



# *Spondias mombin*: biosafety and GC–MS analysis of anti-viral compounds from crude leaf extracts

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## Abstract

Essential oils are combinations of naturally occurring phytochemicals that, alone or in synergy with other compounds, can therapeutically attenuate disease-causing viral infections such as SARS-CoV-2, Ebola, and Marburg viruses. This study aimed to investigate the biosafety of and identification of antiviral phytochemicals of *Spondias mombin* (Linn) leaf crude extracts by GC–MS analysis. GC–MS analysis showed that the compound concentrations were dependent on the polarity of extracting solvent SMH (34), SMDCM (36), SMEA (12), SME (50) and SMM (36). Toluene (15.13%) and di-isooctyl phthalate (14.21%) were identified as the basic constituents of SMH. In SMDCM, n-nonadecanol-1 (19.64%) and eicosane (13.93%) were the main compounds, while in SMEA it was ethanol, 2-butoxy-(83.29%). Both SME and SMM showed the presence of Tetradecyl trifluoroacetate (15.43%). Pentadecanoic acid (12.18%), Propane, 2,2-diethoxy- (33.83%) and o-Xylene (15.87%). The identified antiviral compounds in the crude extracts, were D-Limonene (1.33%), p-Cymene (1.31%), Thymol (0.50%) and Carvacrol (0.87%) in SMM and SMH extracts, with Phytol, acetate being a common constituent in all the essential oils, except SMEA. In vitro cytotoxicity studies of crude *S. mombin* leaf extracts were performed using the MTT method in three cell lines: MCF-7, A-549 and HEK-293, with IC<sub>50</sub> values between 15.91 and 178.5 µg/mL. Therefore, the results indicated that crude extracts from *S. mombin* leaves had low toxicity and could be used safely. Compared with hexane extracts (1), methanol extracts have more compounds with antiviral properties (3) and can be used as reusable therapeutic candidates, natural dietary supplements or in the fight against SARS-CoV-2, Ebola and Marburg viruses. This can be valuable in pharmaceutical preparations of drug candidates for the treatment of these viruses.

**Keywords** GC–MS · *Spondias mombin* · Plant extract · Essential Oils · Cytotoxicity

## Introduction

Essential oils are naturally occurring mixtures of phytochemicals that, alone or in synergy with other compounds, therapeutically attenuate disease-causing viral infections such as SARS-CoV-2, Ebola, and Marburg viruses (Obi-Egbedi et al. 2012; Aromolaran and Badejo 2014). Natural products such as plant extracts, either as pure compounds or as standardized extracts, offer unlimited opportunities for

new drug discovery due to the unparalleled availability of chemical diversity. According to the World Health Organization (WHO), more than 80% of the world's population relies on traditional medicine for their basic health needs (Santos Sampaio et al. 2018). Due to their natural properties, medicinal plants have been used to treat many diseases. Consequently, research has grown into the field of examining the potential properties and uses of terrestrial plant extracts to create potential nanomaterial-based drugs to treat diseases including cancer and viruses. Researchers have identified plant species with anticancer and antiviral properties, with a focus on plants used in herbal medicine in developing countries (Sabiou et al. 2015). However, there is a need to identify new drug candidates with chemotherapeutic, chemoprotective, and antiviral properties with little or no side effects on normal cells (Ishola et al. 2018).

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Despite the high availability of medicinal plant materials, relatively few have been thoroughly investigated, with reports on the detailed mechanisms of the adsorption process being limited (Obi-Egbedi et al. 2012).

*Spondias mombin* (S.M.), an essential oil-bearing plant, is a fructiferous tree that thrives in rainforests (Aromolaran and Badejo 2014) and coastal areas and is dispersed throughout the tropical regions of South America, Africa and Asia. *Spondias mombin* (*S. mombin*) has been used extensively in folk medicine and is known by various names (Aromolaran and Badejo 2014; Santos Sampaio et al. 2018). The *S. mombin* plant attains a height of 15–22 m and is commonly used to cure many diseases due to its potent bioactive principles, including tannins, saponins, flavonoids, phenolics and anthraquinone glycosides (Sabiu et al. 2015; Ishola et al. 2018).

This study aims to identify antiviral phytochemical compounds by using Gas Chromatography–Mass Spectrometry (GC–MS) in crude extracts of *S. mombin* (S.M.), serially and exhaustively extracted with increasing polarity.

The WHO has produced and licensed for use a variety of vaccinations as the number of infections rises (Li et al. 2021; Li and Lu 2020; Carvalho et al. 2021; Burgess et al. 2021; Mahase 2021). Notable vaccines (Connors et al. 2021) being provided in various regions of the world include; mRNA-1273 (Moderna) (World Health Organization 2021a), BNT162b2 (Pfizer/BioNTech) (World Health Organization 2021b; Liu et al. 2021), ChAdOx1 nCoV-19 (Astrazenac/Oxford) (Soto 2020; Voysey et al. 2021), (rAd26)/rAd5 (Sputnik V) (Soto 2020; Tulleken 2021; Jones and Roy 2021), and Janssen Ad26.COVS.2.S (Johnson and Johnson) (Douguih 2021). Few medicines such as Chloroquine, Hydroxychloroquine, Remdesivir (GS-5734), Favipiravir, Ivermectin and Lopinavir/Ritonavir have also been extensively investigated and repurposed for the treatment of SARS-CoV-2 based on their previously reported potential as antiviral therapeutics (Savarino et al. 2003; Dan et al. 2020; Colson et al. 2020; Barlow et al. 2020; Dong et al. 2020; Wang et al. 2020; Awadasseid et al. 2021; Tu et al. 2020). The Food and Drug Administration subsequently approved Remdesivir, Ronapreve (Mahase 2021) to treat COVID-19 in hospitalized adults and pediatric patients (Goldman et al. 2021; Beigel et al. 2020).

To further investigate in vitro cytotoxic properties of these crude leaf extracts of S.M. using, (3-[4,5-dimethylthiazol-2-yl]-2,5 diphenyl tetrazolium bromide) (MTT) assay in three different cell lines: liver hepatocellular carcinoma cell lines, breast cancer (MCF-7), adenocarcinoma human alveolar basal

epithelial cells (A-549) and human embryonic kidney cells (HEK-293).

## Materials and methods

### Collection of plant materials, reagents and instrumentations

The leaves of the medicinal plant *S. mombin* were collected from the Cape Coast (Coordinates: 5.114467, – 1.287032) Central Region of Ghana on 25/06/2019 at 11:30 am Central African Time with an average temperature of 25 °C, average humidity of 80% and an average pressure of 1009 bar. The plants were identified and authenticated at the Herbarium in the School of Life Sciences at the University of KwaZulu-Natal, Westville Campus. All chemicals and solvents used in this research work were of analytical reagent grade (A.R. grade) and purchased from Sigma Aldrich and Merck Millipore, South Africa. Hexane and ethyl acetate were purchased from Associated Chemical Enterprises, Dichloromethane, methanol, ethanol, 3-(4,5-Dimethyl-2-thiazolyl)-2,5-diphenyl-2H-tetrazolium bromide (MTT) salt Dulbecco's Modified Eagle Medium (DMEM), Eagle's Minimum Essential Medium (EMEM) and all other cell culture reagents, including Whatman filter paper were purchased from Sigma Aldrich, South Africa. Rotary evaporator and Soxhlet apparatus (Büchi-Germany) and Analytical balance (Lasec, South Africa) are some instrumentations used in the experiments.

### Methodology

Various experimental procedures in this research were carried out after collecting the materials, such as the extractions using five different solvents of increasing polarity, preliminary phytochemical screening, Gas Chromatography–Mass Spectrometry (GC–MS) and in vitro cytotoxicity assay.

### Study design

Preliminary phytochemical screening was carried out using the standard methods as reported (Dahiru et al. 2006; Kumar et al. 2007; Kwon et al. 2006; Parekh and Chanda 2007; Sengar et al. 2015; Wu et al. 2014; Iqbal et al. 2015). In brief, the leaves were air-dried at room temperature and pulverized to a fine powder. *S. mombin* leaf powder (20 g) was weighed using

an analytical balance and placed in the extractor of the Soxhlet apparatus with a total of 600 mL solvent measured into the distillation flask Serial exhaustive extraction method at a temperature of 70 °C, in triplicate, and involved successive extractions with bulk solvents of different polarities, namely hexane, dichloromethane, ethyl acetate, ethanol and methanol. The increasing solvent polarity was to ensure that a wide range of compounds with varying polarities was extracted (Nadembega et al. 2011). Each solvent extract was concentrated and dried using a rotary evaporator, and preserved in the refrigerator for future use in phytochemical screening and in vitro, isolation, purification and characterization (Rukshana et al. 2017). Interpretation of mass spectrum GC–MS was conducted using the National Institute Standard and Technology (NIST) database, having more than 62,000 patterns.

**Specifications:** Analysis by GC–MS using PerkinEmler Gas Chromatography (Clarus 580) equipped with MSD mass spectrometer (Clarus SQ8S) instrument with built-in autosampler. Column: Elite-5MS (30 m × 0.25 mm id × 0.25 µm). The oven temperature is programmed from 37 to 320 °C at a rate of 18–25 °C/min and held for 0.5 and 1.85 min at 18 and 320 °C, respectively. The injector temperature: 250 °C and MS Ion Source temperature: 280 °C with a full scan and solvent delay of 0–2.30 min. MS Scan Range was m/z 35–500 min 0.10 s. One microlitre of the samples was injected in He carrier gas at a 20 mL/min split flow.

The name of the compound, molecular weight (g/mol), molecular formula, retention time, percentage area (Area%), class of phytochemical, biological activity and reference (s) of the compounds were ascertained.

The MTT assay was performed using standard methods described by Hassan et al. (2019), with the crude extract (*S. mombin*) *in-vitro* cytotoxicity being evaluated in three different cell lines, the liver hepatocellular carcinoma cell lines-breast cancer (MCF-7), adenocarcinoma human alveolar basal epithelial cells (A-549) and human embryonic kidney cells (HEK-293) that were incubated. All three cells were sustained at 37 °C in a humidified atmosphere of 5% CO<sub>2</sub> in the air. The tested crude was dissolved in the cell culture media serving as a stock solution and diluted in the culture medium, resulting in 20, 40, 60, 80 and 100 µg/mL (Hassan et al. 2019). All three cell lines were transferred homogeneously ( $2.5 \times 10^3$ ) into a 96-well plate and incubated for 24 h to allow cell adherence. The ultimate treatment concentrations were reached by

refilling the 96-well plate with a fresh culture medium (100 µL per well) that was composed of suitable concentrations of the studied solution crude extract. The culture medium alone was used for the control, while the blank contained only the culture medium in the 96-well plate with no cells. After 48 h of incubation, the culture medium and the studied samples in each well were withdrawn and substituted with fresh culture medium and MTT solution (5 mg/mL in PBS pH 7.4). The media and MTT solution were withdrawn after 4 h of incubation and immediately thereafter 100 µL of dimethyl sulfoxide was combined with each 96-well to solubilize the MTT formazan. The optical density of samples in the 96-well was measured on a microplate spectrophotometer (Spectrostar Nano, Germany) at a wavelength of 540 nm (Hassan et al. 2020), and the IC<sub>50</sub> was calculated based on the previously reported method (Damiani et al. 2019). All experiments were completed in three replicates, the percentage (%) cell viability being calculated according to Eq. 1.

$$\% \text{ Cell Viability} = \frac{(\text{A540 nm treated cells})}{(\text{A540 nm untreated cells})} \times 100 \quad (1)$$

## Results and discussions

### Percentage yield and phytochemical screening

Table 1 presents the yield of *S. mombin* leaf extract obtained from hexane (SM-HEX), dichloromethane (SM-DCM), ethyl acetate (SM-EtOAc), ethanol (S.M.- EtOH) and methanol (S.M.- MeOH).

The result in Table 2 presents a preliminary phytochemical screening of *S. mombin* alcoholic crude leaf extracts. According to Nworu et al. (2007), the methanolic leaf extract of S.M. contained flavonoids, tannins, saponins, glycosides and triterpenes. Another author reported tannins, anthraquinones, flavonoids, cardiac glycosides and saponins being identified in SM-EtOH and SM-MeOH crude leaf extracts (Ayoka et al. 2005).

The experiments were done in triplicates and the preliminary phytochemical screening of the crude extract of *S.M* leaf indicated a positive test for anthraquinones derivatives, alkaloids, tannins, terpenoids, saponins and Cardiac glucoside, which are in line with reports by Ayoka et al.

**Table 1** The amount of extract per solvent of extraction from 20 g S.M. dry powder

	SM-HEX extract	SM-DCM extract	SM-EtOAc extract	SM-EtOH extract	SM-MeOH extract	Total
Yield	2.072 g	0.1493 g	0.1802 g	0.1740 g	0.1801 g	2.7556 g
% Yield	10.36%	0.75%	0.90%	0.87%	0.90%	13.78%

**Table 2** Preliminary phytochemical screening leaf extracts of *S. mombin*

Class of phytochemicals	Tests performed	<i>S. mombin</i> L. leaf extracts				
		<i>SM-HEX</i>	<i>SM-DCM</i>	<i>SM-EtOAc</i>	<i>SM-EtOH</i>	<i>SM-MeOH</i>
Alkaloids	Meyer	–	–	–	+	+
Anthraquinones derivatives	Borntrager's test	–	+	+	+	+
Steroids	Liebermann–Burchard test	+	+	+	–	–
Terpenoids	Liebermann–Burchard test	–	–	+	+	+
Saponins	Frothing	–	–	–	+	+
Flavonoids	Sulfuric acid test	+	–	–	–	–
Tannins	Ferric chloride test	+	+	+	+	+
Cardiac glucosides	Keller Killian	+	+	+	+	+

Legend: present (+), absent (–)

(2005) and Igwe et al. (2010). The absence of flavonoids and steroids was also reported by Igwe et al. (2010) and Maduka et al. (2014) in a study of anti-oxidant and microbial inhibitory effects of *S. mombin* leaf and stem bark extracts.

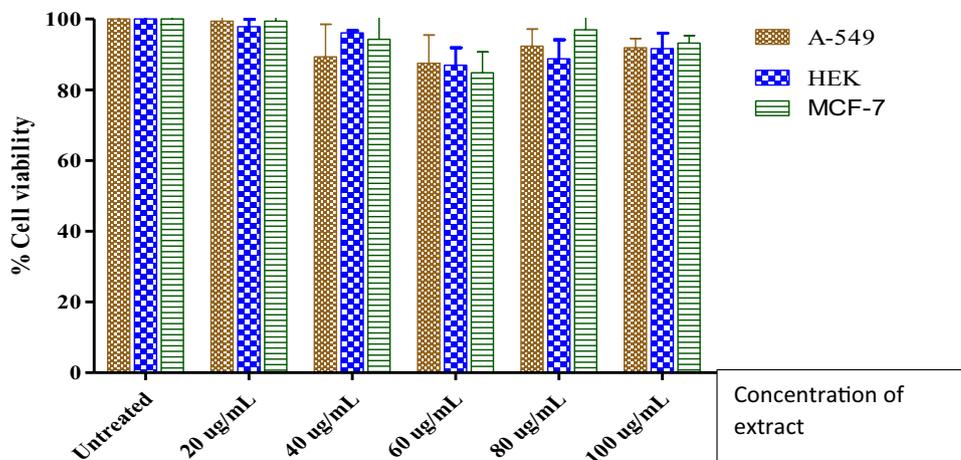
### In vitro cytotoxicity of *S. mombin* in the different solvents of extraction

Biosafety is an essential criterion to establish the nontoxic dosages of any compound for biomedical applications. An MTT assay-based cytotoxicity study was employed to evaluate and quantify the cytotoxicity of newly extracted plant crude based on a previously used plant-reported method (Sonawane et al. 2015). The in vitro cell viability of the crude in Hex, DCM, EtOAc, EtOH and MeOH was studied in three different cell lines: liver hepatocellular carcinoma cell lines- breast cancer (MCF-7), adenocarcinoma human alveolar basal epithelial cells (A-549) and human embryonic kidney cells (HEK-293).

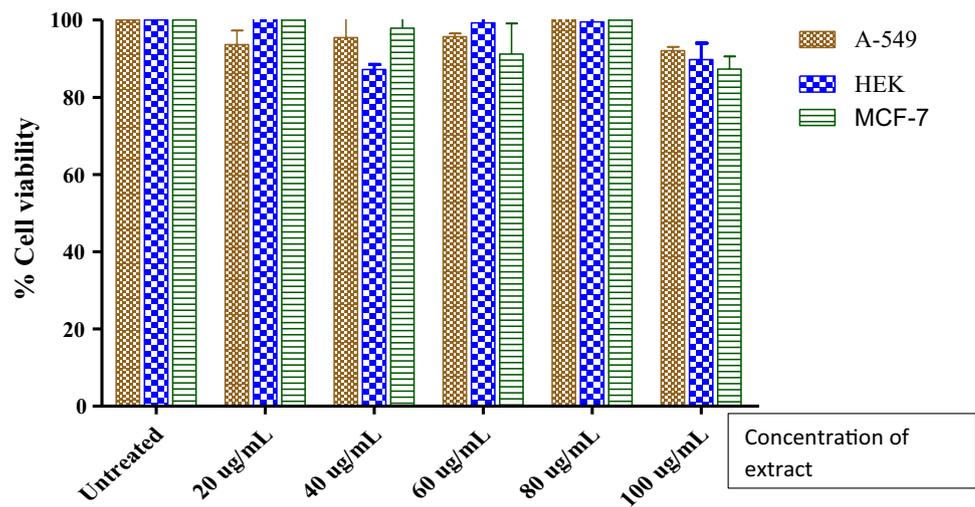
### In vitro cytotoxicity of *S. mombin* in SM-EtOAc

Generally, the MTT assay demonstrated that the crude plant extract was safe in all three cell lines, as shown in Fig. 1A. The crude extracts demonstrated cell viability from 81.89 to 100% across all concentration from 20 to 100  $\mu\text{g}/\text{mL}$  for A-549 cell line. In addition, the cell viability of the crude in HEK-293 was revealed to be from 78.43 to 100% viability in all crude extract concentrations, and for MCF-7, the cell viability was revealed to be from 73.47 to 100%. These results revealed that the crude leaf extract of SM-EtOAc shows that the cells were more viable in MCF-9 when compared to HEK-293 and A-549 cell lines. Therefore, these results suggested that cell viability was higher and more potent in MCF-7, and is greater than 75% viability, with ambiguous inhibitory concentration ( $\text{IC}_{50}$ ) ranging from 41.78, 60.67 and 59.71  $\mu\text{g}/\text{mL}$  in A-549, HEK-293 and MCF-7, which indicated that SM-EtOAc did not induce cytotoxicity on the cells.

**Fig. 1** Cell viability of S.M.-EtOAc leaf extract tested in three different cell lines: liver hepatocellular carcinoma cell lines—breast cancer (MCF-7), adenocarcinoma human alveolar basal epithelial cells (A-549) and human embryonic kidney cells (HEK-293)



**Fig. 2** Cell viability of SM-MeOH leaf extract tested in three cell lines: liver hepatocellular carcinoma cell lines—breast cancer (MCF-7), adenocarcinoma human alveolar basal epithelial cells (A-549) and human embryonic kidney cells (HEK-293)



### In vitro cytotoxicity of *S. mombin* in SM-MeOH

As indicated in Fig. 2, the cell viability of the *S. mombin* methanolic (SM-MeOH) crude leaf extract was shown to be viable, with greater than 75% cell viability across all concentrations (20–100  $\mu\text{g/mL}$ ). The cell viability was shown to be from 82 to 100% in A-549, 86–100% in HEK-293 and 78% to 100% in MCF-7. These results demonstrated that the *S. mombin* crude extracted using methanol was more viable in MCF-7 than in A-549 and HEK-293 (MCF-7 > A-549 > HEK-293). The inhibitory concentration ( $\text{IC}_{50}$ ) ranges from 36.42 to 15.91 and 178.5  $\mu\text{g/mL}$  in A-549, HEK and MCF-7 respectively, which indicated that SM-MeOH did not induce cytotoxicity on the cells.

### In vitro cytotoxicity of *S. mombin* in SM-EtOH

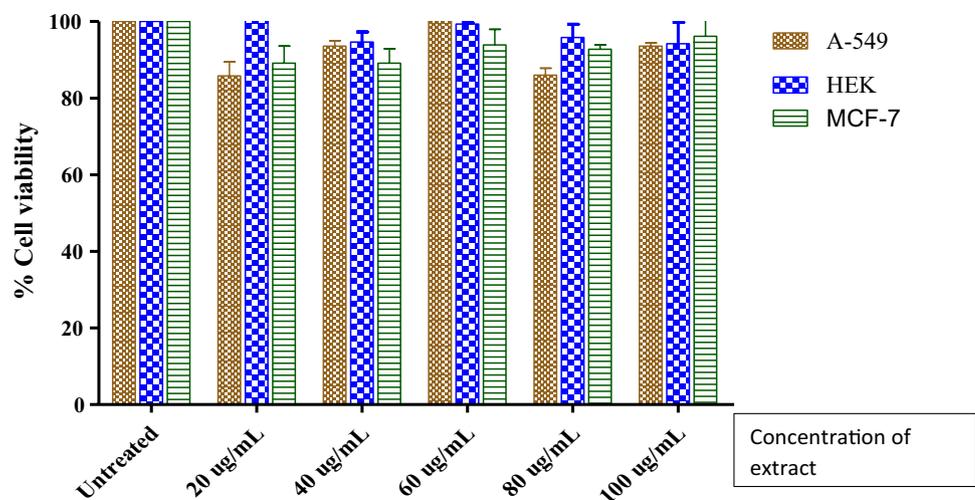
After exposure to the leaf extracted with ethanol (SM-EtOH), the viability of the cells was assessed by quantifying

crystalline blue formazan formation (Abo et al. 1999). Figure 3 presented the breakdown of the cytotoxicity assay of the plant extract against the A549, HEK 293 and MCF-7 cells. The results indicate a high percentage of cell viability, from 83 to 100% for A549 cells, 93–100% for HEK-293, and 82–100% for MCF-7 cells for all concentrations of nanoplexes tested. The results confirm the biosafety of the plant extract, with greater than 75% cell viability indicating nontoxicity of the material to mammalian cells (Sikwal et al. 2016). The inhibitory concentration ( $\text{IC}_{50}$ ) ranges from 19.59 to 15.81 and 51.55  $\mu\text{g/mL}$  in A-549, HEK and MCF-7 respectively, which indicated that SM-EtOH did not induce cytotoxicity on the cells.

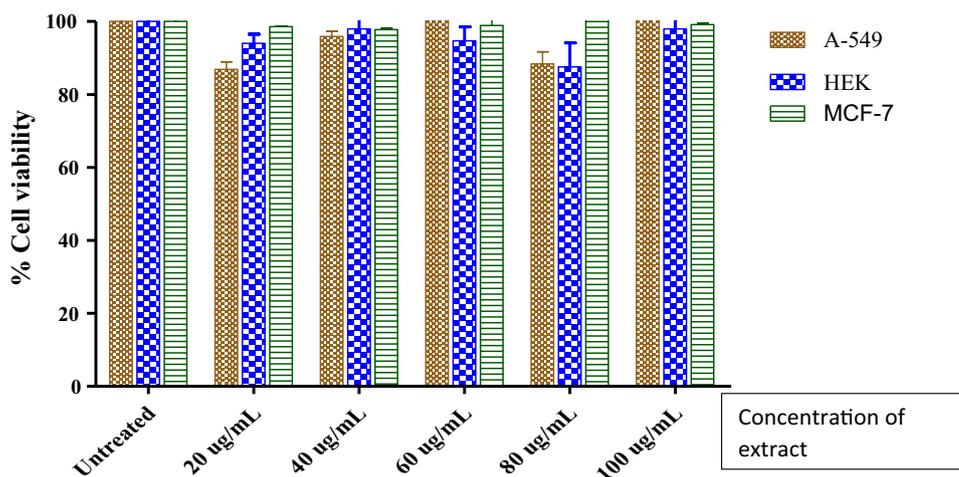
### In vitro cytotoxicity of *S. mombin* in SM-HEX

The viability of cells after exposure to the SM-HEX was assessed by quantifying crystalline blue formazan formation (Abo et al. 1999). Figure 4 shows the breakdown of

**Fig. 3** Cell viability of plant (crude) extract in SM-EtOH tested in three cell lines: liver hepatocellular carcinoma cell lines—breast cancer (MCF-7), adenocarcinoma human alveolar basal epithelial cells (A-549) and human embryonic kidney cells (HEK-293)



**Fig. 4** Cell viability of SM-HEX leaf extract (crude) is tested in three cell lines: liver hepatocellular carcinoma cell lines—breast cancer (MCF-7), adenocarcinoma human alveolar basal epithelial cells (A-549) and human embryonic kidney cells (HEK-293)

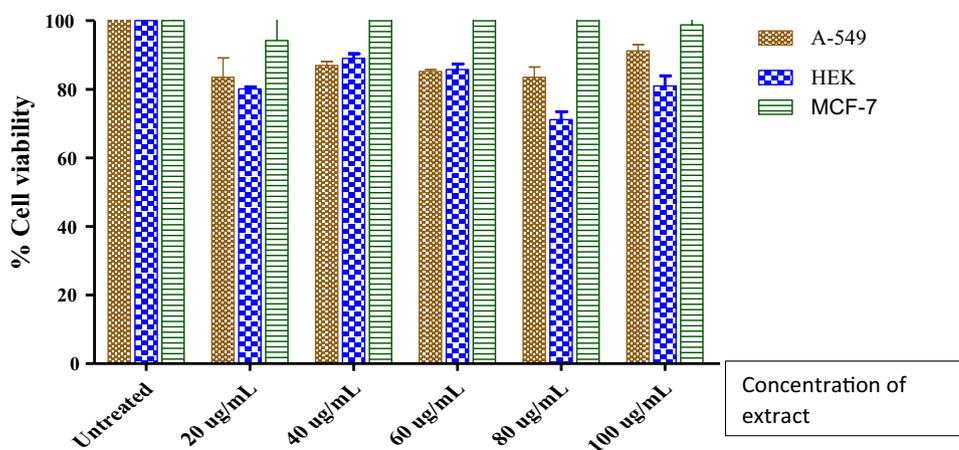


the cytotoxicity assay of the plant extract against the A549, HEK-293 and MCF-7 cells. The results indicate a high percentage of cell viability, from 84 to 100% for the A549 cells, 8%–100% for the HEK-293, and 91–100% for the MCF-7 cells for all concentrations of nanoplexes tested. The results confirm the biosafety of the plant extract, with greater than 75% cell viability indicating the nontoxicity of the material to all the cell lines (Sikwal et al. 2016). The inhibitory concentration ( $IC_{50}$ ) ranges from 38.55 to 35.56 and 60.52  $\mu\text{g}/\text{mL}$  in A-549, HEK and MCF-7 respectively, which indicated that SM-HEX did not induce cytotoxicity on the cells.

#### In vitro cytotoxicity of *S. mombin* in DCM

Figure 5 shows the breakdown of the cytotoxicity assay of SM-DCM leaf extract against the A549, HEK 293 and MCF-7 cells. The results indicate a high percentage of cell viability, from 78 to 94% for A549 cells, 68–89% for HEK-293, and 86–100% for MCF-7 (MCF-7 > A-549 > HEK-293) for all concentrations of plant extract tested. The

**Fig. 5** Cell viability of *S.M.*-DCM crude extract tested in three cell lines: liver hepatocellular carcinoma cell lines—breast cancer (MCF-7), adenocarcinoma human alveolar basal epithelial cells (A-549) and human embryonic kidney cells (HEK-293)



inhibitory concentration ( $IC_{50}$ ) ranges from 113.2 to 56.48 and 16.39  $\mu\text{g}/\text{mL}$  in A-549, HEK and MCF-7 respectively, which indicated that DCM did not induce cytotoxicity on the cells. The results confirm the biosafety of the plant extract, indicating the nontoxicity of the material to the mammalian cells (Sikwal et al. 2016). Based on these results, the plant extract can therefore be considered nontoxic and safe for cancer treatment.

#### GC–MS analysis of *S. mombin* L. crude leaf extracts

The suitability of the process for extracting phytochemicals from medicinal plants is an important step in determining the class of phytochemicals and their biological activities. The target compound can exist in different extraction solvents, ranging from polar to non-polar solvents. However, the choice of extraction solvent mainly depends on toxicity and availability (Zandoná et al. 2020). Therefore, the choice of five solvents with increasing polarity was made to be able to extract as many as possible phytochemicals that may possess antiviral properties.

The GC–MS analysis of *S. mombin* crude leaf extracts revealed various phytoactive compounds as shown in the “Appendix” (Tables 3, 4, 5, 6, 7 and Figs. 6, 7, 8, 9, 10, 11, 12). Some of the common compounds found in all the extracts are Phytol, phytol acetate, Diisooctyl phthalate, Eicosane, Tetradecane, Ascorbic acid and Phthalic acid. Toluene (15.13%) and Diisooctyl phthalate (14.21%) were identified as the significant components of SMH. However a monoterpene, occurring in the Hexane crude extract of *S. mombin*, Carvacrol with molecular mass of 150.22 g/mol and a formula of  $C_{10}H_{14}O$  was identified and has been reported by other authors to possess antiviral effect against herpes simplex virus types 1 (HSV-1), SARS-CoV-2, immunomodulatory and anti-inflammatory (Elbe et al. 2020; Javed et al. 2020). In addition to Carvacrol being identified as antiviral, other antiviral phytochemicals were also identified in polar crude extracts such as, methanol leaf extracts of *S. mombin* (SMM). These compounds with reported antiviral activities were D-Limonene has antiCoVID properties (Zahi et al. 2015; Panikar et al. 2021), p-Cymene, Thymol. This confirms the traditional use of leaf extracts of *S. mombin* in the treatment of viral diseases (Osuntokun 2019; Abubakar et al. 2022; Maria et al. 2022).

In SMDCM, n-Nonadecanol-1(19.64%) and Eicosane (13.93%) are the major compounds, while in SMEA, Ethanol, 2-butoxy- (83.29%). Both SME and SMM showed Tetradecyl trifluoroacetate (15.43%), Pentadecanoic acid (12.18%), Propane, 2,2-diethoxy- (33.83%) and o-Xylene (15.87%).

The SME extract chromatogram analysis revealed the highest number of compounds, i.e., 51, whereas the SMEA extract revealed the lowest number at 12, with almost all exhibiting various pharmacological activities. Phytol is an acyclic diterpene alcohol that shows anti-cancer, anti-microbial and anti-oxidant activities. Hexadecanoic acid, or palmitic acid, is known to possess anti-cancer, anti-inflammatory and anti-microbial activity. Eicosane, an alkane, is also known to exhibit anti-tumor and anti-cancer effects (Sivasubramanian and Brindha 2013). Tetradecane is an alkane that possesses anti-fungal, anti-tumor, anti-cancer and anti-proliferative activities (Guo et al. 2008; Erenler et al. 2016). While the majority of the compounds are yet to be illustrated in detail, more research attempts are needed to isolate, characterize and assess these compounds from *S. mombin* leaves to justify their various pharmacological relevance.

## Conclusion

Results obtained from the phytochemical compounds of essential leaf oils (EO) from *S. mombin* (Linn) indicated that a total of 169 compounds was identified in SMH, SMDCM, SMEA SME, and SMM. Identified antiviral compounds in the EOs, were D-Limonene (1.33%), p-Cymene (1.31%), Thymol (0.50%) in SMM and Carvacrol (0.87%) in SMH extracts respectively. Phytol, acetate is a common constituent in all the essential oils except SMEA.

The in vitro cytotoxicity study of EOs of SM leaf extracts MCF-7, A-549 and HEK-293 indicate low toxicity. The *S. mombin* essential oils showed  $IC_{50}$  ranges between 15.91 and 178.5  $\mu\text{g/mL}$ . The methanolic extract has the most compounds with antiviral properties that can be used as repurposing therapeutic candidates, natural supplements, or valuable for drug formulation in the fight against SARS-CoV-2, Ebola and Marburg viruses.

Further isolation of essential oil compounds, in silico and in vitro SARS-CoV-2 and Ebola or Marburg virus with A-549/HeLa infected cell lines from these essential oils is under process. Overall, this *S. mombin* leaf extract is an auspicious and treasured source of different bioactive compounds that could have tremendous health benefits.

To the best of our knowledge, Carvacrol and 11-Methyldodecanol have been identified for the first time in *S. mombin* leaf extract.

## Appendix



Preliminary phytochemical screening of *S. mombin* leaf extracts.

***Spondias mombin*: biosafety and GC–MS analysis of anti-viral compounds from Crude Leaf Extracts”**

Fractionated leaf extracts of *S. mombin*.



*Spondias mombin* dried leaf powder.  
See Tables 3, 4, 5, 6 and 7.

**Table 3** The GC–MS analysis data of SMH chromatogram with biological activity

Peak	Name	Molecular weight (g/mol)	Molecular formula	Retention time (min)	Area%	Class of phytochemical	Biological activity	References
1	Heptane, 2-methyl-	114.23	C <sub>8</sub> H <sub>18</sub>	3.569	3.06	Hydrocarbon	–	–
2	Toluene	92.14	C <sub>7</sub> H <sub>8</sub>	3.629	15.13	Hydrocarbon	Anxiolytic effect, depressant effect Induces locomotor hypersensitivity	Armenta-Reséndiz et al. (2019), Rangel-Sánchez et al. (2014)
3	Heptane, 2,4-dimethyl-	128.25	C <sub>9</sub> H <sub>20</sub>	3.960	2.63	Hydrocarbon	–	–
4	Tetrachloroethylene	165.8	C <sub>2</sub> Cl <sub>4</sub>	4.077	4.10	Chlorinated hydrocarbon	Anticonflict effect Anthelmintic activity in vivo in dogs	Umezū et al. (1997), Miller (1966)
5	2,4-Dimethyl-1-heptene	126.24	C <sub>9</sub> H <sub>18</sub>	4.463	0.43	Hydrocarbon	–	–
6	Ethylbenzene	106.16	C <sub>8</sub> H <sub>10</sub>	4.775	0.79	Aromatic	–	–
7	o-Xylene	106.16	C <sub>8</sub> H <sub>10</sub>	4.897	2.49	Aromatic	–	–
8	Cyclopropane, 2-chloro-1,1,3-trimethyl-	118.6	C <sub>6</sub> H <sub>11</sub> Cl	5.203	2.67	Chlorinated Alkane	–	–
9	Nonane	128.25	C <sub>9</sub> H <sub>20</sub>	5.240	0.30	Hydrocarbon	–	–
10	Benzene, 1-ethyl-3-methyl	162.27	C <sub>12</sub> H <sub>18</sub>	6.132	0.32	Aromatic	–	–
11	Undecane	156.31	C <sub>11</sub> H <sub>24</sub>	6.614	0.98	Hydrocarbon	–	–
12	Benzene, 1-methyl-3-propyl-	134.22	C <sub>10</sub> H <sub>14</sub>	7.370	0.31	Aromatic	–	–
13	Benzene, 1,4-diethyl-	134.22	C <sub>10</sub> H <sub>14</sub>	7.454	0.61	Aromatic	–	–
14	5-Chloropentanoic acid, 6-ethyl-3-octyl ester	276.84	C <sub>15</sub> H <sub>29</sub> ClO <sub>2</sub>	7.713	0.40	Aromatic	–	–
15	Undecane, 4,7-dimethyl-	184.36	C <sub>13</sub> H <sub>28</sub>	7.970	0.38	Hydrocarbon	–	–
16	2-Isopropyl-5-methyl-1-heptanol	172.31	C <sub>11</sub> H <sub>24</sub> O	10.490	0.44	Primary alcohol	–	–
17	Carvacrol	150.22	C <sub>10</sub> H <sub>14</sub> O	10.537	0.87	monoterpene	Antiviral effect against herpes simplex virus types 1 (HSV-1), SARS-CoV-2, immunomodulatory, anti-inflammatory, and antiviral effects	Elbe et al. (2020), Javed et al. (2020)
18	11-Methyldodecanol	200.36	C <sub>13</sub> H <sub>28</sub> O	10.591	0.73	Alcohol	–	–
19	1-Tetradecene	196.37	C <sub>14</sub> H <sub>28</sub>	11.552	2.24	Hydrocarbon	–	–
20	Tetrapentacontane, 1,54-dibromo-	917.2	C <sub>54</sub> H <sub>108</sub> Br <sub>2</sub>	13.266	0.37	Hydrocarbon	–	–
21	Dichloroacetic acid, 4-hexadecyl ester	353.4	C <sub>18</sub> H <sub>34</sub> Cl <sub>2</sub> O <sub>2</sub>	13.623	0.40	–	–	–
22	n-Nonadecanol-1	284.5	C <sub>19</sub> H <sub>40</sub> O	29.505	7.81	Primary alcohol	–	Ahmad et al. (2021)

Table 3 (continued)

Peak Name	Molecular weight (g/mol)	Molecular formula	Retention time (min)	Area%	Class of phytochemical	Biological activity	References
23 Tetradecanoic acid	228.37	C <sub>14</sub> H <sub>28</sub> O <sub>2</sub>	15.514	1.40	Carboxylic acid	Antimicrobial, antispasmodic and anti-inflammatory effects	Sosa et al. (2016)
24 Heneicosane	296.6	C <sub>21</sub> H <sub>44</sub>	15.898	0.35	Saturated hydrocarbon	Antimicrobial activity	Vanitha et al. (2020)
25 Phytol, acetate	338.6	C <sub>22</sub> H <sub>42</sub> O <sub>2</sub>	16.394	0.55	Terpene Alcohol	Anti-oxidant and Antimicrobial	Syeda and Riazunnisa (2020)
26 2-Pentadecanone, 6,10,14-trimethyl-	268.5	C <sub>18</sub> H <sub>36</sub> O	16.489	0.55	Ketone	Has antibacterial effect in curing diarrhoea	Nayak et al. (2020)
27 l-(+)-Ascorbic acid 2,6-dihexadecanoate	652.9	C <sub>38</sub> H <sub>68</sub> O <sub>8</sub>	18.813	8.70	Vitamin-C	Anticancer	Hase et al. (2017)
28 n-Tetracosanol-1	354.7	C <sub>24</sub> H <sub>50</sub> O	19.292	4.88	Alcohol	Antioxidant	Amudha et al. (2018)
29 Phytol	296.5	C <sub>20</sub> H <sub>40</sub> O	22.563	1.80	Diterpene alcohol	Antioxidant, anticonvulsant and antinociceptive effects	Silva et al. (2014), Santos et al. (2013), Costa et al. (2012), Shibula and Velavan (2015)
30 Linoleic Acid	280.4	C <sub>18</sub> H <sub>32</sub> O <sub>2</sub>	23.658	3.34	fatty acids	Anti-bacteria action against the growth of Staphylococcus aureus NCTC 8325	Greenway and Dyke (1979)
31 cis-9-Hexadecenal	238.41	C <sub>16</sub> H <sub>30</sub> O	23.945	4.01	mono-unsaturated fatty-aldehyde	Antimelanogenic	Kumari and Shankar (2021)
32 Stearic Acid	284.5	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	24.88	2.58	Acid	Immunomodulatory effect	Bergamo et al. (2014)
33 1-Heptacosanol	396.7	C <sub>27</sub> H <sub>56</sub> O	25.953	3.59	Alcohol	Antibacterial, nematocidal and antioxidant effects	Murugan and Iyer (2014)
34 Diisooctyl phthalate	390.6	C <sub>24</sub> H <sub>38</sub> O <sub>4</sub>	32.768	14.21	Plasticizer	Anti-microbial activity, and inhibited the growth of food-borne harmful bacteria	Naz et al. (2021), Yang et al. (2020)

**Table 4** The GC–MS analysis data of SMDCM chromatogram with biological activity

Peak Name	Molecular weight (g/mol)	Molecular formula	Retention time (min)	Area%	Class of phytochemical	Biological activity	References
1 Toluene	92.14	C <sub>7</sub> H <sub>8</sub>	3.631	9.80	Hydrocarbon	Anxiolytic effect, depressant effect and Induces locomotor hypersensitivity	Armenta-Reséndiz et al. (2019), Riegel and French (1999)
2 Cyclopropanecarboxylic acid, 2-pentyl ester	156.22	C <sub>9</sub> H <sub>16</sub> O <sub>2</sub>	4.006	0.14	–	–	–
3 Ethanol, 2-butoxy-	118.17	C <sub>6</sub> H <sub>14</sub> O <sub>2</sub>	5.350	1.23	Alcohol	–	–
4 Cyclobutane, 1,1-bis (1-methyl-ethenyl-, trans	136	C <sub>10</sub> H <sub>16</sub>	7.085	3.34	Hydrocarbon	–	–
5 Trans-p-Mentha-2,8-dienol	152.23	C <sub>10</sub> H <sub>16</sub> O	9.157	1.45	Monoterpene alcohol	Anti-proliferative and antioxidant	Alam et al. (2019), Brito et al. (2012), Bayala et al. (2018)
6 Cyclohexene,3-(3-methyl-1-butanyl)-,(E)-	150	C <sub>11</sub> H <sub>18</sub>	9.732	0.19	Hydrocarbon	–	–
7 Cis-p-mentha-1(7),8-dien-2-ol	152.23	C <sub>10</sub> H <sub>16</sub> O	9.775	0.16	Monoterpene alcohol	Anti-proliferative and antioxidant	Brito et al. (2012), Bayala et al. (2018)
8 2-Hydroxy-iso-butyrophenone	164.2	C <sub>10</sub> H <sub>12</sub> O <sub>2</sub>	10.416	0.56	Ketone	–	–
9 Tetradecene	196.37	C <sub>14</sub> H <sub>28</sub>	11.632	2.16	Hydrocarbon	–	–
10 Ferimine	194.18	C <sub>10</sub> H <sub>10</sub> O <sub>4</sub>	12.260	0.33	Plastersizer	–	–
11 Propioldrazone, 3-benzoylamino-N2-(2-hydroximinol-1-methylpropylideno)	290	C <sub>14</sub> H <sub>18</sub> N <sub>4</sub> O <sub>3</sub>	12.630	0.63	–	–	–
12 Pentadecane	212.41	C <sub>15</sub> H <sub>32</sub>	12.715	0.22	Hydrocarbon	Anti-microbial against Leishmania parasites	Bruno et al. (2015)
13 Phenol, 2,4-bis(1,1-dimethyl-ethyl)-	206.32	C <sub>14</sub> H <sub>22</sub> O	12.903	0.15	Monoterpene	Antifungal activity	Rangel-Sánchez et al. 2014)
14 2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R)-	180	C <sub>11</sub> H <sub>16</sub> O <sub>2</sub>	13.255	0.26	Furan	Antidiabetic and anti-obesity property	Carranza et al. (2020)
15 Hexamethylene diacrylate	226.27	C <sub>12</sub> H <sub>18</sub> O <sub>4</sub>	13.598	3.91	–	–	–
16 n-Pentadecanol	228.41	C <sub>15</sub> H <sub>32</sub> O	13.673	2.83	Aliphatic alcohol	–	Naquvi et al. (2014)
17 Heptadecane	240.5	C <sub>17</sub> H <sub>36</sub>	12.742	0.46	Hydrocarbon	Antioxidant activity	Shyamala and Manikandan (2019)
18 Dihydrojasnone	166.26	C <sub>11</sub> H <sub>18</sub> O	13.912	0.27	Aromatic compound	–	–
19 3-Benzoyl-2-t-butyl-4-methyl-oxazolidin-5one	261	C <sub>15</sub> H <sub>19</sub> NO <sub>3</sub>	14.175	0.60	–	–	–
20 Heptadecane,3-methyl-	254.5	C <sub>18</sub> H <sub>38</sub>	15.527	0.21	–	–	–
21 Tridecane,3-methylene	196.37	C <sub>14</sub> H <sub>28</sub>	15.721	6.52	Hydrocarbon	–	–
22 n-Nonadecanol-1	284.5	C <sub>19</sub> H <sub>40</sub> O	15.816	19.64	Primary alcohol	–	–
23 Phytol, acetate	338.6	C <sub>22</sub> H <sub>42</sub> O <sub>2</sub>	16.393	1.06	Terpene Alcohol	Antioxidant and Antimicrobial	Syeda and Riazunnisa (2020)

Table 4 (continued)

Peak Name	Molecular weight (g/mol)	Molecular formula	Retention time (min)	Area%	Class of phytochemical	Biological activity	References
24 2-Pentadecanone,6,10,14-trimethyl-	268.5	C <sub>18</sub> H <sub>36</sub> O	16.484	0.29	–	–	–
25 Octadecane,4-methyl-	268.5	C <sub>19</sub> H <sub>40</sub>	16.484	0.15	–	–	–
26 7-Octadecyne, 2-methyl-	264.5	C <sub>19</sub> H <sub>36</sub>	16.754	0.29	–	–	–
27 Phytol	296.5	C <sub>30</sub> H <sub>40</sub> O	17.052	0.49	Diterpene alcohol	cancer-preventive Antimicrobial, anti-inflammatory anti-diuretic Antioxidant	Silva et al. (2014), Santos et al. (2013), Costa et al. (2012), Shibula and Velavan (2015)
28 7,9-Di-tert-butyl-1-oxaspiro(4,5)deca-6,9-diene-2,8-dione	276.4	C <sub>17</sub> H <sub>24</sub> O <sub>3</sub>	17.631	0.72	Flavonoid	Antibacterial activity	Monisha and Virmala (2018)
29 Hexadecanoic acid,methyl ester	270.5	C <sub>17</sub> H <sub>34</sub> O <sub>2</sub>	17.834	1.56	Fatty acid methyl ester	Anti-oxidant, decrease blood cholesterol, anti-inflammatory	Belakhdar et al. (2015)
30 Phthalic acid,butyl hexyl ester	306.4	C <sub>18</sub> H <sub>26</sub> O <sub>4</sub>	18.099	2.11	phthalate ester	–	–
31 1-(+)-Ascorbic acid 2,6-dihexadecanoate	652	C <sub>38</sub> H <sub>68</sub> O <sub>8</sub>	18.505	0.25	Acid	Anti-oxidant and anti-inflammatory	Dulara et al. (2019)
32 Tridecane,3-methylene-	196.37	C <sub>14</sub> H <sub>28</sub>	18.699	5.99	Hydrocarbon	–	–
33 n-Tetracosanol-1	354.7	C <sub>24</sub> H <sub>50</sub> O	19.105	13.85	Alcohol	Anti-oxidant	Amudha et al. (2018)
34 Nonadecane, 2,3-dimethyl-	296.6	C <sub>21</sub> H <sub>44</sub>	20.873	0.39	Hydrocarbon	–	–
35 1-Heptacosanol	396.7	C <sub>27</sub> H <sub>56</sub> O	25.934	4.02	Alcohol	Anti-bacterial, antioxidant and nematocidal effect	Naquvi et al. (2014), Sultana et al. (2010)
36 Eicosane	282.5	C <sub>20</sub> H <sub>42</sub>	26.230	13.93	Hydrocarbon	Anti-oxidant and anti-inflammatory effect	Dulara et al. (2019)

**Table 5** The GC–MS analysis data of SMEA chromatogram with biological activity

Peak	Name	Molecular weight	Molecular formula	Retention time (min)	Area%	Class of phytochemical	Biological activity	References
1	Tetrachloroethylene	164	C <sub>2</sub> C <sub>14</sub>	4.107	0.40	Chlorinated hydrocarbon	Anticonflict effect Anthelmintic activity in dogs	Umezu et al. (1997), Miller (1966)
2	Alpha-Hydroxyisocaproic acid	132	C <sub>6</sub> H <sub>12</sub> O <sub>3</sub>	4.190	0.46	Acid	–	–
3	2-Hexanol, 2-methyl-	116	C <sub>7</sub> H <sub>16</sub> O	4.530	1.61	Primary alcohol	–	–
4	Ethanol, 2-butoxy-	118	C <sub>6</sub> H <sub>14</sub> O <sub>2</sub>	5.453	83.29	Primary alcohol	Antibacterial effect	Elshafie et al. (2017)
5	(R)-(-)-2,2-Dimethyl-1,3-dioxolane-4-methanol	132	C <sub>6</sub> H <sub>12</sub> O <sub>3</sub>	5.898	0.24	–	–	–
6	Ethane, pentachloro-	200	C <sub>2</sub> HC <sub>15</sub>	6.442	0.32	Chlorinated hydrocarbon	Anti-parasitic effect on <i>F. hepatica</i>	Bartlet (1976)
7	Ethanol, 1-(2-butoxyethoxy)-	162	C <sub>8</sub> H <sub>18</sub> O <sub>3</sub>	9.151	11.12	–	–	–
8	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7	180	C <sub>11</sub> H <sub>16</sub> O <sub>2</sub>	13.249	0.37	Ketone	Antifungal, antioxidant and antibacterial effect	Anyasor et al. (2015)
9	Dihydrojasmane	166	C <sub>11</sub> H <sub>18</sub> O	13.905	0.23	Aromatic compound	–	–
10	4-[3,4-Dimethoxycyclohexyl]-n-butanol	216	C <sub>12</sub> H <sub>24</sub> O <sub>3</sub>	14.087	0.41	Alcohol	–	–
11	1,9,12,15-Octadecatetraene, 1-methoxy-	276	C <sub>19</sub> H <sub>32</sub> O	14.169	0.29	–	–	–
12	Acetic acid, 2-(2,2,6-trimethyl-7-oxa-bicyclo[4.1	238	C <sub>14</sub> H <sub>22</sub> O <sub>3</sub>	15.878	1.25	–	–	–

### GC–MS chromatogram: crude leaf extracts *S. mombin* (Linn)

See Figs. 6, 7, 8, 9, 10, 11 and 12.

Table 6 The GC-MS analysis data of SME chromatogram with biological activity

Peak	Name	Molecular weight	Molecular formula	Retention time (min)	Area%	Class of phytochemical	Biological activity	References
1	Leucinic acid	132.16	C <sub>6</sub> H <sub>12</sub> O <sub>3</sub>	4.185	1.41	Valeric acid	-	-
2	Cyclohexene, 3-(1-methyl-ethyl)-	124.22	C <sub>9</sub> H <sub>16</sub>	5.072	0.33	-	-	-
3	Ethanol, 2-butoxy-	118.17	C <sub>6</sub> H <sub>14</sub> O <sub>2</sub>	5.397	0.68	Primary alcohol	-	-
4	Silane, [3-(2,3-epoxypropoxy)propyl]ethoxydimethyl-	218.36	C <sub>10</sub> H <sub>22</sub> O <sub>3</sub> Si	6.505	1.40	-	-	-
5	tert-Butyldimethylsilyl 2-acetoxyacetate	232.35	C <sub>10</sub> H <sub>20</sub> O <sub>4</sub> Si	9.089	0.53	-	-	-
6	Undecane, 6-ethyl-	184.36	C <sub>13</sub> H <sub>28</sub>	10.135	0.52	-	-	-
7	Nonane, 3-methyl-5-propyl-	184.36	C <sub>13</sub> H <sub>28</sub>	10.483	0.91	Hydrocarbon	Prevent osteoporosis	Diwan et al. (2011)
8	Phenol, 3-methyl-5-(1-methyl-ethyl)-, methylcarbamate	207.27	C <sub>12</sub> H <sub>17</sub> NO <sub>2</sub>	10.601	0.45	Alkylbenzene	-	-
9	Dodecane, 2,6,11-trimethyl-	212.41	C <sub>15</sub> H <sub>32</sub>	11.345	0.39	Alkane	-	-
10	1-Tetradecene	196.37	C <sub>14</sub> H <sub>28</sub>	11.545	2.07	Hydrocarbon	-	-
11	Tetradecane	198.39	C <sub>14</sub> H <sub>30</sub>	11.627	1.64	Hydrocarbon	Antimicrobial, antipyretic, anthelmintic, and Tuberculosis effect	Diwan et al. (2011)
12	Dodecane, 2,6,11-trimethyl-	212.41	C <sub>15</sub> H <sub>32</sub>	12.711	1.15	Alkane	-	-
13	Phenol, 2,4-bis(1,1-dimethyl-ethyl)-	206.32	C <sub>14</sub> H <sub>22</sub> O	12.929	0.60	Phenol	Antifungal activity	Diwan et al. (2011)
14	2-(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-	180.24	C <sub>11</sub> H <sub>16</sub> O <sub>2</sub>	13.250	0.92	Ketone	Antifungal, antioxidant and antibacterial effects	Anyasor et al. (2015)
15	Tetradecyl trifluoroacetate	310.39	C <sub>16</sub> H <sub>28</sub> F <sub>3</sub> O <sub>2</sub>	13.662	15.43	-	-	-
16	Heptadecane, 2,6,10,15-tetramethyl-	296.6	C <sub>21</sub> H <sub>44</sub>	13.730	2.23	Hydrocarbon	-	-
17	2-Undecenal	168.28	C <sub>11</sub> H <sub>20</sub> O	13.775	0.60	Hydrocarbon	Anti-leishmanial, Antibacterial and Anti-cancer activities	Diwan et al. (2011)
18	1,2-Oxathiane, 6-dodecyl-, 2,2-dioxide	304.5	C <sub>16</sub> H <sub>32</sub> O <sub>3</sub> S	13.899	0.37	-	Antimicrobial activity	Anbukumar et al. (2021)
19	Benzene, (1-pentylhexyl)-	232.4	C <sub>17</sub> H <sub>28</sub>	14.040	0.29	Alkylbenzene	-	Shoubaky and Salem (2014)
20	Benzene, (1-butylheptyl)-	232.4	C <sub>17</sub> H <sub>28</sub>	14.081	0.64	Alkylbenzene	-	Shoubaky and Salem (2014)
21	Benzene, (1-propyloctyl)-	232.4	C <sub>17</sub> H <sub>28</sub>	14.187	0.61	Alkylbenzene	-	Shoubaky and Salem (2014)
22	Benzene, (1-ethylnonyl)-	232.4	C <sub>17</sub> H <sub>28</sub>	14.416	0.38	Alkylbenzene	-	Shoubaky and Salem (2014)
23	Benzene, (1-methyldecyl)-	232.4	C <sub>17</sub> H <sub>28</sub>	14.812	0.46	Alkylbenzene	-	Shoubaky and Salem (2014)
24	Benzene, (1-pentylheptyl)-	232.4	C <sub>17</sub> H <sub>28</sub>	15.038	0.38	Alkylbenzene	-	Shoubaky and Salem (2014)
25	Benzene, (1-butylloctyl)-	232.4	C <sub>17</sub> H <sub>28</sub>	15.095	0.41	Alkylbenzene	-	Shoubaky and Salem (2014)

Table 6 (continued)

Peak	Name	Molecular weight	Molecular formula	Retention time (min)	Area%	Class of phytochemical	Biological activity	References
26	Benzene, (1-propylonyl)-	232.4	C <sub>17</sub> H <sub>28</sub>	15.244	0.61	Alkylbenzene	–	Shoubaky and Salem (2014)
27	6,9,12-Octadecatrienoic acid, phenylmethyl ester, (Z,Z,Z)-	368.6	C <sub>23</sub> H <sub>36</sub> O <sub>2</sub>	15.523	1.16	–	Antioxidant, anti-inflammatory, antimicrobial, pesticide and cancer-preventive activity	Al-Rubaye et al. (2017)
28	E-7-Octadecene	252.5	C <sub>18</sub> H <sub>36</sub>	15.730	0.69	Hydrocarbon	–	–
29	Heptadecane, 2,6,10,15-tetra-methyl-	296.6	C <sub>21</sub> H <sub>44</sub>	15.876	4.42	Alkane	–	–
30	Benzene, (1-methylundecyl)-	246.4	C <sub>18</sub> H <sub>30</sub>	16.039	0.44	Alkylbenzene	–	Shoubaky and Salem (2014)
31	Benzene, (1-pentylloctyl)-	260.5	C <sub>19</sub> H <sub>32</sub>	16.253	0.50	Alkylbenzene	–	Shoubaky and Salem (2014)
32	Phytol, acetate	338.6	C <sub>22</sub> H <sub>42</sub> O <sub>2</sub>	16.375	5.91	Terpene Alcohol	Antioxidant and Antimicrobial	Syeda and Riazunnisa (2020)
33	Phytone	268.5	C <sub>18</sub> H <sub>36</sub> O	16.465	3.55	Ketone	–	–
34	Benzene, (1-propyldecyl)-	260.5	C <sub>19</sub> H <sub>32</sub>	16.555	0.57	Alkylbenzene	–	–
35	Phytol	296.5	C <sub>20</sub> H <sub>40</sub> O	16.733	4.32	Diterpene Alcohol	Cancer-preventive Antimicrobial anti-inflammatory anti-diuretic Antioxidant	Silva et al. (2014), Santos et al. (2013), Costa et al. (2012), Shibula and Velavan (2015) Figueiredo et al. (2018), Yan et al. (2020)
36	Geranyl benzoate	258.35	C <sub>17</sub> H <sub>22</sub> O <sub>2</sub>	17.607	1.06	–	–	Al-Salman (2019)
37	2-Piperidinone, N-[4-bromo-n-butyl]s-	234.13	C <sub>9</sub> H <sub>16</sub> BrNO	17.735	0.69	Amide/delta-lactams	Antimicrobial activity	–
38	Pentadecanoic acid, methyl ester	270.5	C <sub>17</sub> H <sub>34</sub> O <sub>2</sub>	17.810	1.90	–	Antioxidant, antimicrobial and fungal activity	Elaiyaraja and Chandramohan (2016)
39	Pentadecanoic acid	242.4	C <sub>15</sub> H <sub>30</sub> O <sub>2</sub>	18.695	12.18	Fatty acid	–	–
40	n-Tridecan-1-ol	200.36	C <sub>13</sub> H <sub>28</sub> O	19.130	0.36	Fatty Alcohol	Natural mosquito control agent	Arora and Kumar (2018)
41	n-Nonadecanol-1	284.5	C <sub>19</sub> H <sub>40</sub> O	19.233	7.16	Fatty Alcohol	–	–
42	Eicosane	282.5	C <sub>20</sub> H <sub>42</sub>	19.366	1.66	Hydrocarbon	Anti-oxidant and antiinflammatory effect	Dulara et al. (2019)
43	Methyl 5,12-octadecadienoate	294.5	C <sub>19</sub> H <sub>34</sub> O <sub>2</sub>	21.906	0.83	–	–	–
44	Dehydroisoholtzia ketone	164.2	C <sub>10</sub> H <sub>12</sub> O <sub>2</sub>	22.100	0.40	–	–	–
45	Oxirane, hexadecyl-	268.5	C <sub>18</sub> H <sub>36</sub> O	22.140	1.28	Epoxide	Adhesive	Elangovan et al. (2015)
46	Heptadecanoic acid, 16-methyl-, methyl ester	298.5	C <sub>19</sub> H <sub>38</sub> O <sub>2</sub>	23.059	1.03	–	Potent skin cancer activity	Kandasamy et al. (2012)
47	trans-Undec-4-enal	168.28	C <sub>11</sub> H <sub>20</sub> O	23.549	1.92	Hydrocarbon	–	–
48	7-Hexadecyn-1-ol	238.41	C <sub>16</sub> H <sub>30</sub> O	23.779	3.92	Alcohol	–	–
49	n-Tetracosanol-1	354.7	C <sub>24</sub> H <sub>50</sub> O	25.839	5.89	Alcohol	Anti-oxidant effect	Amudha et al. (2018)
50	2-Bromotetradecane	277.28	C <sub>14</sub> H <sub>29</sub> Br	26.103	1.31	–	–	–

Table 7 The GC-MS analysis data of SMM chromatogram with biological activity

Peak Name	Molecular weight	Molecular formula	Retention time (min)	Area%	Class of phytochemical	Biological activity	References
1 Propane, 2,2-diethoxy-	132.2	C <sub>7</sub> H <sub>16</sub> O <sub>2</sub>	3.622	33.83	-	-	-
2 Cyclotrisiloxane, hexamethyl-	222.46	C <sub>6</sub> H <sub>18</sub> O <sub>3</sub> Si <sub>3</sub>	4.182	0.52	-	Antimicrobial, antioxidant, antibacterial activity	Ismail et al. (2019)
3 2-Hexanol, 2-methyl-	116.2	C <sub>7</sub> H <sub>16</sub> O	4.646	0.84	Alcohol	-	-
4 Ethylbenzene	106.16	C <sub>8</sub> H <sub>10</sub>	4.790	2.92	Alkylbenzene	-	-
5 o-Xylene	106.16	C <sub>8</sub> H <sub>10</sub>	4.933	15.87	Alcohol	-	-
6 Ethanol, 2-butoxy-	118.17	C <sub>6</sub> H <sub>14</sub> O <sub>2</sub>	5.460	0.48	-	-	-
7 Propanoic acid, 2,2-dimethyl-, 2-ethyl-hexyl ester	214.34	C <sub>13</sub> H <sub>26</sub> O <sub>2</sub>	6.075	0.51	-	-	-
8 2-Hexyl-1-octanol	214.39	C <sub>14</sub> H <sub>30</sub> O	6.150	1.16	Alcohol	-	-
9 Benzene, 1-ethyl-3-methyl-	120.19	C <sub>9</sub> H <sub>12</sub>	6.190	1.14	-	-	-
10 Heptane, 5-ethyl-2-methyl-	142.28	C <sub>10</sub> H <sub>22</sub>	6.235	0.82	-	-	-
11 2',6'-2',6'-Dihydroxyacetophenone, bis(trimethylsilyl) ether	296.51	C <sub>14</sub> H <sub>24</sub> O <sub>3</sub> Si <sub>2</sub>	6.306	0.76	-	Antimicrobial activity	Pradhan and Dubey (2021)
12 Decane	142.28	C <sub>10</sub> H <sub>22</sub>	6.645	6.75	Alkane	-	-
13 Benzene, (1-methylpropyl)-	134.22	C <sub>10</sub> H <sub>14</sub>	6.850	0.80	Alkylbenzene	-	-
14 Decane, 4-methyl-	156.31	C <sub>11</sub> H <sub>24</sub>	6.939	1.40	-	-	-
15 PseudoCumene	120.19	C <sub>9</sub> H <sub>12</sub>	7.024	2.16	-	-	-
16 D-Limonene	136.23	C <sub>10</sub> H <sub>16</sub>	7.114	1.33	-	AntiCOVID and Antimicrobial activity	Zahi et al. (2015), Panikar et al. (2021)
17 1-Decanol, 2-hexyl-	242.44	C <sub>16</sub> H <sub>34</sub> O	7.165	1.23	-	-	-
18 Indane	118.18	C <sub>9</sub> H <sub>10</sub>	7.225	0.23	-	-	-
19 Benzene, 1-methyl-3-propyl-	134.22	C <sub>10</sub> H <sub>14</sub>	7.396	1.01	-	-	-
20 Benzene, 1,4-diethyl-	134.22	C <sub>10</sub> H <sub>14</sub>	7.478	3.10	-	-	-
21 Benzene, (1,3,3-trimethylonyl)-	246.4	C <sub>18</sub> H <sub>30</sub>	7.596	1.79	-	-	-
22 Benzene, 1-ethyl-2,3-dimethyl-	134.22	C <sub>10</sub> H <sub>14</sub>	7.731	0.68	-	-	-
23 Benzene, 1-methyl-3-(1-methylethyl)-	134.22	C <sub>10</sub> H <sub>14</sub>	7.773	0.89	-	-	-
24 p-Cymene	134.22	C <sub>10</sub> H <sub>14</sub>	7.854	1.31	Monoterpene	AntiCovid-19, antioxidant, anti-inflammatory, antiparasitic, antidiabetic, antifungal and antitumor activities	Panikar et al. (2021), Balabbib et al. (2021), Sharifi-Rad et al. (2018)
25 Undecane	156.31	C <sub>11</sub> H <sub>24</sub>	7.984	4.12	-	-	-
26 Benzenepropanal, beta.-methyl-	148.2	C <sub>10</sub> H <sub>12</sub> O	8.056	0.56	-	-	-
27 Spiro[3.5]nona-5,7-dien-1-one, 5,9,9-trimethyl	176.25	C <sub>12</sub> H <sub>16</sub> O	8.124	1.18	-	-	-
28 Benzene, 2,4-diethyl-1-methyl-	148.24	C <sub>11</sub> H <sub>16</sub>	8.185	0.32	-	-	-

Table 7 (continued)

Peak	Name	Molecular weight	Molecular formula	Retention time (min)	Area%	Class of phytochemical	Biological activity	References
29	Benzene, 1,2,4,5-tetramethyl-	134.22	C <sub>10</sub> H <sub>14</sub>	8.335	0.48	-	-	-
30	10-Chloro-1-decanol, pentafluoropropionate	338.74	C <sub>13</sub> H <sub>20</sub> ClF <sub>5</sub> O <sub>2</sub>	8.505	0.47	-	-	-
31	Benzene, 1,3-diethyl-5-methyl-	148.24	C <sub>11</sub> H <sub>16</sub>	8.600	0.78	-	-	-
32	1,7,7-Trimethyl-2-vinylbicyclo[2.2.1]hept-2-ene	162.27	C <sub>12</sub> H <sub>18</sub>	8.815	0.32	-	-	-
33	Dodecane	170.33	C <sub>12</sub> H <sub>26</sub>	9.266	0.67	-	-	-
34	Pentadecane	212.41	C <sub>15</sub> H <sub>32</sub>	10.486	0.49	-	-	-
35	Thymol	150.22	C <sub>10</sub> H <sub>14</sub> O	10.595	0.50	Monoterpene	Antimicrobial, anti-inflammatory and antioxidant activity Antiviral SARS-CoV-2	Braga et al. (2006), Rolta et al. (2021), Abdelli et al. (2021), Kulkarni et al. (2020)
36	Phytol, acetate	338.6	C <sub>22</sub> H <sub>42</sub> O <sub>2</sub>	16.375	0.29	Terpene Alcohol	Anti-oxidant and Anti-microbial	Syeda and Riazumma (2020)

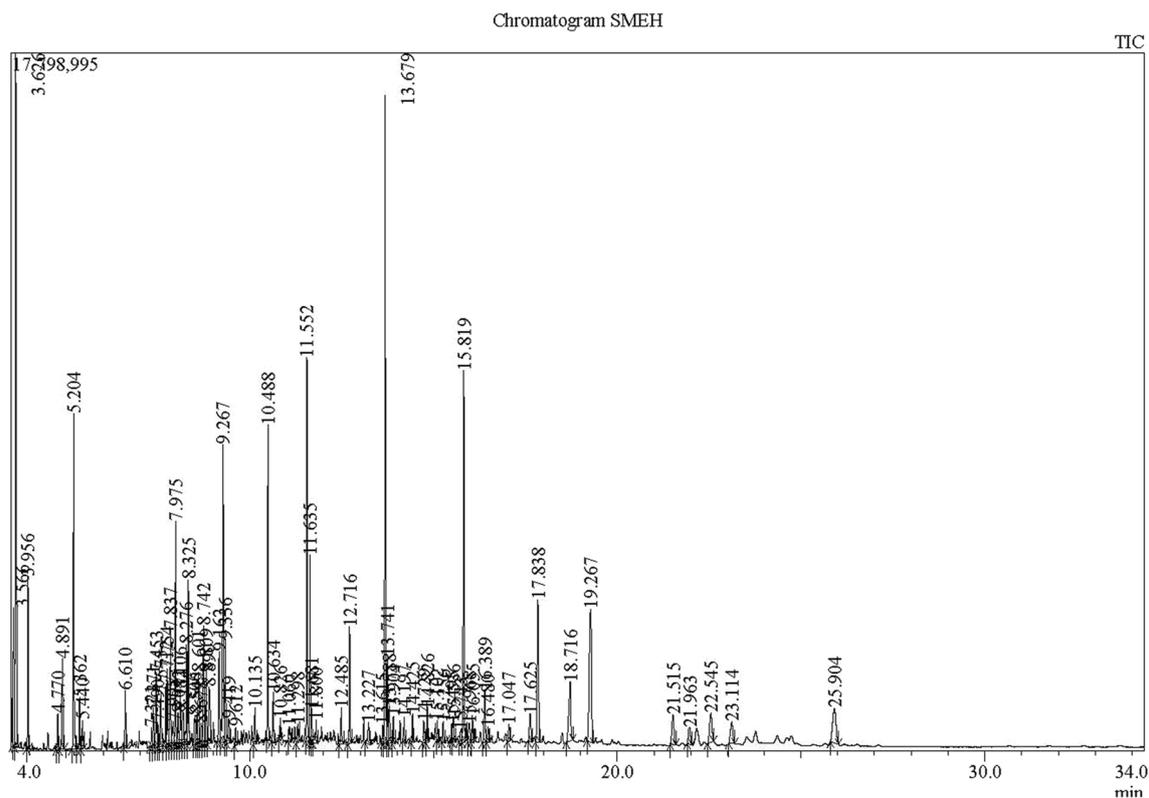


Fig. 6 GC-MS chromatogram for SMEH

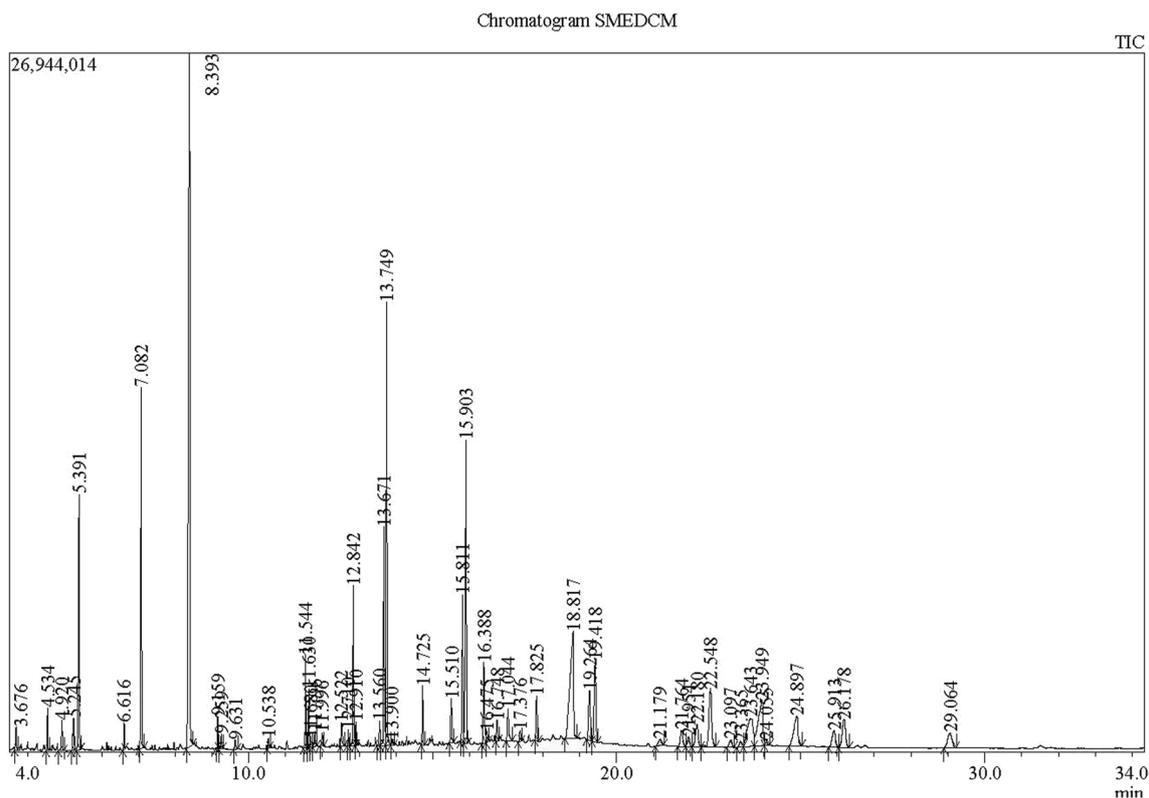


Fig. 7 GC-MS chromatogram for SMEDCM

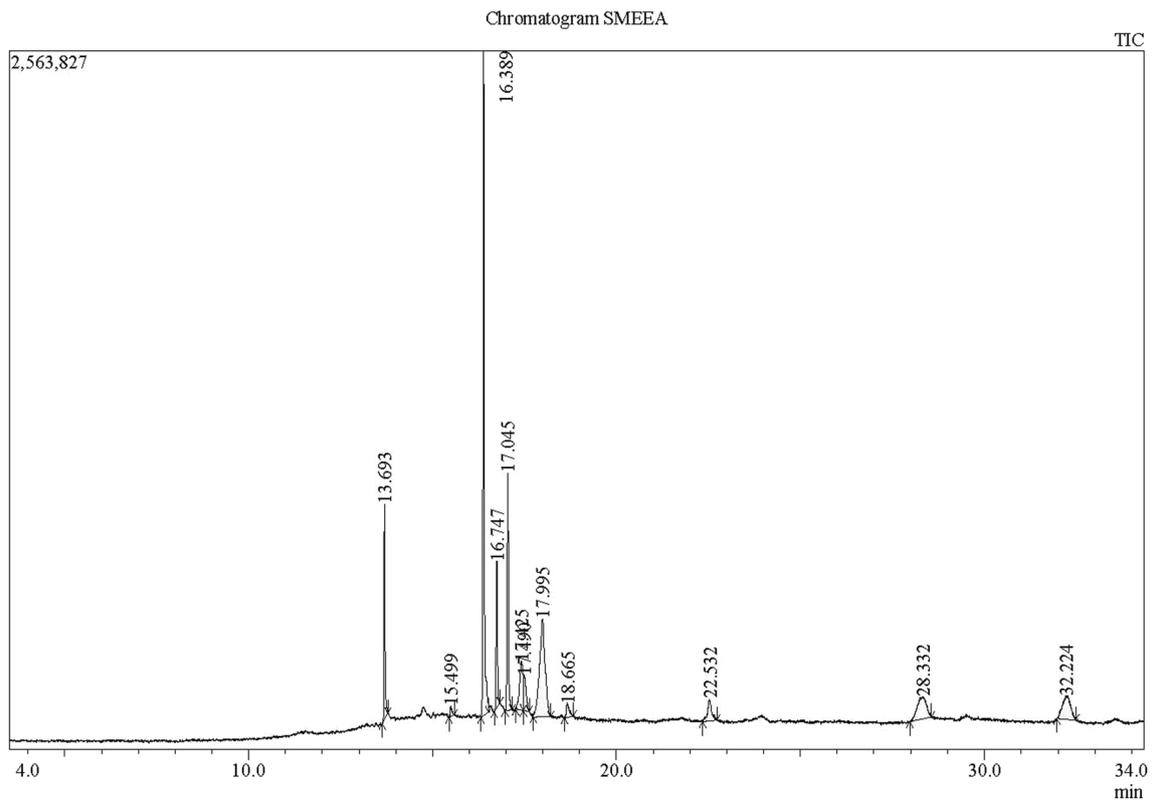


Fig. 8 GC-MS chromatogram for SMEEA

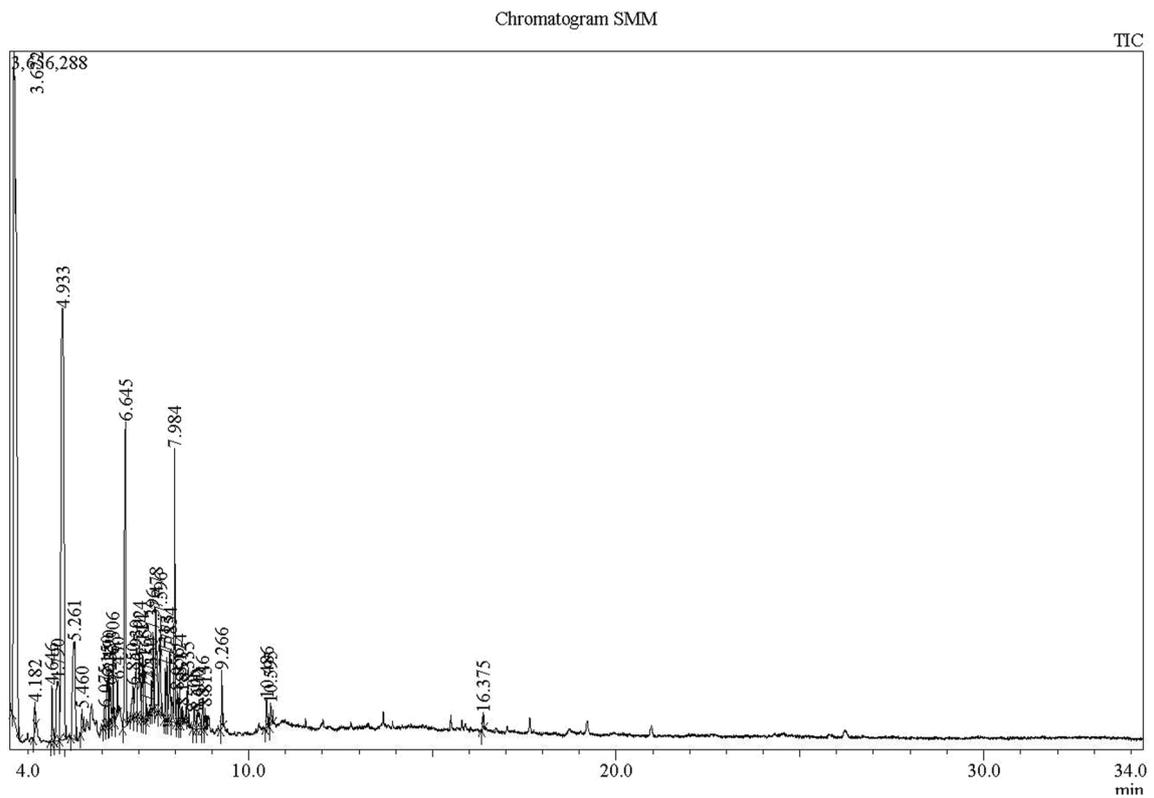


Fig. 9 GC-MS chromatogram for SMM

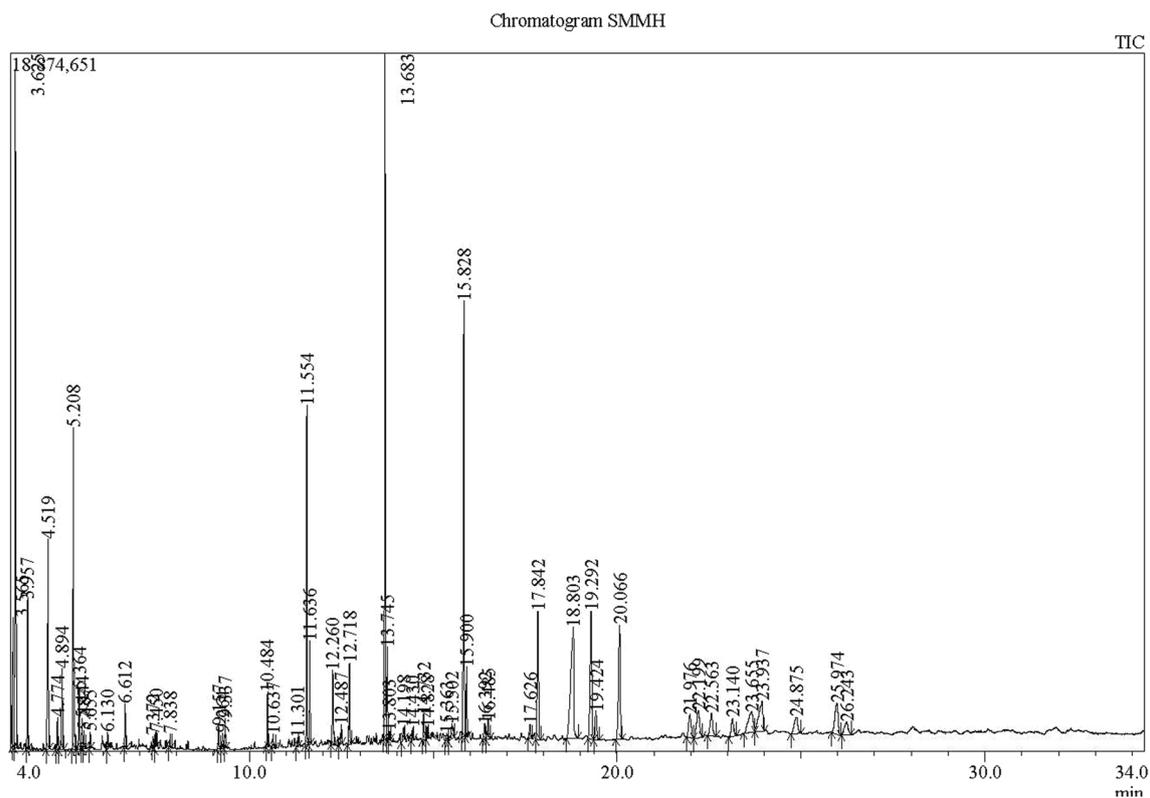


Fig. 10 GC-MS chromatogram for SMMH

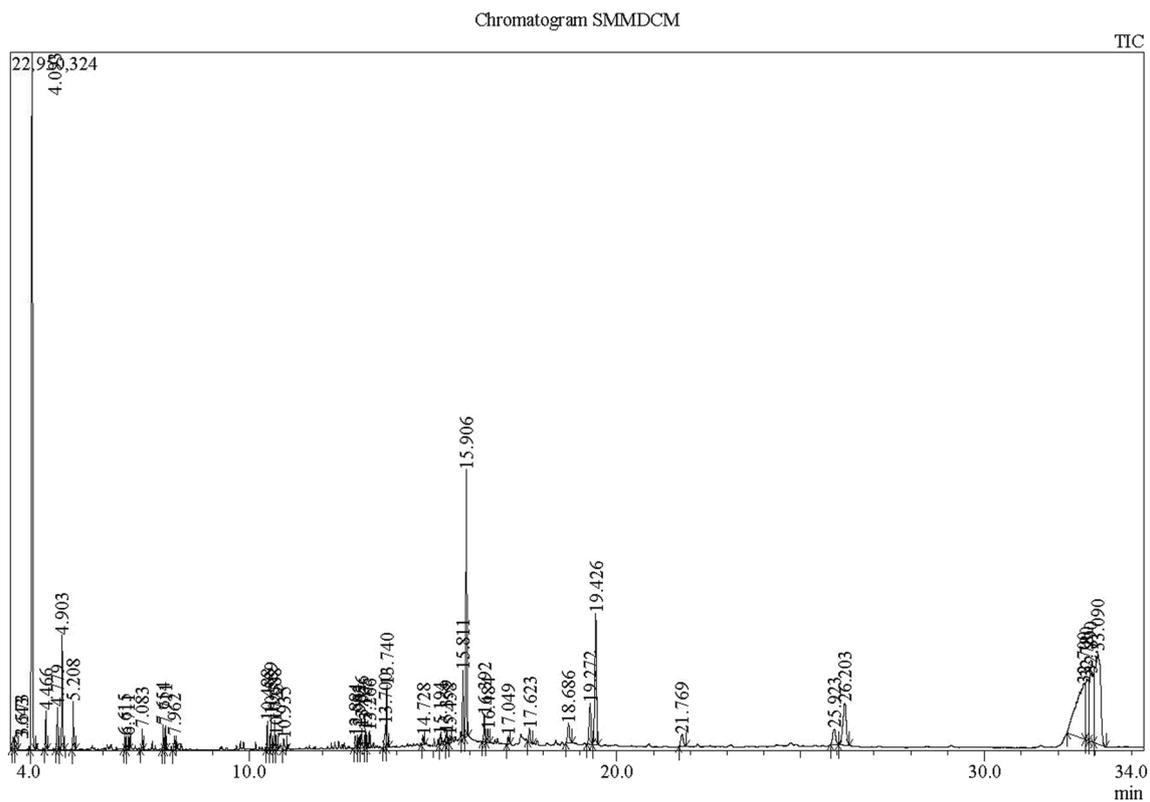


Fig. 11 GC-MS chromatogram for SMMDCM

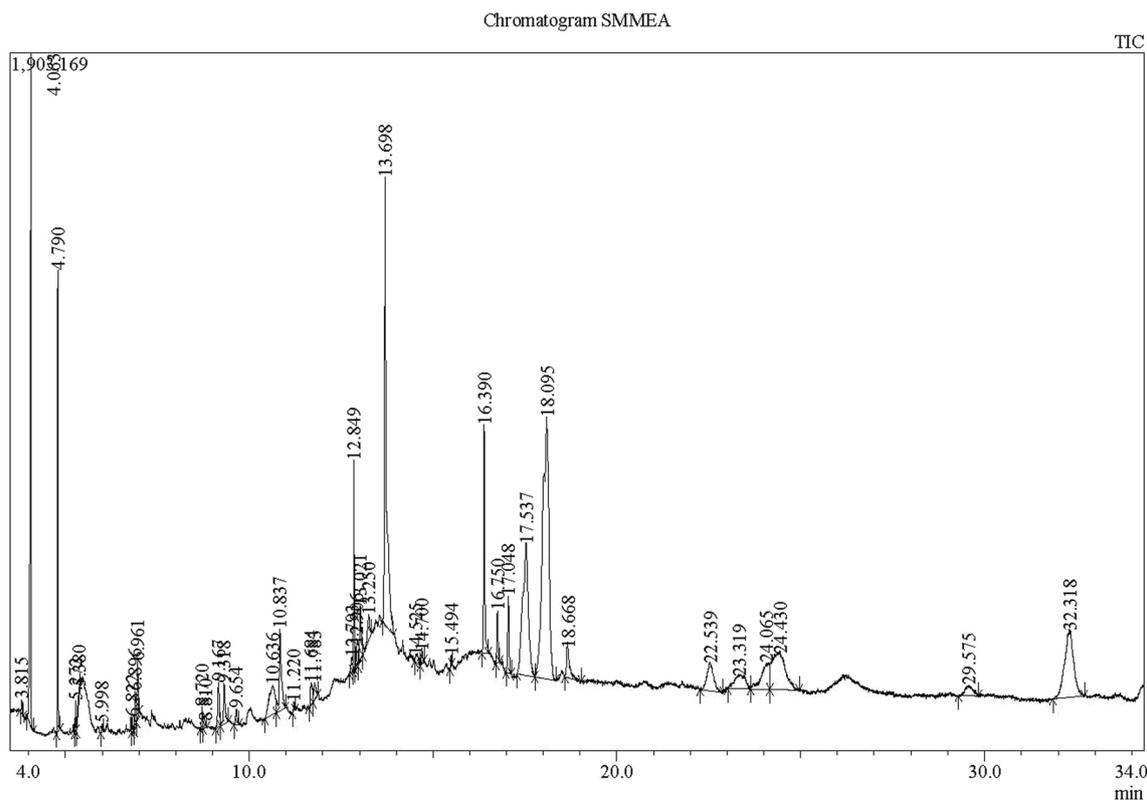


Fig. 12 GC–MS chromatogram for SMMEA

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## Declarations

**Ethical statement** This article does not contain any studies involving animals and human participants performed by any of the authors.

**Conflict of interest** Akwasi Boadu has no conflict of interest. Rajshekhar Karpoomath has no conflict of interest. Manimbulu Nlooto has no conflict of interest.

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