Chemical Composition and Antioxidant Activities of the Essential Oils of *Eugenia caryophyllata* from Northern Buru Island, Moluccas

Hanoch Julianus Sohilait,[a,*] Healthy Kainama,[b,*] and Martha Kaihena [c]

The essential oils of clove species *Eugenia caryophyllata* (COs) from northern Buru Island were obtained by steam distillation. The chemical components were characterized by gas chromatography (GC-FID) and GC-mass spectrophotometry (MS). In total, five components were identified in COs of leaf, four components were identified in COs of bud, and two components were identified in COs of stem. The main constituents are eugenol (77.1% to 78.8%) and β-caryophyllene (17.0% to 19.6%) in COs of leaf. The main components are eugenol (76.1% to 87.7%) and eugenyl acetate (6.0% to18.9%) in COs of bud and the only main component was eugenol (94.2% to 97.6%) in COs of stem from Waihani (COsWh), Ilath (COsI) and Waimoli (COsWm). The antioxidant activity COs of leaf, bud, and stem was identified using DPPH and ABTS assay. Results showed that COsI of bud had higher ABTS free radical scavenging (IC_{50} value: 0.09 ± 0.61 µg/mL) than the eight samples in the study. When this activity was compared with synthesis antioxidant of BHT (IC_{50} value: 0.83 ± 0.54 µg/mL) and AA (IC_{50} value: 0.81 ± 1.64 µg/mL), the COs in three locations show potential of being used as an antioxidant.

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Contact information: a: Department of Chemistry, Faculty of Mathematic and Natural Science, Universitas Pattimura, Ambon, Indonesia; *b*: *Department of Chemistry Education, Faculty of Training and Science Education, Universitas Pattimura Ambon, Indonesia; c: Department of Biology, Faculty of Mathematic and Natural Science, Universitas Pattimura Ambon, Indonesia*;
** Corresponding authors*: *nokesohilait@yahoo.com*; *healt_kainama@yahoo.com*

INTRODUCTION

The clove tree, *Eugenia caryophyllata* Thunb. (Myrtaceae), grows naturally in Moluccas Islands, East Indonesia, and are cultivated in Tanzania, Madagascar, Sri Lanka, India, Malaysia, Brazil, Jamaica, and Guinea (Ozturk and Ozbek 2005). *Eugenia caryophyllata* (synonym: *Syzygium aromaticum*) commonly known as clove, is a medium size tree (8 to 12 m) from the Myrtaceae family native from the Moluccas islands in east Indonesia. For centuries, the trade of clove and the search for this valuable spice stimulated the economic development of this Asia region (Kamatou *et al*. 2012). Clove can be used in cooking, either whole or in ground form. The spice is used in Europa and Asia, and in Indonesia, it is used for cigarettes, known as "kreteks" (Alma *et al*. 2007). Furthermore, the essential oil of clove (clove oils/ COs) has been widely used as spice and is well known for its medicinal properties. *Eugenia caryophyllata* (Clove) is considered an important medicinal plant with a wide range of biological activities, such as anti-bacterial or anti-oxidant activities (Barakat 2014). Therefore, many researchers have been looking for scientific

evidence for the use of clove extracts and essential oil. Some evidence of antioxidant, analgesic, antipyretic, and sedative effects (Rojas *et al*. 2014) have been found and include, treatments for dengue fever and larvicidal issues (Lopes *et al*. 2020); antioxidant effects (Lee and Shibamoto 2001; Sohilait and Kainama 2019; Kaur *et al*. 2019; Alfikri *et al.* 2020) and anti-inflammatory effects (Han and Parker 2017).

Three types of essential oils are available from clove (*E. caryophyllata*): clove leaf oils, clove stem oil, and clove bud oil. Each has a different chemical composition and flavour. Clove leaf oils contain eugenol (81.06% to 86.04%) and β-caryophyllene (11.95% to 16.16%). Clove stem oils contain eugenol (97.20% to 98.83%), bud clove oil contain eugenol (81.13% to 84.44%) and eugenyl acetate (11.60% to 15.02%) (Sohilait 2015). The differences of the essential oil composition are affected by different environmental and genetic factors, chemotypes, and soil condition of the plants, which may influence the composition (Salleh and Ahmad 2016; Sohilait *et al*. 2021; Kainama *et al*. 2023).

Several studies reported that the amount of chemical composition contained in COs varied according to the location where it was grown. COs of bud in Turkey contained 18 compounds (Alma *et al*. 2007), CO of leaf contained 38 compounds and COs of bud in Bangladesh 31 compounds (Bhuyan *et al*. 2010). The chemical composition of COs of bud contained 10 compounds, COs leaves had 9 compounds and 10 compounds were found in COs stem in Madagascar. The COs of bud were 22 compounds, and COs of leaf contained 21 compounds in India (Srivastava *et al*. 2005). In addition, the free radical scavenging DPPH activity of COs of bud in India and Tunisia has been reported by Mahboubi *et al.* (2015) and Chaieb *et al*. (2007).

However, the chemical composition of COs bud, leaf, and stem from northern Buru Island, Moluccas-Indonesia and free radical scavenging activity of DPPH and ABTS have not been reported by various sources. This research is important so that it can provide information regarding the quality and quantity of the phytochemicals of essential oils of COs from the locations where *E. caryophyllata* is abundant. On the other hand, this information can also explain the free radical scavenging activity of DPPH and ABTS for COs containing five to seven compounds in these sites.

EXPERIMENTAL

Materials

Fresh cloves from the tree were collected December 2021 from Waihani, Ilath, and Waimoli Village, northern Buru Island, Moluccas and were deposited in the Organic Chemistry Laboratory, Universitas Pattimura, Ambon. The chemicals used in the study were anhydrous sodium sulfates p.a (E. Merck, Darmstadt, Germany), DPPH (2,2-diphenyl-1-picrylhydrazyl), ABTS (2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid diammonium salt), standard BHT(butylated hydroxytoluene), and AA (ascorbic acid). The tools used in this study were a set steam distillation, Gas Chromatograph GC-2010, Shimadzu, Kyoto, Japan, and GC-Mass Spectrophotometer (GC-MS QP-2010 Plus, Shimadzu, Kyoto, Japan).

Methods

Isolation of COs from leaf, bud and stem

To obtain clove essential oils (COs), dried plant materials from the leaf, bud, and stem of *Eugenia caryophyllata* (Table 1) were steam-distilled using a steam distiller for 6

h. The product of essential oil was then dried over anhydrous Na_2SO_4 and cool-stored in a brown bottle for further analysis. The essential oils yield of *Eugenia caryophyllata* was calculated by the method described by Taipabu *et al*. (2022), defined using Eq. 1:

$$\text{Yield (\%)} = \text{COs weight (g)/Sample weight (g) x 100} \quad (1)$$

Gas Chromatography Analysis

The COs was analyzed using a Shimadzu QP-2010, equipped with an FID and Rtx-5 using a fused silica capillary column (30 m × 0.25 mm ID, film thickness 1.0 µm). Oven temperature was set at 60 °C for 5 min, and heating was programmed from 60 to 180 °C at a rate of 20 °C for 5 min, and from 280 °C for 4 min, injector temperature was 270 °C; detector temperature was 280 °C; pressure of carrier nitrogen gas at inlet was 7 psi, split ratio 20:1, and the volume injection was 0.5 µL.

Gas Chromatography-Mass Spectrometry Analysis

The GC-MS analyses of COs were performed using Shimadzu QP-2010 Plus at 70 eV and 320 °C with auto sampler. The system was equipped with Rtx-5 using a fused silica capillary column (30 m x 0.25 mm ID, film thickness 1.0 µm). Oven temperature was set at 60 °C for 5 min, then program heated from 60 to 220 °C at a rate of 10 °C for 5 min, and 280 °C for 4 min, with the injector temperature set at 270 °C. The carrier gas used was helium at flow rate of 1.40 mL/min. The spectra were scanned from 40 to 600 *m/z*, at split ratio of 20, and ion source temperature of 225 °C. The spectrum of the unknown component was compared with that of known components stored in the Wiley 7 library and retention index (Adams 2007), name, molecular weight, and structure of the component of the test materials were ascertained.

DPPH and ABTS Radical Scavenging Activity

The DPPH and ABTS assays were performed according to the method described by Sohilait *et al*. (2019). The BHT and AA were used as positive controls. The percentage of radical scavenging activities was calculated as follows:

$$\text{Inhibition (\%)} = (\text{Absorbance}_{\text{control}} - \text{Absorbance}_{\text{sample}})/\text{Absorbance}_{\text{control}} \times 100 \quad (2)$$

The result was expressed as IC_{50}, the concentration of the extract to scavenge at 50% of the DPPH and ABTS radical scavenging activities.

RESULTS AND DISCUSSION

Performance of COs from Northern Buru Island

The yield of COs in bud, leaf, and stem were distilled from dry plant material using steam distillation, and the results are shown in Table 1. Results show that the highest COs yields were 8.21% (w/w), in bud 4.22% (w/w) in leaf oil, and 3.75% (w/w) in stem oil from Waihani village. Sohilait (2015) reported the yield of bud (7.05% w/w), leaf (3.21%, w/w), and stem (3.58%, w/w) of *E. caryophyllata* from Amboina Island, Moluccas. Other studies have also reported the yield of bud as 7.1 ± 0.8% (Hassine *et al*. 2021).

Table 1. Yield of COs from Northern Buru Island

Sample	Sample Weight (g)			COs Weight (g)			Yield (wt%)		
	Bud	Leaf	Stem	Bud	Leaf	Stem	Bud	Leaf	Stem
COsWh	1000.10	1289.09	2517.83	82.12	54.41	94.40	8.21	4.22	3.75
COsI	1442.69	2058.72	1002.60	116.60	40.08	18.06	8.08	1.95	1.80
COsWm	1000.08	1254.20	1440.42	70.30	38.48	44.04	7.03	3.07	3.06

COsWh: sample of Waihani village; COsI: Ilath village; COsWm: Waimoli village

Chemical Composition of COs in Leaf

Chemical composition of COs in leaf from Northern Buru were analyzed by GC and GC-MS. There were four components in COsWh, five components in COsI, and three components in COsWm (Table 2, Fig. 1), according to their elution order on the Rtx-5 column. The main constituent eugenol in COsWh (77.1%), COsI (78.8%), and COsWm (78.7%) of leaf and the second main constituent was β-caryophyllene (19.6%, 17.0% and 18.3%, respectively). Other studies have also reported high concentrations of eugenol in leaf clove oil 81.0% to 86.0% at growing area Amboina Island (Sohilait *et al*. 2019) and 78.3% in West Amboina Island (Sohilait *et al*. 2018).

Table 2. Chemical Composition of leaf in CosWh, COsI, and COsWm

No.	RT (min)	KI [*, a]	Compounds	COsWh (%)[b]	COsI(%)[b]	COsWm (%)[b]
1	8.478	1031	1,8-Cineole (**1**)	-	1.0	-
2	14.650	1359	Eugenol (**2**)	77.1	78.8	78.7
3	15.542	1419	β-Caryophyllene (**3**)	19.6	17.0	18.3
4	16.008	1454	α-Humulene (**4**)	2.1	2.0	2.0
5	16.875	1523	Eugenyl acetate (**5**)	0.77	-	-
6	17.783	1583	Caryophylleneoxide (**6**)	-	1.1	-

Kovats Index literature, ***** a): Adam, 2007, b) Concentration of compounds base on GC-FID peaks

Chemical Composition of COs in Bud

The chemical composition of COs in bud were analyzed the same method as used for the previous sample, which is by their elution order on the Rtx-5 column. The COsWh and COsI contain four components but COsWm was composed of three components (Table 3; Fig. 1). Both COsWh (77.7%) and COsWm (76.1%) showed that same concentration of eugenol as main component in bud. However, the eugenol concentration in COsI (87.7%) was higher than the two previously mentioned samples. Other studies have also reported high concentrations of eugenol in bud clove oil 81.1% to 86.0% (Sohilait 2015), 49.7% (Bhuiyan *et al*. 2010), and 84.6% (Lopes *et al*. 2010).

Table 3. Chemical Composition of Bud in COsWh, COsI, and COsWm

No.	RT (min)	KI * [a]	Compounds	COsWh (%)[b]	COsI (%)[b]	COsWm (%)[b]
1	14.650	1359	Eugenol (**2**)	77.7	87.7	76.1
2	15.542	1419	β-Caryophyllene (**3**)	6.0	5.6	5.0
3	16.008	1454	α-Humulene (**4**)	0.8	0.7	-
4	16.875	1523	Eugenyl acetate (**5**)	14.8	6.0	18.9

Kovats Index literature, *a): Adam, 2007, b) Concentration of compounds base on GC-FID peaks

Chemical Composition of COs in Stem

The data from GC/GC-MS of COs in stem from Buru showed that the presence of eugenol in the three samples had similar concentration (94.2% to 97.6%). However, COsWh contains phenyl propene group of eugenol (**2**) (94.2%) and chavicol (7) (0.4%), sesquiterpene hydrocarbon (α-humulene (**4**), at; 0.5%) and sesquiterpene oxygenated component (caryophyllene oxide at 0.6%), though these were at low concentrations (Table 4, Fig. 3G, 3H, 3I). Other studies have also reported high concentrations of eugenol in stem clove oil 97.2% to 98.8% from Amboina Island (Sohilait 2015).

Table 4. Chemical Composition of Stem in COsWh, COsI, and COsWm

No.	RT (min)	KI * [a]	Compounds	COsWh (%)[b]	COsI (%)[b]	COsWm (%)[b]
1	12.930	1250	Chavicol (**7**)	0.4	-	-
2	14.650	1359	Eugenol (**2**)	94.2	97.6	95.9
3	15.542	1419	β-Caryophyllene(**3**)	4.3	2.4	3.6
4	16.008	1454	α-Humulene (**4**)	0.5	-	-
5	17.783	1583	Caryophylleneoxide (**5**)	0.6	-	-

Kovats Index literature, * a): Adam, 2007, b) Concentration of compounds base on GC-FID peaks

Eugenol (**2**) is the main component in different plant parts that produce COs from Waihani, Ilath, and Waimoli villages. Eugenol (**2**), β-caryophyllene (**3**), α-humulene (**4**) and eugenyl acetate (**5**), were observed as the four versatile common components present in leaf, bud, and stem oils with variations in percent content (Tables 2, 3, and 4).

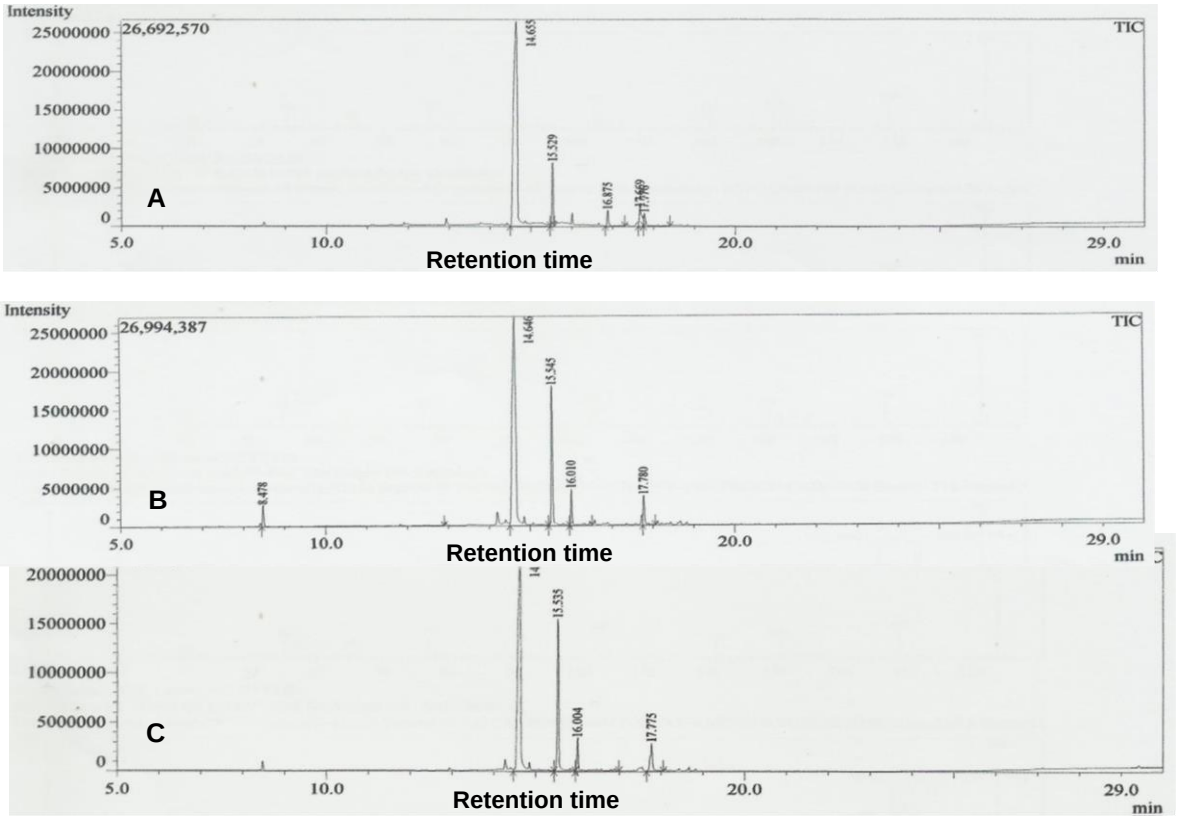


Fig. 1. GC Chromatogram of COsWh (A); COsI (B); and COsWm (C) of leaf

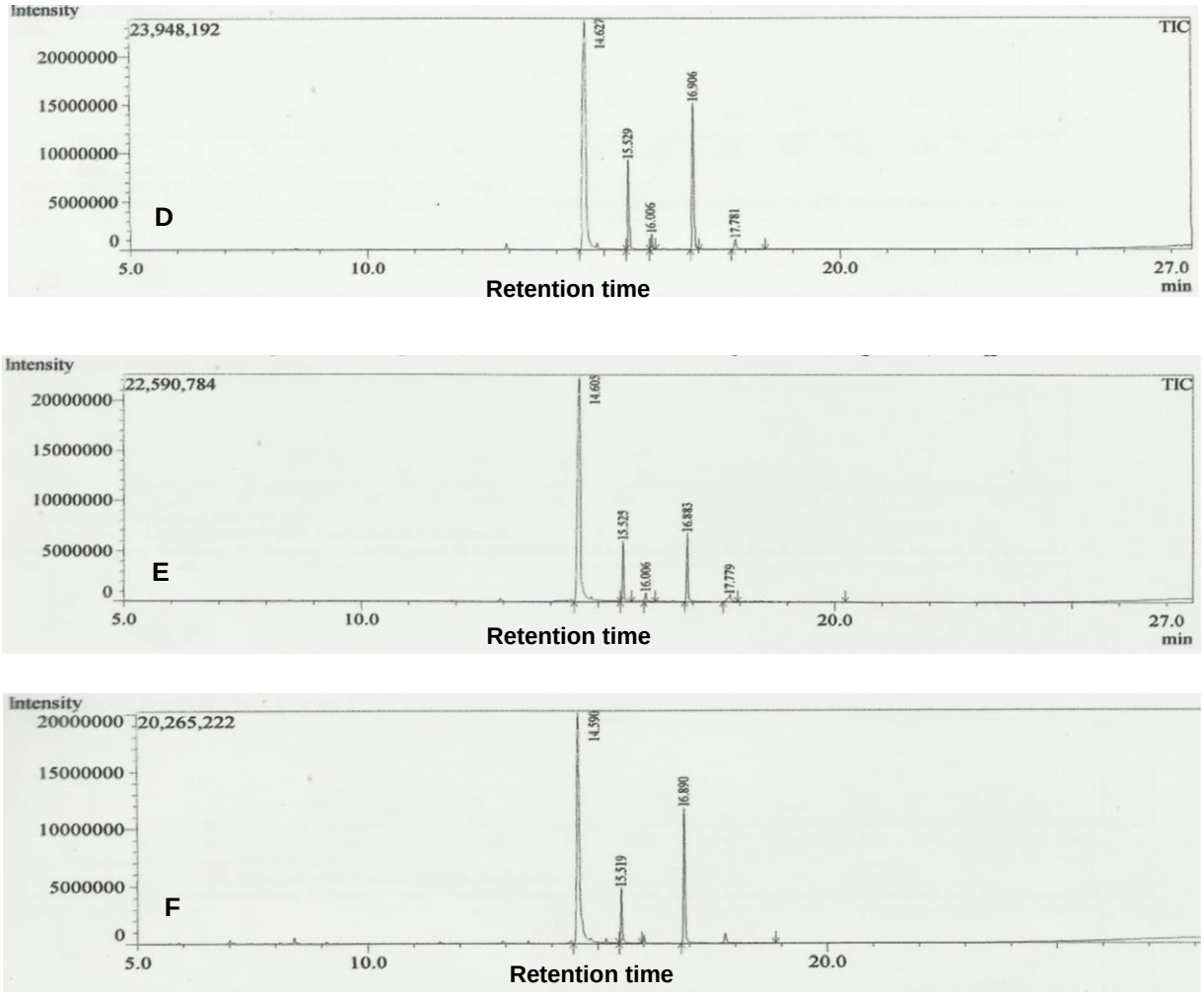


Fig. 2. GC Chromatogram of COsWh (D); COsI (E); and COsWm (F) of bud

The study reveals that except for eugenol (**2**) as the main component, the composition of the oil differs from earlier reports (Srivastava *et al*. 2005; Razafimamonjison *et al*. 2014) and may, therefore, be treated as different chemotypes. The high concentration of eugenol in leaf, buds, and stem oil makes it potentially useful in the medicines because they exhibit antibacterial, antifungal, anti-inflammatory activity, insecticidal, and antioxidant properties, and it is used traditionally as flavoring agent and antimicrobial material in food (Kamatau *et al*. 2012; Lopes *et al*.2020; González *et al*. 2021).

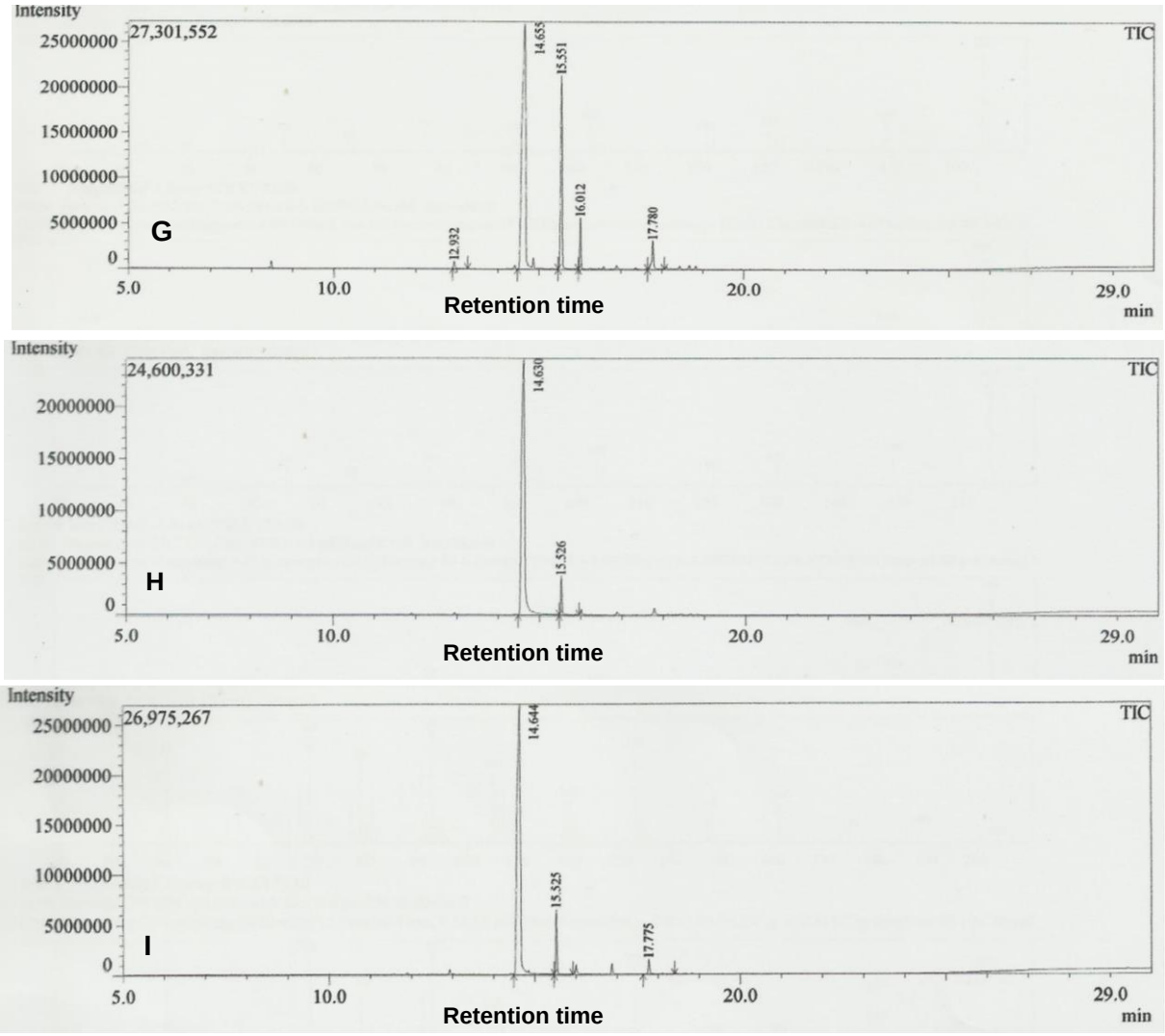


Fig. 3. GC Chromatogram of COsWh (G); COsI (H); and COsWm (I) of stem

Furthermore, COs were identified from northern Buru Island and there were a total of seven secondary metabolites. The compound structures are shown in Fig. 4. The compounds contained in COs from northern Buru show that the essential oils in this area are dominated by the phenylpropene (77.9 to 96.0%) followed by the sesquiterpenes hydrocarbon (2.4 to 21.7%). Compound 1,8-cineole is an oxygenated monoterpene (1.0%) only found in COsI of leaf. The oxygenated sesquiterpenes were found in small amounts in COsI of leaf (1.14%) and COsWh of stem (0.5%), respectively. However, monoterpene hydrocarbon is not found in all plant tissues of COsWh, COsI, and COsWm. The highest concentration of phenylpropene compounds can be found in COsI stem (97.6%) and the lowest content is COsWh leaf (77.9%). A COsWh leaf contained the highest sesquiterpene hydrocarbon (21.7%) among the nine samples analyzed. Based on the analysis of areas where plants grow and the tissues that can produce COs, it can be explained that COsI contains almost all terpene compound groups except for monoterpene hydrocarbons (Fig. 5).

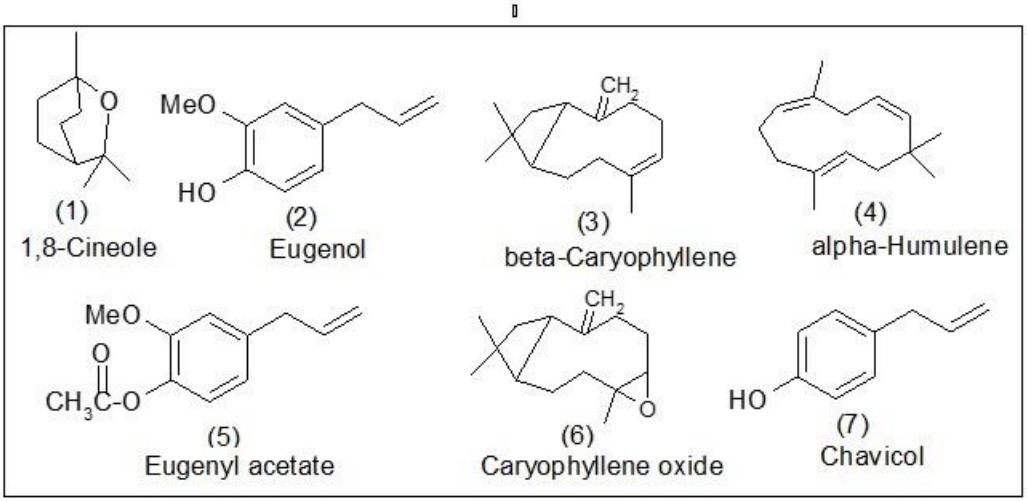


Fig. 4. Structure compounds in COs from northern Buru Island

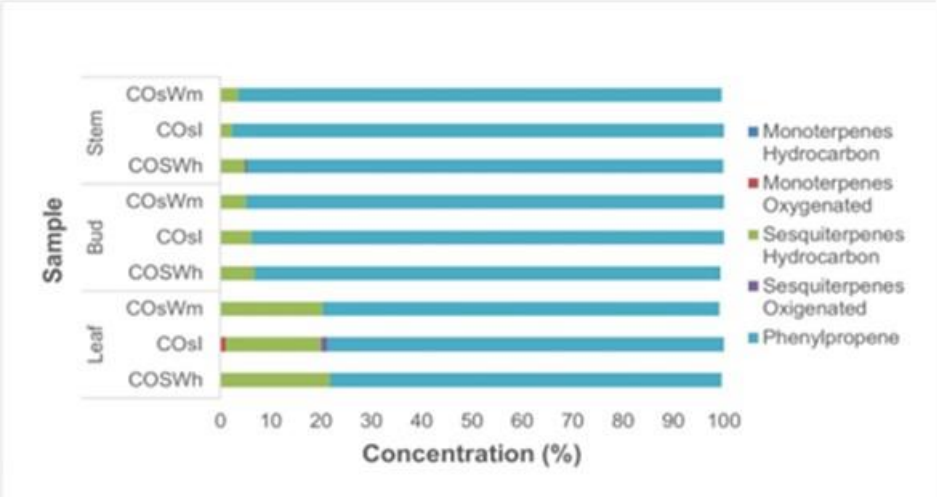
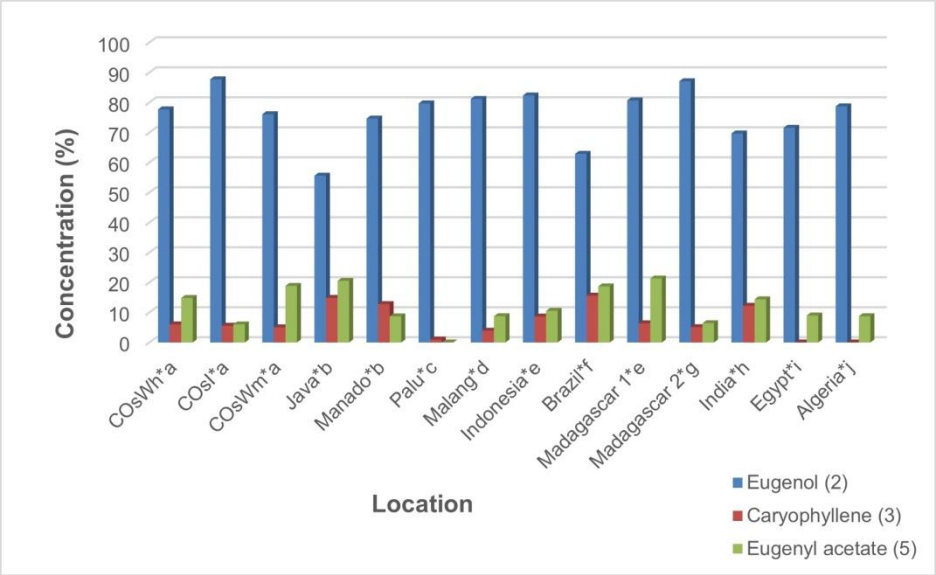


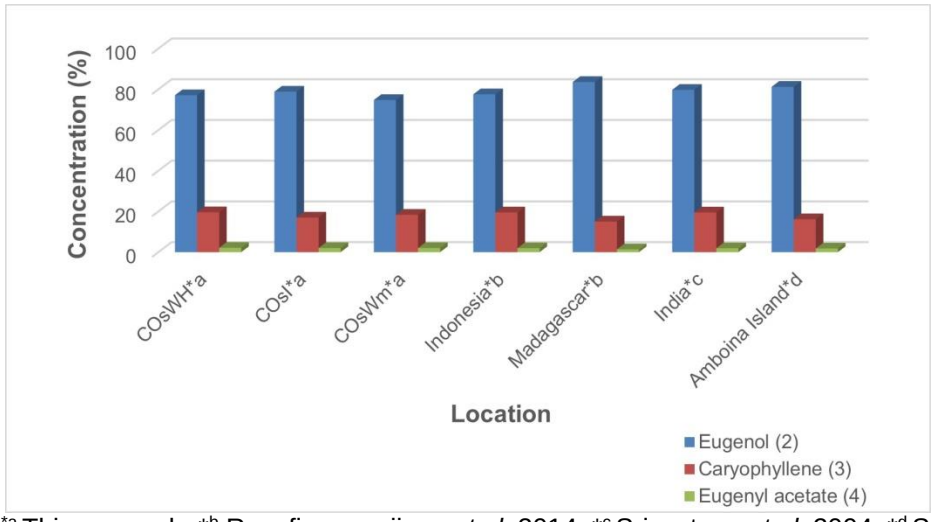
Fig. 5. Profile concentration of terpene in COs northern Buru Island

Geographic condition and environmental factors strongly influence the essential oil chemical composition (Xie *et al.* 2011). Chemical composition of COs of bud in several growth areas and results from this research (Fig. 6) showed the concentration of the major compound of eugenol (**2**) in CosI as 87.7%, which is similar to Madagascar 2 (87.1%). The concentration of eugenyl acetate (**5**) in COsWm (18.9%) was higher than the two samples analyzed in this study and the areas where *E. caryophyllata* grow compared to Manado (8.70%), Malang (8.74%), Indonesia (10.55%), Madagascar 2 (6.40%), India (14.38%), Egypt (8.99%), and Algeria (8.74). However, the concentration of eugenyl acetate (**5**) in COsWm is lower than Java (20.5%) and Madagascar 1 (21.3) and similar to Brazil (18.69%). The presence of eugenyl acetate (**5**) in COsWh (14.8%) was not noticeably different from that in India (14.4%). Concentrations of β-caryophyllene (3) in COsWh (6.03%), COsI (5.56%), and COsWm (5.04%) were not noticeably different from Madagascar 2 (5.1%) but were higher than Palu (0.96%) and Malang (3.92%).



*a This research, *bAmelia *et al*. 2017; *c Tahir *et al*. 2020; *d Prianto *et al*. 2013; *e Razafimamonjison *et al*. 2014; *f de Oliviera et al. 2016; *g Gonzales *et al*. 2016; *h Kaur *et al*. 2019; *i Nassar *et al*. 2007; *j Selles *et al.* 2020

Fig. 6. Concentrations of major compounds in COs Bud of northen Buru Island and other locations

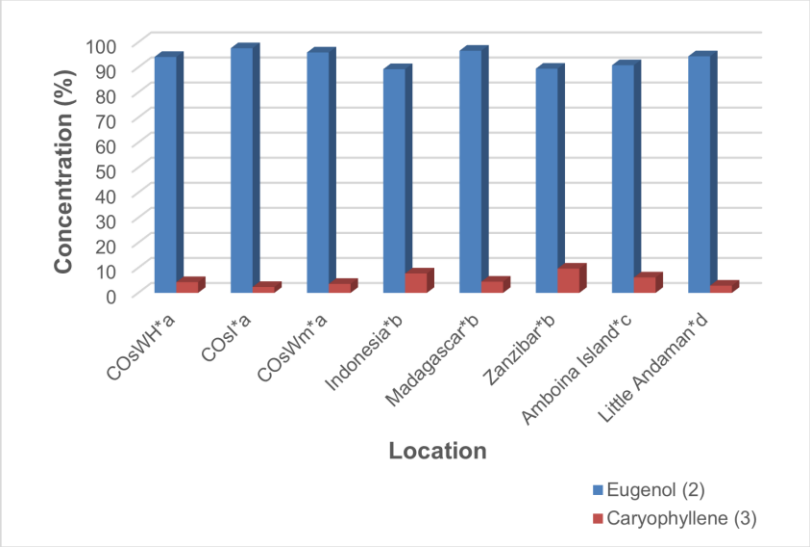


*a This research, *b Razafimamonjison *et al*. 2014; *c Srivastava *et al*. 2004; *d Sohilait *et al*. 2018

Fig. 7. Concentrations of major compounds in COs leaf of northern Buru Island and other locations

The concentration of eugenol (**2**) COs of leaf *E. caryophylata* in Fig. 7 showed that WOsI > COsWh > COsWm. The concentration of (**2**) as the main compound in COsI (78.8%) is similar to that in India (79.7%) and Indonesia (77.5%) but lower than Amboina Island (81.2%) and Madagascar (83.6%). The composition of caryophyllene (**3**) in COsWh (19.6%) is the same as COs Indonesia (19.5%) and India (19.5%) and higher than Madagascar (15.0%) and Amboina Island (16.2%). The chemical composition of eugenyl acetate (**4**) was the same in COs of leaf from three locations in northern Buru island, COsWh (2.12%), COsI (2.02%) and COsWm (2.03%) respectively. Likewise, when

compared to the other four locations, Indonesia (1.93%), Madagascar (1.39%), India (1.90%) and Amboina Island (1.80%) did not show a noticeable difference.



[*a] This research, [*b]Razafimamonjison *et al*. 2014; [*c] Raina *et al*. 2001; [*d] Sohilait *et al*. 2015

Fig. 8. Concentration of major compounds in COs Stem of Northen Buru Island and other locations

Figure 8 shows that major components are eugenol (**2**) and caryophyllene (**3**) COs of stem in northern Buru Island. The eugenyl acetate (**5**) component was not found in COsWh, COsI, and COsWm, respectively. The composition of eugenol (**2**) in COsI of stem (97.6%) was higher than COsWh (94.2%) and COsWm (96.0%) also at five other locations. However, the concentration of eugenol (**2**) in COsWh of stem is similar to Little Andaman (94.4%) but higher than Indonesia (89.3%), Zanzibar (89.5%), and Amboina Island (90.8%). The concentration of caryophyllene (**3**) component, in COsWh (4.30%) is similar to Madagascar (4.48%) but lower than Zanzibar (9.70%), Indonesia (7.75%) and Amboina Island (6.16%).

Antioxidant Activities of COs from Northern Buru Island

The antioxidant activity of COs from Buru was evaluated by DPPH and ABTS assays. Free radical scavenging effect of COs in various concentration is shown in Figs. 9 and 10. Concentrations of 159.73 to 0.623 µg/mL were found for DPPH; and 99.0 to 1.54 µg/mL for ABTS. Data show the mean SD (n = 3) for each experiment performed in triplicate; $p < 0.01$ for comparisons of BHT and AA as positive control.

The COsWh of bud (97.21 ± 0.58%) and COsWm of bud (97.21 ± 0.13%) showed the same inhibition of DPPH free radical scavenging at the highest concentration (159.7 µg/mL). The inhibition of DPPH at concentration 0.623 µg/mL (lowest concentration), COsWm of bud (47.03 ± 0.33%) was higher than the nine samples used in this study, even the positive control (Fig. 9). Furthermore, COsWh of bud showed higher DPPH activity, as indicated by the IC_{50} value 0.09 ± 0.37 µg/mL (Table 5).

This study shows data on COs northern Buru inhibition of ABTS free radical at highest concentration (99 µg/mL) in the range of 96.02 ± 0.30 to 99.74 ± 0.23%. There is no remarkable difference when compared with BHT and AA as positive controls. The COs

in bud at different locations showed the same inhibition of COsWh and COsWm, namely 99.74 ± 0.23%. However, at the lowest concentration in the ABTS assay (1.54 µg/mL), COsI of bud inhibition (37.06 ±0.64 µg/mL) was higher than all samples and controls used in study (Fig.10). Calculation based on the equation of the interpolation curve, the free radical scavenging activity of ABTS COsI of bud (IC_{50} value 0.09 ± 0.61 µg/mL) is higher than samples evaluated in this study. These data also showed that COsI of bud is higher than AA (IC_{50} value 0.81 ± 1.64 µg/mL) and BHT (IC_{50} value 0.83 ± 0.54 µg/mL) as positive controls (Table 5).

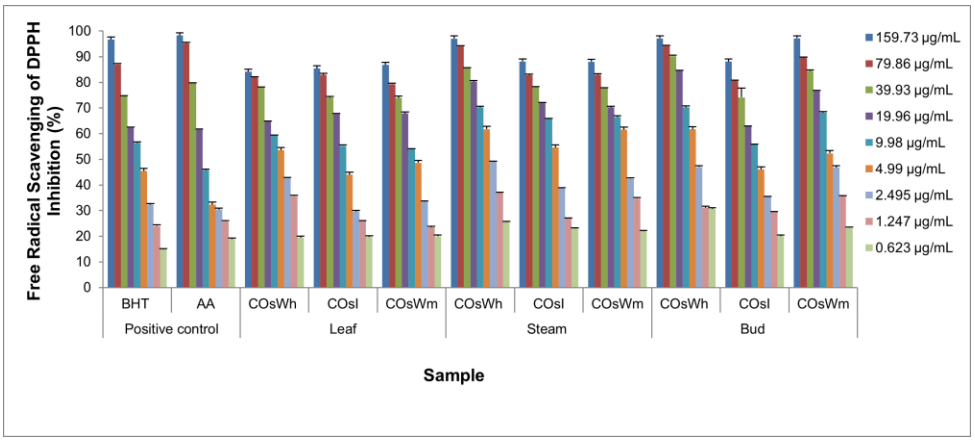


Fig. 9. Inhibition (%) of DPPH Free radical scavenging of Cos

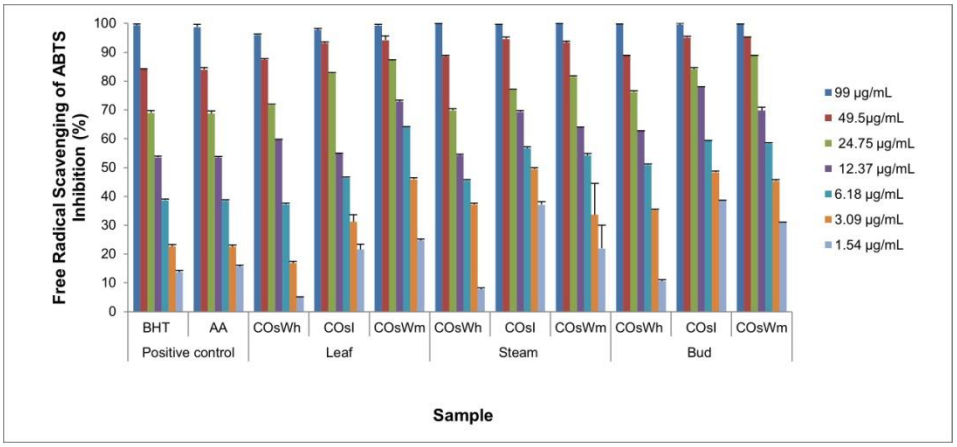


Fig. 10. Inhibition (%) of ABTS Free radical scavenging of COs

The results obtained provide information that COs from different plant parts have free radical scavenging activity of DPPH and ABTS in strong classification. This excellent performance is dominated by COs of bud, followed by stem and leaf. The COs of bud contain high eugenol (76.10% to 87.74%), and there is also eugenyl acetate (14.82% to 18.86%) from phenyl propene group. Both compounds synergize with proton donors in powerful free radical scavenging action. Phenolics compound have favorable antioxidant activity naturally (Wang *et al.* 2022).

Free radical scavenging (IC_{50} value) depends on chemical composition of essential oils. The large quantities of phenyl propene serve as main compounds in clove oil,

increasing its activity (Mahboubi *et al.* 2015). The hydroxyl group available in eugenol on the aromatic ring is responsible for the antioxidant activity. The phenolic compounds transfer electrons or hydrogen atoms and neutralize them to free radicals, resulting in a blocked oxidative process (Gonzales *et al.* 2022). The eugenol (**2**), eugenyl acetate (**5**), and β-caryophyllene (**3**) as the main components of clove oil play a very important role, as shown in this research. The present research data shows concentrations of (**2**) (77.7%), (**3**) (6.03%), and (**5**) (14.8%) and synergistic effects in COsWh (IC_{50} 0.09 ± 0.37 μg/mL) of bud that against DPPH free radical is higher than COsI (IC_{50} 0.64 ± 0.54 μg/mL) and CosWm (IC_{50} 0.09 ± 0.37 μg/mL). COsI of bud, as well as against ABTS free radical (0.09 ± 0.61 μg/mL) with concentration 87.7% of (**2**), 6.02 % of (**5**), (5.56%) of (**3**) (Table 5). This data is strengthened by the values of Pearson correlation coefficients (*r*) between chemical composition and free radical scavenging DPPH and ABTS. High positive correlations between free radical scavenging DPPH activity of COsWh bud and major compounds are (**2**) (r: 0.85, $p < 0.01$); (**3**) (r: 0.87, $p < 0.01$); (**5**) (r: 0.70, $p < 0.01$) respectively. This relationship also applies to COsI and ABTS free radical scavenging DPPH activity which are (**2**) (r: 0.98, $p < 0.01$); (**3**) (r: 0.73, $p < 0.01$); (**5**) (r: 0.65, $p < 0.01$).

Table 5. IC_{50} Value of COs from Northern Buru Island

COs/Standard	DPPH		ABTS	
	Inhibition (%)	IC_{50} Value (μg/mL)	Inhibition (%)	IC_{50} Value (μg/mL)
COsWh of Leaf	84.15 ± 0.44	0.48 ± 0.36	96.02± 0.30	0.85 ± 0.30
COsWh of Bud	97.21 ± 0.58	0.09 ± 0.37	99.74± 0.23	0.68 ± 0.47
COsWh of Stem	97.08 ± 0.45	0.29 ± 0.19	99.54± 0.17	0.76 ± 0.42
COsI of Leaf	85.50 ± 0.33	0.69±0.29	97.75± 0.46	0.70 ± 0.38
COsI of Bud	88.12 ± 0.01	0.64±0.54	99.44± 0.17	0.09 ± 0.61
COsI of Stem	88.20 ± 0.34	0.46±0.33	99.59± 0.46	0.23 ± 0.48
COsWm of Leaf	86.77 ± 0.33	1.31±0.33	99.23± 0.40	0.32 ± 0.18
COsWm of Bud	97.21 ± 0.13	0.32±0.44	99.74± 0.23	0.31 ± 0.24
COsWm of Stem	88.03 ± 0.44	0.23±0.13	99.64± 0.70	0.58 ± 0.25
BHT (positive control)	96.73 ± 0.16	0.67 ± 0.45	99.34±0.43	0.83 ± 0.54
AA (positive control)	98.32 ± 0.35	0.70 ± 0.23	98.71±0.93	0.81 ± 1.64

The location of plants during their growth will influence chemical composition and bioactivity (Kainama *et al.* 2023). This result showed that COsWh of bud DPPH free radical scavenging abilities of (**2**), (**3**) and (**5**) were higher than COs in Indian plants. In addition, high concentration (**5**) (14.82%) can affect DPPH free radical scavenging, which is more active compared to Indian COs bud (IC_{50} value 2.6 μg/mL).

CONCLUSIONS

1. The essential oils of clove (*Eugenia caryophyllata*) from northern Buru Island were obtained by steam distillation. The chemical composition of COs was analyzed by GC–FID and GC–MS techniques. The main component is eugenol (77.1 to 78.8%) and β-caryophyllene (17.0% to 19.6%) in leaf oil. The main components are eugenol (76.1% to 87.7%) and eugenyl acetate (6.0% to 18.9%) in bud oil and the only one component was eugenol (94.2% to 97.6%) in stem oil from Waihani, Ilath, and Waimoni.

2. The COs of leaf, bud, and stem showed strong activity and strong potential for the application of clove essential oil as a natural DPPH and ABTS free radical scavenging.

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