi, Z. (2010). Antifungal activity of eugenol against Botrytis cinerea. *Tropical Plant Pathology*, *35*, 137-143. https://doi.org/10.1590/S1982-56762010000300001

Xie, Y., Yang, Z., Cao, D., Rong, F., Ding, H., & Zhang, D. (2015). Antitermitic and antifungal activities of eugenol and its congeners from the flower buds of Syzgium aromaticum (clove). *Industrial Crops and Products*, *77*, 780-786. https://doi.org/10.1016/j.indcrop.2015.09.044

Yahaya, U., Abubakar, S., & Salisu, A. (2019). Antifungal activity of Parkia biglobosa extract on pathogenic strain of Candida albicans. *Journal of Applied Sciences*, *19*(3), 235-240. https://doi.org/10.3923/jas.2019.235.240

Yaméogo, J., & Yanogo, I. P. (2023). Impacts And Perceptions Of Climate Variability On Shea (Vitellaria Paradoxa) And African Locust Bean (Parkia Biglobosa) In Burkina Faso. *The Geographical Bulletin*, *1*(64), 17-29.

Yao, D., Zhang, G., Chen, W., Chen, J., Li, Z., Zheng, X., ... & Hu, X. (2021). Pyrogallol and Fluconazole Interact Synergistically In Vitro against *Candida glabrata* through an Efflux-Associated Mechanism. *Antimicrobial Agents and Chemotherapy*, *65*(7), 10-1128. https://doi.org/10.1128/aac.00100-21