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Variation in chemical composition, insecticidal and antioxidant activities of essential oils from the leaves, stem barks, and roots of *Blighia unijugata* (Baker) and *B. sapida* (K. D. Koenig)

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ABSTRACT

Blighia unijugata (Baker) and *B. sapida* (K.D. Koenig) (Sapindaceae) are forest trees widespread in Tropical Africa. They are used in folk medicine for the treatment of rheumatism, cardiovascular diseases, yellow fever, dysentery, and epilepsy. The essential oils of the air-dried leaves, stem bark, and roots of the plants were extracted using the hydrodistillation method and were analyzed by Gas Chromatography-Mass Spectrometry (GC-MS). The antioxidant activity was determined using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical method using α-tocopherol and ascorbic acid as standards. The insecticidal activity of the oils was investigated by determining the percentage mortality of the grain insects (*Sitophilus zeamaise*). Results showed that the yield of the colorless oils (% w/w) from the leaves, stem bark, and roots of the plants were 0.91%, 0.42%, and 0.34% for *B. unijugata* and 0.67%, 0.32%, and 0.48% for *B. sapida*, respectively. The most abundant compounds in *B. unijugata* leaves were pentadecanoic acid (14-methyl-, methyl ester) (38.34%), carbonic acid, propyl-en-2-yl undecylpropyl ester (36.79%), and 9-octadecanoic acid *(Z)*-methyl ester (24.86%). The stem bark was rich in 1,3-dimethoxybenzene (79.01%) while the root contained limonene (20.51%), *trans*-13-octadecanoic acid (16.74 %), and *cis*-vaccenic acid (9.50%). The leaf essential oil of *B. sapida* had hexahydrofarnesyl acetone (21.43%), phytol (20.45%), geosmin (16.258%), and α-ionone (8.271%) as the major constituents. The stem bark had cholesterol (38.66%) as the major constituent while the root contained nonanal (18.09%) and 4-cyclopropyl carbonyl tetradecane (11.6%). The antioxidant analysis revealed that *B. unijugata* root had 83.96% inhibition at 100 mg/ml, while *B. sapida* stem had 79.73% inhibition, better than α-tocopherol. No significant insecticidal activity was observed in the essential oils of the plants against *S. zeamaise* except for *B. sapida* stem which showed 50% toxicity to the maize weevils. However, the two plants can be the source of antioxidant agents against oxidative stress and related diseases.

1. Introduction

Plants play a very important role in human society due to their secondary metabolites which serve as the source of medicine and food (Akerele, 1991; Farnsworth & Soejarto, 1991; Tapsell et al., 2006, Oloyede et al., 2019a, Onanuga & Oloyede, 2021). The biological function of essential oils in a variety of fields has made them very important natural products especially their use in food, cosmetics, deodorants, biocides, insecticides or pharmaceutical industries, personal care, and perfumery (Bakkali et al., 2008; Abdelouaheb & Amadou, 2012; Oloyede et al., 2021). Essential oils are obtained by hydrodistillation, enfleurage, solvent extraction, expression, and CO_2 hypercritical methods. Other modern techniques include microwave distillation, headspace trapping, solid phase micro-extraction (SPME), supercritical fluid extraction (SPE), and simultaneous distillation extraction (SDE). Therapeutic activities of essenti-

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al oils are due to the presence of compounds of terpene origin, olefinic double bonds, and functional groups such as hydroxyl and aldehydes (Bowles, 2003; Edris, 2007; Sell, 2010, Oloyede et al., 2019a, b).

The Sapindaceae (Soapberry) family are trees, shrubs, or lianas widely distributed in tropical to sub-tropical regions. Many of its members contain latex, milky sap, and mildly toxic saponins with soap-like qualities in the foliage and/or the seeds or roots. *Blighia* is a genus of three species of flowering plants (*B. sapida*, *B. unijugata*, and *B. welwitschii*) (Watson & Dalwitz, 2007). *B. welwitschii* however is not available in Nigeria. *B. unijugata* is a shrub that is small to medium-sized, but occasionally grows up to 30 to 35 m tall (Bekele-Tesemma & Tengnäs, 2007; Oderinde et al., 2008) (Figure 1). In some African countries, the leaves are used as vegetables to prepare meals. Various parts of the tree are known to have anti-malarial, sedative, and analgesic properties, for the treatment of rheumatism, kidney pain, and stiffness. Bark pulp is applied as enema or bark decoction is taken to treat fever, and as purgative (Burkill, 2000; Hyde et al., 2002). The wood of *B. unijugata* is commonly used for light construction, furniture, and charcoal production (Katende et al., 1995). Macroscopical investigation showed the stem bark outer layer is greyish and, the inner layer is pale reddish brown, with a disagreeable odor, while microscopical screening revealed the presence of starch grains, trichomes, and sclerenchyma and chemomicroscopic result revealed the presence of lignin, starch, calcium oxalate, cellulose, stone cells (Osuala, 2020). Significant inhibitory activity against the pathogenic microorganisms was observed when the nutritional elements, comparative study of the antimicrobial and cytotoxicity of the root, stem bark, and leaf essential oils, and crude extract of *B. unijugata* was investigated (Oderinde et al., 2008; Adewuyi, et al., 2009). Phytochemical screening showed the presence of polyphenols, polyterpenes, and flavonoids while extract of the fruits and butanol fraction from the leaves of *B. unijugata* was reported not to be toxic in animal studies (Aquaisua et al., 2011; Bléyéré et al., 2013; Osuala, 2020). Hydrogen peroxide and DPPH free radical scavenging activities of leaf, stem bark, root, flower, and fruit of *B. unijugata* was reported by Ajiboye et al. (2017a, b) and the essential oil compositions were reported (Dorcas et al., 2017) indicating the presence of terpenes.

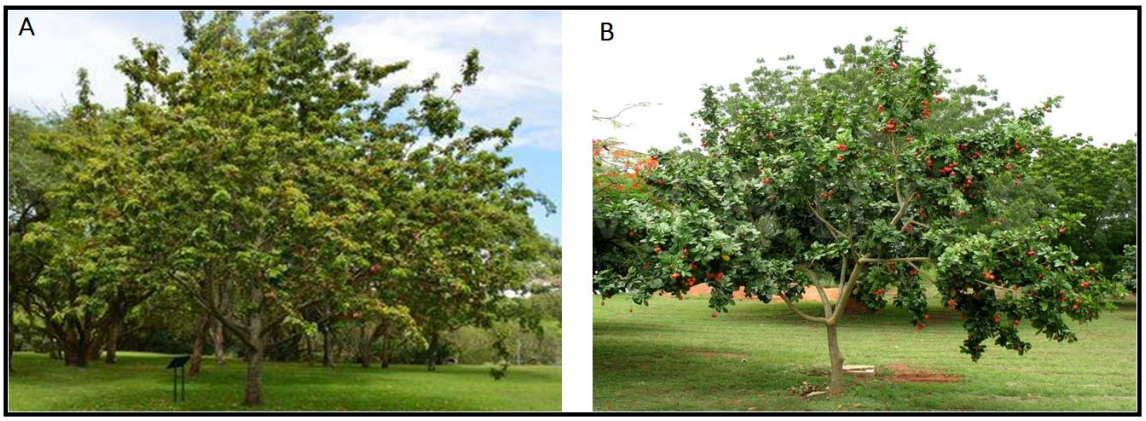

Figure 1. *B. unijugata* (A) and *B. sapida* (B)

B. sapida is a woody perennial multipurpose fruit tree species native to the Guinean forests of West Africa (Orwa et al., 2009) (Figure 1). The fruit of *B. sapida* is edible when fully ripped (Moya, 2001). The fruit produces lather with water and is therefore used for laundering purposes in some West African countries. Crushed fruits are used as fish poison while various preparation and combination of the extract have been made for the treatment of diseases such as dysentery, epilepsy, and yellow fever (Gbolade, 2009; Hamzah et al., 2013). The plant has been reported to be effective against cold, pain, and emulsion properties (Elizabeth et al., 2012) and is used as an insecticidal agent. The extract of the flowers is used as cologne while the pulverized bark is mixed with grounded hot peppers and rubbed on the body as a stimulant. Capsules of the fruits have the property of producing saponins, which lather in water and are used for washing (Ekué et al., 2010). Alkaloids, saponins, cardiac glycosides, reducing sugar, carbohydrates, flavonoids, phenol, and tannin were found in *B. sapida* fruits and root bark (Oyeleke, et al., 2013; Dossou et al., 2014). Aqueous extract of *B. sapida* decreased blood glucose levels of rats in a separate experiment and was the most potent inhibitor of α-amylase while ethanolic extract inhibited α-glucosidase most effectively (Howélé et al., 2010; Kazeem et al., 2013).

These two plants are sometimes used interchangeably and previous work done only focused on *B. unijugata*. Therefore, the main objective of this study is to compare the composition of the essential oil extracted from the leaves, roots, and stem bark of *B. unijugata* and *B. sapida* and to investigate their antioxidants [using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical method] and insecticidal potential using the maize pest (*Sitophilus zeamaise*) method (Oloyede et al., 2019a, b).

2. Materials and methods

2.1. Materials

2.1.1. Plant collection and identification

Fresh leaves, roots, and stem bark of *B. unijugata* and *B. sapida* were collected at the Botanical Garden and identified at the Herbarium, Department of Botany, University of Ibadan, Nigeria in January 2016. The samples were air-dried, pulverized, and placed in air-tight polythene bags before extraction to prevent any loss of volatile components.

2.1.2. Reagents

Hexane, methanol, ascorbic acid, and butylated hydroxyanisole (BHA) were purchased from BDH chemicals. 2,2-Diphenyl-1-picrylhydrazyl (DPPH) were purchased from Sigma-Aldrich.

2.1.3. Equipment/Apparatus

Hydro-distillation flask, Clevenger apparatus, glass condenser, heating mantle, electronic weighing balance (OHAUS), oven (Carbolite), syringes, sample bottles, GC-MS (Agilent Technologies, Model-7890A) gas chromatograph, and UV spectrophotometer (Unico1200 & Perkin Elmer Lambda 25 model, UK).

2.2. Methods

2.2.1. Extraction of essential oil

Hydro-distillation method using the Clevenger apparatus was used to extract oils. 300 g of each of the pulverized samples was weighed and placed in a 5 liter round bottom flask fitted with a heating mantle and water was added until the sample was fully immersed. The extraction process was carried out for 3 hours at a regulated temperature according to the European Pharmacopoeia (1996) specification. The volatile oil trapped in 1.0 ml of hexane was carefully collected using a syringe and placed in a sample vial. The weight of the oil was recorded and stored in the refrigerator until further analysis (Burt, 2004; Oloyede & Egbewole, 2014).

2.2.2. Gas chromatography-mass spectrometry (GC-MS) analysis

The essential oils were analyzed using GC-MS (Agilent Technologies, Model-7890A) gas chromatograph, coupled with a 5975C mass spectrometer (Agilent Technology). The gas chromatograph capillary column type was an HP-5MS, with a column length of 30 m; internal diameter of 0.320 mm, and film thickness of 0.25 µm. The carrier gas used was helium at a constant flow rate of 1.4123 ml/min and an average velocity of 43.311 cm/sec, while the pressure was put at 1.5 psi. Temperature programming started with an initial column temperature set at 80 °C for 2 mins and was increased to 240 °C at the rate of 10 °C/min. The volume of the sample injected was 1µl (Cronin & Caplan, 1987; Baser & Buchbauer, 2015; Onocha et al., 2016).

2.2.3. Identification of components

Flame ionization detector (FID) set at a temperature of 300 °C was used to identify the constituents and their percentage compositions obtained from electronic integration measurement. The peak numbers and relative percentages of the characterized compounds were recorded while the individual components of the oil were identified based on their retention indices determined with reference to *n*-alkanes and by comparison of their mass spectra fragmentation pattern (NIST0.8 L Database/Chem. Station System) with previously reported data (Lawless, 2013; Adams 2007; Las Heras et al., 2003). The peak numbers and relative percentages of the characterized components are given in Tables 1 and 2.

2.2.4. Insecticidal activity assay

Maize weevil insects, *S. zeamais*, that were used in this study were cultured at the Department of Crop Protection and Environmental Biology, University of Ibadan, Nigeria. A standard culture was maintained for 4 weeks without exposure to any insecticide. Following the methods of Karpouhtsis et al. (1998) and Isman (2000) with little modification, six insects (three males and three females) were used for each assay. The insects were placed in air penetrating containers containing 10 g of maize to avoid mortality by suffocation. 0.2 ml of test samples, *B. unijugata* leaves (BUL), *B. unijugata* stem (BUS), *B. unijugata* roots (BUR), *B. sapida* leaves (BSL), *B. sapida* stem (BSS), *B. sapida* roots (BSR), were applied onto a piece of sterilized Whatman filter paper (No: 1) and introduced into the different containers to determine its toxicity against the insects either via inhalation or contact. A control experiment of the same number of insects, but without test samples, was also performed. Three different concentrations of the essential oils, 25%-100% were prepared by serial dilution with hexane. Analysis was done in triplicate. Insects were monitored at 12-72 hours. Percentage (%) mortality of insects was calculated using the formula given below:

$$Corrected\ mortality\ (\%) = 1 - \frac{n\ in\ Co\ before\ treatment\ x\ n\ in\ T\ after\ treatment}{n\ in\ Co\ after\ treatment\ \times\ n\ in\ T\ before\ treatment} x\ 100$$

Where 'n' is insect population, 'T' is treated diet with sample, and 'Co' is control.

2.2.5. Antioxidant screening

The antioxidant activity of the essential oils of the samples was determined using DPPH radical scavenging method. Various concentrations (25 mg/ml – 100 mg/ml) of the test sample (1 ml of each essential oil) were mixed with 0.2 mm methanol-DPPH solution (2.0 ml), prepared by dissolving 7.81 mg of DPPH in 100 ml of methanol. The mixture was shaken vigorously and left to incubate for 30 minutes in the dark at room temperature and the absorbance was then measured at 517 nm and recorded using GS UV-12, UV-VIS spectrophotometer. In its radical form, DPPH absorbs, but upon reduction by an antioxidant species, its absorption reduces. A blank experiment was carried out applying the same procedure without the test sample (DPPH/methanol) and the absorbance was recorded. Ascorbic acid (vitamin C) and α-tocopherol (vitamin E) were used as standard.

2.2.6. Statistical analysis for determination of percentage inhibition in the DPPH analysis

Determination of the free radical scavenging activity in the DPPH analysis of each test oil solution was calculated as percentage inhibition using the equation given below (Ayoola et al., 2008; Onocha et al., 2016):

$$Inhibition\ (\%) = 1 - \frac{Ablank - Asample}{Ablank} x\ 100$$

This was statistically determined from the absorbance measurement of samples. Graph showing % inhibition against concentration (mg/ml) was shown as a column chart in Figure 2.

3. Results and discussion

The percentage yields (w/w) of the colourless essential oils were as follows: BUL: 0.91%, BUS: 0.42%, BUR: 0.34%, BSL: 0.67%, BSS: 0.48%, and BSR: 0.32%.

Three non-terpenoid constituents yielding a total of 99.99% identified in *B. unijugata* leaf essential oil (Table 1) were pentadecanoic acid, 14-methyl, methyl ester (38.34%), carbonic acid, prop-1-en-2-yl undecylpropyl ester (36.79%), and 9-octadecanoic acid *(Z)*-methyl ester (24.86%). Twelve constituents were identified in the root essential oil of *B. unijugata* representing a total of 99.94%, comprising one monoterpene (limonene), and eleven non-terpenoids, mainly acids, esters, and hydrocarbon. The major constituents were: limonene (20.51%), *trans*-13-octadecanoic acid (16.74%), *cis*-vaccenic acid (9.50%), 1,2-benzene dicarboxylic acid, butyl-2-methyl propyl ester (9.74%), and 2,5-norbornanediol (8.36%). Other constituents present were 3-methylheptadecane (7.42%), *cis*-13-octadecanoic acid, *(E)* (6.40%), hexadecanoic acid (5.93%), heptadecanolide (4.0%), octadec-9-enoic acid (3.21%), 9-

octadecanoic acid *(Z)*-2,3-dihydroxy propylester (2.96%) and 9-octadecanoic acid *(E)* (0.32%). Fifteen compounds were detected in the stem bark oil of *B. unijugata* with a non-terpenoid 1,3-dimethoxybenzene as the major constituent (79.01%), however, longifolene (4.22%), 4-penta-1-one, 1-(1H, imidazole-2-yl)-4-isopropenyl cyclo hexanone (2.54%), α-ionone (2.40%), phenol, 4-methoxy-3-methyl (2.20%), *trans*-β-ionone (1.51%), benzothiazole, 3-methyl (1.26%), 2,5-nornanediol (1.18%), hexahydro farnesyl acetone (1.13%), tricosane (0.83%), 1,16-hexadecanediol (0.81%), 5-*t*-butyl-4-methylimidazole (0.78%), 2-piperidine, N-(4-bromo, *n*-butyl) (0.58%), farnesyl acetone (0.57%), and octadecane,1-bromo (0.48%) were detected in trace amount. It was observed that only 2,5-norbornanediol was common to both the root and stem bark essential oil of *B. unijugata* (Table 1). Geranyl acetone is common to root and leaves of *B. sapida*, on the other hand, hexahydrofarnesyl acetone, farnesyl acetone, and cembrene A were found in stem and leaves, whereas the oleic acid was detected in stem and root.

Table 1. Chemical constituents of leaves, stem, and roots of *B. unijugata*

No	RT (min)	Constituents	Composition (%)*		
			BUL	BUS	BUR
1	4.34	1,16-Hexadecanediol	-	0.81	-
2	5.67	Dimethylresorcinol	-	79.01	-
3	11.17	α-Ionone	-	2.40	-
4	11.63	5-*t*-Butyl-4-methylimidazole	-	0.78	-
5	11.73	4-Methoxy-3-methyl phenol	-	2.20	-
6	12.41	β-Ionone	-	1.51	-
7	15.39	2,5-Norbornanediol	-	1.18	8.36
8	15.95	Longifolene	-	4.22	-
9	16.57	Octadecane,1-bromo	-	0.48	-
10	17.85	Limonene	-	-	20.51
11	17.58	4-penta-1-one,1-(1H-Imidazol-2-yl)-4-isopropenylcyclohexanone	-	2.54	-
12	19.30	Hexahydrofarnesyl acetone	-	1.13	-
13	19.36	Carbonic acid, prop-1-en-2-yl undecyl ester	36.79	-	-
14	20.63	Farnesyl acetone	-	0.57	-
15	20.76	Methyl isohexadecanoate	38.34	-	-
16	21.27	Heptadecane, 3-methyl	-	-	7.42
17	21.43	1,2-Benzenedicarboxylic acid, butyl-2-methyl propyl ester	-	-	9.14
18	21.88	Hexadecanoic acid	-	-	5.93
19	23.38	Octadec-9-enoic acid	-	-	3.21
20	23.63	Oleic acid, methyl ester	24.86	-	-
21	23.80	Heptadecanolide	-	-	4.00
22	24.03	9-Octadecanoic acid *(Z)*-2,3-dihydroxypropyl ester	-	-	2.96
23	24.67	*cis*-Vaccenic acid	-	-	9.50
24	25.05	*cis*-13-Octadecenoic acid	-	-	6.40
25	25.23	9-Octadecanoic acid, *(E)*	-	-	0.32
26	26.52	2-Piperidinone, N-(4-bromo *n*-butyl)	-	0.58	-
27	27.88	*trans*-13-Octadecenoic acid	-	-	16.74
28	27.95	Docosane	-	0.25	-
29	29.26	Tricosane	-	0.83	-
30	30.26	Benzothiazole 3-methyl		1.26	
		Total	99.99	99.75	94.49

*Percentages were calculated from the flame ionization detection data. RT: Retention time on HP-5MS column, BUL: *B. unijugata* leaves, BUS: *B. unijugata* stem, BUR: *B. unijugata* roots.

Seventeen components were identified in the leaf essential oil of *B. sapida* and contained hexahydrofarnesyl acetone (21.43%), phytol (20.45%), geosmin (16.25%), and α-ionone (8.27%) as the major constituents, while β-ionone (4.41%), β-eudesmol (2.20%), aromadendrene (1.36%), coniferyl aldehyde and *n*-propyl ether (2.94%) were also detected. Twenty-one compounds were observed in *B. sapida* stem essential oil, with the major constituent being cholesterol (38.66%). The root with twenty two compounds had nonanal (18.09%), 4-cyclopropyl carbonyl tetradecane (11.60%), 3-methyl-4-(phenylthio)-2-prop-2-enyl-2,5-dihydro thiophene-1,1-dioxide (11.49%) as the principal components, while *n*-hexanoic acid (9.60%), ascorbyl palmitate (1.35%), (2)-6-octadecanoic acid (0.87%), 6-methoxy-2,7,8-trimethyl-2-(4,8,12-trimethyl decyl) chroman (0.52%), stearin (0.34%), and oleic acid (0.26%) were also found in the oil. Cembrene A, hexahydrofarnesyl acetone, and farnesyl acetone were common to both *B. sapida* stem and leaf, while oleic acid was present in *B. sapida* stem and root and was also detected in *B. unijugata* (Table 2).

Mass fragmentation of the major and common components in *B. unijugata* and *B. sapida* leaves, stem bark, and root were also presented in Table 3.

3.1. Insecticidal activity

The use of natural products from plants is considered a useful alternative for the control of stored grain pests (Ho et al., 1996; Isman, 2000). Essential oils have also been employed as green pesticides (Obeng-Ofori & Reichmuth, 1999; Koul et al., 2008). The result of the insecticidal analysis showed that only *B. unijugata* leaves (BSL) and *B. sapida* stem (BSS) showed 50% and 33.3% toxicity to the maize weevils, respectively (Table 4) and justify their use in ethnomedicine as pesticides. The low insecticidal activity observed may have been due to the absence of certain phytochemicals such as α-pinene, β-pinene, camphor, linalool, and other related compounds in the plant essential oils which were reported to be responsible for the toxicity of stored grain pests (Huang et al., 1998; Lee et al., 2001). The extracts may however have pronounced activity.

3.2. Antioxidant activity

The essential oils scavenged free radicals in the antioxidant screening assay, with comparable activity to the standards, ascorbic acid, and α-tocopherol. *B. unijugata* root (83.96%) showed the highest activity and better activity than α-tocopherol (75.08%) but lower than ascorbic acid (91.60%) at 100 mg/ml. BUL and BUS exhibited 79.19% and 77.28% inhibition at the same concentration, respectively. The activity of *B. sapida* stem essential oil (79.73%) was

higher than that of the root (79.61%) at 100 mg/ml. The leaf essential oil gave the least antioxidant activity (74.96%). These values are comparable to that of one of the standard, α-tocopherol (75.08%) but less than that of ascorbic acid (91.60%). The antioxidant activities of the oils were concentration-dependent (Figure 2). The results indicated that the essential oils are good scavengers of free radicals as the reduction in absorbance values were observed in all the oil samples after incubation in DPPH.

Table 2. Chemical composition of the essential oil of leaves, stem, and roots of *B. sapida*

No	RT (min)	Constituents	Composition (%)*		
			BSL	BSS	BSR
1	3.464	Benzene acetyldehyde	-	1.93	-
2	3.514	2,4,4-Trimethyl-2-hexen-1-ol	2.21	-	-
3	3.802	Octan-1-ol	-	-	3.82
4	4.322	Nonanal	-	-	18.09
5	4.328	1,10-Decanediol	-	4.71	-
6	4.335	2-Methyl cyclohexanol	2.44	-	-
7	5.427	*(E)*-Non-2-enal	-	-	3.89
8	5.724	Veratrole	-	2.98	-
9	5.701	Butylcyclopentane	-	-	2.89
10	5.902	*(2E,4E)*-3,7-Dimethyl-2,4-octadiene	-	1.74	-
11	7.304	Acetaldehyde, (3,3-dimethylcyclohexylidene)	1.17	-	-
12	8.877	*(E,E)*-2,4-Decadienal	-	-	4.33
13	10.649	Geosmin	16.25	-	-
14	11.174	α-Ionone	8.26	-	-
15	11.715	Geranyl acetone	3.14	-	3.07
16	11.733	3,5-Dimethyl-1-adamantanol	-	3.81	-
17	12.373	Eremophylene	-	3.66	-
18	12.417	β-Ionone	4.41	-	-
19	13.878	*cis*-Chrysanthemol	-	1.54	-
20	14.82	Caryophyllene oxide	-	-	1.47
21	15.546	β-Eudesmol	2.20	-	-
22	16.157	Aromandendrene	1.35	-	-
23	16.558	Coniferyl aldehyde, *n*-propyl ether	2.94	-	-
24	17.575	2-(1H-Imidazole-5-yl)bicyclo[1,1,1]pentan-2-ol	-	1.51	-
25	19.297	Hexahydrofarnesyl acetone	21.44	2.03	-
26	20.607	Farnesyl acetone	7.01	1.34	-
27	21.101	Isophytol	1.21	-	-
28	21.27	Cembrene A	1.19	3.03	-
29	21.803	*n*-Hexanoic acid	-	-	9.60
30	21.929	Ascorbyl palmitate	-	-	1.35
31	21.998	1-Hexacosene	-	-	2.03
32	22.106	*(Z)*-9-Tricosene	-	-	1.73
33	22.112	Verticilla-4(20),7,11-triene	-	2.59	-
34	22.232	1,3,6,10-Cyclo-tetra decatetraene-3,7,11-trimethyl	-	1.99	-
35	23.194	2-Octylcyclopropaneoctanal	-	-	3.26
36	23.291	Stearin	-	-	0.34
37	23.819	Phytol	20.45	-	-
38	23.823	*(E)*-3-Eicosene	-	2.15	-
39	24.287	Linoleic acid	-	0.76	-
40	25.03	Petroselinic acid	-	0.13	-
41	25.242	Dodecyl myristate	-	0.19	-
42	26.501	Oleic acid	-	1.30	0.27
43	27.348	*(E)*-Octadecanoic acid	-	-	4.79
44	27.399	(2)-6-Octadecanoic acid	-	-	0.88
45	27.954	Vinyl stearyl ether	-	0.60	-
46	27.956	*trans*-13-Octadecenoic acid	2.41	-	-
47	28.652	Squalene	-	-	10.75
48	29.35	2-Hydroxy-cyclopentadecanone	-	1.23	-
49	29.364	Batilol	1.83	-	-
50	29.802	3-Methyl-4-(phenyl thio)-2-prop-2-enyl-2,5-dihydrothiophene-1,1-dioxide	-	-	11.5
51	30.93	Cholesterol	-	38.66	-
52	31.05	δ-Tocopherol	-	-	3.09
53	31.073	*O*-Acetyl-δ-tocopherol	-	-	0.734
54	31.302	4-Cyclo propyl carbonyl tetradecane	-	-	11.6
55	31.997	6-Methoxy-2,7,8-trimethyl-2-(4,8,12-trimethyl decyl)chroman	-	-	0.522
		Total	100	75.88	87.15

*Percentages were calculated from the flame ionization detection data. RT: Retention time on HP-5MS column, BSL: *B. spida* leaves, BSS: *B. spida* stem, BSR: *B. unijugata* roots.

The presence of oxygenated terpenoids has been reported to greatly enhance the free radical scavenging activity of plant metabolites (Fang et al., 2002; Oloyede & Egbewole, 2014; Ajiboye et al., 2017a, b; Oloyede et al, 2019a, b) and may also be responsible for the observed activity in these plants.

4. Conclusions

Comparative evaluation of the chemical composition of the essential oils from the leaves, stem bark, and roots of *B. unijugata* and *B. sapida* showed that they were rich in oxygenated terpenoids and non-terpenoids with few monoterpenes such as limonene (20.51%) in *B. unijugata*. 2, 5-norbornanediol was found to be present in both the root and stem bark essential oils of the plant. Cembrene A, hexahydrofarnesyl acetone, and farnesyl acetone were found in both *B. sapida* stem and leaf. Oleic acid was also found in *B. unijugata* and *B. sapida* stem and root. Toxic effects against the maize weevil, *S. zeamaise*, and significant free radical scavenging when compared to standards of ascorbic acid and α-tocopherol justify the use of the plants in traditional medicine.

Table 3. Mass fragmentation of the major and common components in *B. unijugata* and *B. sapida* leaves, stem bark, and root

No	Major/Common constituents	Molecular formula	Molecular weight (g/mol)	Mass fragmentation *(m/z)* values
1	*trans*-13-Octadecenoic acid	$C_{18}H_{34}O_2$	282	282 (M^+) ($C_{18}H_{34}O_2)^+$, 264, 246, 235, 222, 211, 196, 180, 165, 151, 137, 123, 111, 97, 83, 69, 60, 55, 41, 29, 15
2	Limonene	$C_{10}H_{16}$	136	136 (M^+), ($C_{10}H_{16})^+$, 121, 107, 93, 79, 68, 55, 53, 41, 39, 29
3	Oleic acid, methyl ester	$C_{19}H_{36}O_2$	296	296 (M^+) ($C_{19}H_{36}O_2)^+$, 278, 264, 246, 207, 193, 180, 152, 137, 129, 111, 97, 87, 83, 74, 69, 59, 55, 41, 33, 29, 15
4	Carbonic acid, prop-1-en-2-yl undecyl ester	$C_{15}H_{28}O_3$	256	256 (M^+) ($C_{15}H_{28}O_3)^+$, 199, 154, 126, 111, 97, 85, 71, 67, 57, 43, 29
5	Methyl isohexadecanoate	$C_{17}H_{34}O_2$	270	270 (M^+) ($C_{17}H_{34}O_2)^+$, 239, 227, 213, 199, 185, 171, 157, 143, 129, 111, 97, 87, 74, 69, 59, 55, 43, 29
6	Dimethyl resorcinol	$C_8H_{10}O_2$	138	138 (M^+) ($C_8H_{10}O_2)^+$, 123, 109, 95, 78, 65, 57, 52 43, 29
7	Cholesterol	$C_{27}H_{40}O$	386	386 (M^+) ($C_{27}H_{40}O)^+$, 386, 353, 301, 275, 255, 247, 231, 173, 159, 145, 133, 119, 107, 95, 81, 69, 57, 43
8	Hexahydrofarnesyl acetone*	$C_{18}H_{36}O$	268	250 (M^+) ($C_{18}H_{36}O)^+$, 250, 210, 124, 109, 95, 58, 43, 29
9	Phytol	$C_{20}H_{40}O$	296	296 (M^+) ($C_{20}H_{40}O)^+$, 137, 123, 111, 95, 81, 71, 57, 43, 29
10	Nonanal	$C_9H_{18}O$	142	142 (M^+) ($C_9H_{18}O)^+$,114, 98, 95, 82, 70, 57, 41, 39, 29
11	Geosmin	$C_{12}H_{22}O$	182	182 (M^+)($C_{12}H_{22}O)^+$,125, 112, 97, 83, 71, 69, 57, 69, 57, 56, 53, 43, 39
12	2,5-Norbornanediol*	$C_7H_{12}O_2$	128	128 (M^+) ($C_7H_{12}O_2)^+$, 126, 110 (100), 95, 92, 88, 86, 84, 83, 82, 81, 79, 73, 71, 68, 67, 65, 57, 54, 49, 45, 43, 41
13	Geranyl acetone*	$C_{13}H_{22}O$	194	194 (M^+) ($C_{13}H_{22}O)^+$, 151, 136, 125, 121, 109, 95, 83, 79, 69, 43, 41,39, 27
14	Farnesyl acetone*	$C_{18}H_{30}O$	262	262 (M^+) ($C_{18}H_{30}O)^+$, 138, 125, 121, 107, 95, 93, 81, 69, 43,41, 29
15	Cembrene A*	$C_{20}H_{32}$	272	272 (M^+) ($C_{20}H_{32})^+$, 257, 229, 187, 173, 159, 145, 133, 119, 105, 91, 79, 67, 55
16	Oleic acid*	$C_{18}H_{34}O_2$	282	282 (M^+) ($C_{18}H_{34}O_2)^+$, 264, 151, 125, 131, 111, 97, 83, 69, 57, 55, 43, 41, 39, 29, 27

*Compounds that are common to the leaf, stem bark, and root essential oils. The individual components of the oil were identified based on their retention indices determined with reference to *n*-alkanes and by comparison of their mass spectra fragmentation pattern (NIST0.8 L Database/Chem. Station System) with previously reported data. Structures were obtained from the literature (Lawless, 2013; Adams, 2007; Las Heras et al., 2003).

Table 4. Insecticidal activity of *B.unijugata* and *B. sapida* leaves, stem, and root essential oil on *S. zeamaise**

T (hr)	Insects killed at different concentrations (%)																		
	BUL			BUS			BUR			BSL			BSS			BSR			C
	25	50	100	25	50	100	25	50	100	25	50	100	25	50	100	25	50	100	
3	-	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
12	-	-	-	-	-	-	1	-	-	-	-	-	-	-	-	-	-	1	-
18	2	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-	-	-
24	-	-	-	-	-	-	-	-	-	-	1	-	1	-	1	-	-	-	-
36	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
48	-	-	-	-	-	-	-		-	-	-	-	-	1	-	-	-	-	-
72	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-
No-i	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	-
No-d	3	1	0	0	0	0	0	0	0	0	1	1	1	1	2	0	0	1	-
% M	50.0	16.7	0	0	0	0	16.7	0	0	0	16.7	16.7	16.7	16.7	33.3	0	0	16.7	-

*BUL: *B. unijugata* leaves. BUS: *B. unijugata* stem bark, BUR: *B. unijugata* roots, BSL: *B. sapida* leaves, BSS: *B. sapida* stem bark, BSR: *B. sapida* roots, T: Time (hours), No-i = Number of insects used, No-d = Number of dead insects, % M = Percentage mortality.

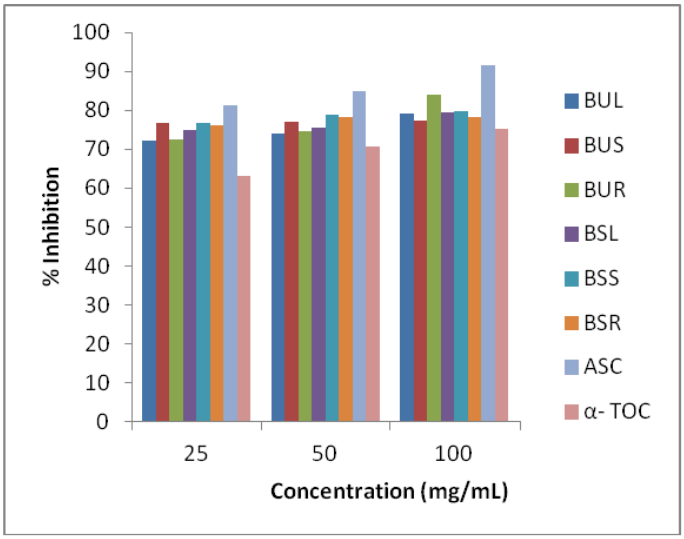

Figure 4. Percentage inhibition of DPPH free radical scavenging activities of BUL (*B. unijugata* leaves), BUS (*B. unijugata* stem bark), BUR (*B. unijugata* roots), BSL (*B. sapida* leaves), BSS (*B. sapida* stem bark) and BSR (*B. sapida* roots), standards ascorbic (ASC) and α-tocopherol (α-TOC).

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Conflict of interest

The authors confirm that there are no known conflicts of interest.

Statement of ethics

In this study, no method requiring the permission of the "Ethics Committee" was used.

Availability of data and materials

All data generated or analyzed during this study are included in this published article.

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Supplementary File

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