ISOLATION AND STRUCTURE ELUCIDATION OF SECONDARY METABOLITE COMPOUNDS FROM *Curcuma aeruginosa*

Alsya Firdausi Nuzula[1], Alfinda Novi Kristanti[1,2*], Nanik Siti Aminah[1,2], Yoshiaki Takaya[3], Marisa Nur Sahadatin[1], Andika Pramudya Wardana[1]
[1]Department of Chemistry, Faculty of Science and Technology, Universitas Airlangga, Kampus C UNAIR, Jl. Mulyorejo, Surabaya 60115, East Java, Indonesia
[2]Biotechnology of Tropical Medicinal Plants Research Group, Universitas Airlangga, Jl. Dharmahusada Permai, Surabaya 60115, East Java, Indonesia
[3]Faculty of Pharmacy, Meijo University, Tempaku-468-8502, Nagoya, Japan
[*]Email: *alfinda-n-k@fst.unair.ac.id*

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Abstract

Black turmeric (*Curcuma aeruginosa*) is Indonesian medicinal plant belonging to the *Zingeberaceae* family, mostly found on Java. This study aimed to determine the new molecular structures of the compounds isolated from black turmeric originating from Indonesia. Isolation of the secondary metabolite compounds was initiated by solid-liquid extraction using the maceration method with ethanol solvent, followed by liquid-liquid extraction using the partition method with *n*-hexane and dichloromethane as solvents. Separation and purification were carried out using vacuum liquid chromatography (VLC) and gravity column chromatography (GCC). The isolated compounds were characterized using spectroscopic methods, including UV-Vis, IR, and NMR spectroscopy. The isolation resulted in three sesquiterpenoid compounds, *(E)-7-isopropyl-4,10-dimethylcyclodec-10-ene-5,8-dione* (*curdione*), *(1R,10R)-Epoxy-(-)-1,10-dihydrocurdione,* and *zedoalactone B,* together with two common terpenes, *ethyl tetradecanoate* and *1-hexadecene*.

Keywords*: black turmeric, Curcuma aeruginosa, medicinal plant, sesquiterpenoids, Zingeberaceae*

Introduction

Indonesia is tropical rainforest country with high biodiversity, with approximately 11% of plant species found on the Earth's surface (Widhyani *et al*., 2017). Indonesia's tropical climate has led to the development of plants that are used to treat various diseases. Curcuma is local plant with the potential to be studied is *Curcuma*. *Curcuma* is a genus of the *Zingiberaceae* family (ginger), which has more than 1200 species, almost all of which grow in tropical forests, particularly in Southeast Asia. One of the plant species of the genus *Curcuma* is the most popular in the community for treating various diseases, including *Curcuma aeruginosa* and black turmeric. Studies on *C. aeruginosa* are gaining attention, as extracts and isolated compounds from this plant can also act as antioxidant, antibacterial, anti-inflammatory, and antipyretic agents (Safitri *et al*., 2017 dan Simoh *et al*., 2018).

Black turmeric contains a variety of secondary metabolites in its rhizome and leaves. The rhizome and leaves were aromatic, indicating the presence of volatile constituents. Black turmeric rhizomes contain saponins, tannins, terpenoids, flavonoids, steroids, and polyphenols (Akarchariya *et al*., 2017; Harnis *et al*., 2022; Fitria *et al*., 2019). In addition, approximately 51% of sesquiterpenes and 18% of monoterpenes

from the ethanol extract of *C. aeruginosa* Roxb were identifiedusing GC-MS analysis (Nurcholis *et al*., 2021).

The compounds identified from black turmeric have various biological activities, including antirheumatic, anti-inflammatory, and antibacterial (Akarchariya *et al*., 2017 and Alonso-Amelot, 2016). Fresh and dried black turmeric leaves mostly essential oil isolated using GC-MS were 1,8-cineole (25.12 %), curzerene (9.01 %) germacrone (4.91 %) and camphor (6.55 %) (Abdul *et al*., 2021). The essential oils showed good antibacterial and antifungal activity against *S. mutants, S. aureus, B. subtilis, E. coli, A. niger* and *C. albicans* (Wahyuni *et al*., 2017, Abdul *et al*., 2021, and Sari *et al*., 2022).

Based on the high potency of black turmeric, the aim of this study was to determine the molecular structures of compounds isolated from black turmeric originating in East Java, Indonesia. To date, the isolation of secondary metabolites from native Indonesian plants has not yet been reported. In this study, extraction was carried out in steps using several solvents to obtain three kinds of extracts, namely *n*-hexane, dichloromethane, and ethyl acetate. Thus, this study attempted to study the diversity of secondary metabolites found in the rhizomes of this species, which were extracted using solvents.

The dichloromethane extract resulted in isolating five pure compounds from sesquiterpenoids and terpenes groups, starting from *n*-hexane extract resulted *curdione,* resulting in *1-hexadecene, ethyl tetradecanoate, (1R,10R)-Epoxy-(-)-1,10 dihydrocurdione*, the ethyl acetate extract resulted *zedoalactone B,* through Separation and purification processes were performed by vacuum liquid chromatography and gravity column chromatography techniques.

Research Methods

Materials

Samples of rhizome *C. aeruginosa* plants were collected in February 2021 from Jogorogo village, Ngawi, East Java, Indonesia, and a voucher specimen (UA-ZCa290622) was deposited at the Herbarium of Universitas Airlangga. The rhizome was cleaned, air-dried in the shade, cut into small pieces, and milled.

The organic solvents used in this research were *n*-hexane, dichloromethane (DCM), ethyl acetate (EtOAc), and methanol (MeOH) of technical grade, which had been distilled. A precoated silica gel GF_{254} 0.25 mm (Merck) plate was used for thin layer chromatography (TLC), and anisaldehyde-sulfuric acid reagent was sprayed on the TLC plate for visualization. Silica gel 60 GF_{254} was used for vacuum liquid chromatography (VLC), and silica gel G_{60} 0,040 – 0,63 mm (Merck) was used for gravity column chromatography (GCC).

Instrumentations

Secondary metabolite compounds were characterized using a UV-Vis Shimadzu 1800 spectrophotometer, a BRUKER Alpha II ECO ATR-FTIR spectrophotometer, and BRUKER 600 MHz NMR spectroscopy.

Extraction and isolation

The rhizome of *C. aeruginosa* was ground to a coarse powder and weighted to 4.4 kg. The powder was placed into maceration vessels, which were used for maceration with ethanol as a solvent for 3 × 24 h. After filtration, the filtrate was evaporated using a rotary vacuum evaporator to obtain the thick ethanol extract. The ethanol extract was partitioned thrice using a separating funnel with *n*-hexane:ethanol (1:1). The remaining ethanol extract was partitioned again using DCM:ethanol (1:2) three times. The results of the partitioning process were obtained *n*-hexane and DCM extracts.

The *n*-hexane extract (35 g) was separatedusing VLC, starting with 100% *n*-hexane until it became *n*-hexane:DCM (2:8) with an increased polarity gradient. The separation produced 12 main fractions (A-M). The main fraction C of the VLC results was separated by GCC I using a mixture of *n*-hexane:DCM eluent (1:1) to produce five subfractions (C1-C5). Subfraction C1 was then separated with CCG II using *n*-hexane:DCM eluent (9:1) to obtain **compound 1**.

The DCM extract (35 g) was separated using VLC, starting with *n*-hexane:DCM (9:1) until it became DCM 100% with an increased polarity gradient. This separation produced 16 main fractions (A1-A9 and B1-B7). The combination of fractions A7 and B3 was further separated usingGCC I with *n*-hexane:DCM as the eluent (7:3). The separation resulted in 13 subfractions. Subfraction 8 was separated using GCC II with *n*-hexane:EtOAc (99:1) and GCC III with *n*-hexane:DCM eluent (4:6) until **compound 2**. To determine the purities of the two compounds, three different eluent systems were tested. A flow diagram of this study is shown in Figure 1.

Characterization

The isolated compounds were prepared and dissolved in dichloromethane (DCM). Then, every 1 mg sample was subjected to UV-Vis spectroscopy at a wavelength (λ) of 200-500 nm to determine the maximum wavelength that showed the characteristics of the compound (Kafle, 2020) and IR spectroscopy at a wave number range of 400-4000 (cm^{-1}) to determine the functional groups contained in the structure of a compound (Dash *et al*., 2021). Moreover, 5 mg samples were dissolved in a deuterated solvent that could be utilized forNMR spectroscopy analysis at the chemical shift range around δ_H of 1-10 ppm and δ_C of 1-200 ppm (Lo *et al*., 2021).

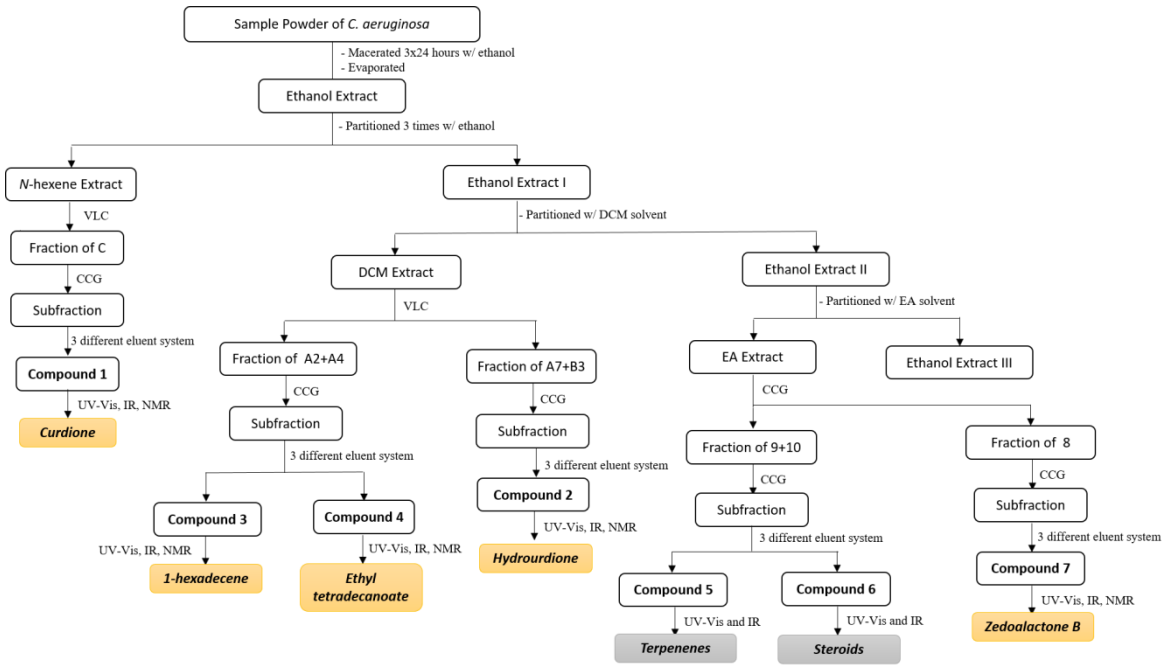

Figure 1. The flow diagram of research

Results and Discussion

Determination of secondary metabolite compounds

Every 35 g of *n*-hexane and DCM extract was separated using VLC and GCC. The first separation process using VLC used a mixture of *n*-hexane and dichloromethane eluents, which were gradually increased in polarity. The separation results were analyzed by thin-

layer chromatography (TLC). The fractions that contained the same Rf on spot TLC from VLC could be combined to produce some main fractions for *n*-hexane extract (A-M) and DCM extract (A1-A9 and B1-B7).

Fraction C from *n*-hexane extract was further separatedusing GCC with *n*-hexane:DCM (1:1) as the eluent and hexane:DCM (9:1) as the eluent. The separation resulted in one spot on TLC with the same Rf, namely **compound 1** which formed a white powder and weighed 90 mg. Subsequently, it was checked with three different eluent systems: a mixture of *n*-hexane:DCM (2:8), *n*-hexane:acetone (8:2), and *n*-hexane:DCM:chloroform (2:4:4). The results show one spot withconsecutive Rf values of 0.28, 0.43, and 0.78.

The combined fraction of A7 and B3 from the DCM extract was separated using GCC I with *n*-hexane:DCM (7:3) eluent. The subfractions were further separated by performing GCC II with *n*-hexane:EtOAc (99:1) and then separated using GCC III with *n*-hexane:DCM (4:6) as the eluent. Finally, the separation resulted in a single spot on TLC with the same Rf, which is **compound 2** which formed a yellowish white crystalline structure and weighed 40 mg. Subsequently, the isolated compounds were tested using three different eluents. The eluent was mixed with *n*-hexane:acetone (9:1), *n*-hexane: chloroform (1:1), and *n*-hexane: chloroform (5:3:2). The results show one spot with consecutive Rf values of 0.37, 0.54, and 0.60.

Compound 1

The UV-Vis spectrum of **compound 1** showed a maximum wavelength of 219 nm.Absorbance was determined to be 0.347. This indicated that compound 1 did not have a conjugated system in its structure (Atmoko *et al*., 2018). The IR spectrum showed absorption bands withwavenumbers of 2966-2858 cm^{-1} (C-H sp^3), 1767 cm^{-1} (C=O ketones), and 1695 cm^{-1} (C=C alkenes).

Analysis of the ^{13}C-NMR spectrum of **compound 1** (Table 1) completed with DEPT 135 and DEPT 90 showed two quaternary carbon signals from a ketone group (C=O) at δ_C 214.2 and 211.0 ppm, one quaternary carbon signal from a vinylic group (C=C) 129.8 ppm, one methine carbon signal (CH) at δ_C 131.5 ppm and three carbon signals methine (CH) at δ_C 53.5; 46.7; and 29.9 ppm, four carbon signals of the methylene (CH_2) group at δ_C 55.8; 44.2; 33.9; and 26.3 ppm, as well as four carbon signals from the methyl group (CH_3) δ_C 16.5; 18.4; 19.7; and 21.0 ppm. The presence of 15 carbon signals is a characteristic of the sesquiterpenoid class (Zhang *et al*., 2022).

Analysis of ^{1}H-NMR spectrum of **compound 1** (Table 1) in $CDCl_3$ 600 MHz indicated the presence of four proton signals in the methine (CH) group which consisted one proton sgnal from the vinylic methine group at δH 5.15 (1H, m) ppm as well as three proton signals from other methine groups at δ_H 2.32 (1H, m), 2.83 (1H, ddd, *J* = 9.17; 9.17; 2.36 Hz), and 1, 86 (1H, m) ppm. Moreover, four proton signals of the methylene (CH_2) group showed at δ_H 2.92; 3.05 (2H, d, *J*=12 Hz), 2.37 (1H, dd, *J*= 16.62; 2.28 Hz); 2.68 (1H, m), 2.08 (2H, m), and 1.56; 2.12 (2H, m) ppm, as well as four proton signals from the methyl group (CH_3), among others: two proton signals from the methyl group adjacent to the methine group formed an isopropyl fragment at δ_H 0.94 (3H, d, *J*= 6.68 Hz) ppm and 0.87 (3H, d, *J*= 6.59 Hz) ppm, a proton signal from a methyl group adjacent to a methyl group at δ_H 0.97 (3H, d, *J*=6.94 Hz) ppm, and a signal from methyl group adjacent to the quaternary carbon at δ_H 1.65 (3H, s) ppm.

Table 1. NMR data of **compound 1**

Number of C	δ_H (ppm) HSQC	δ_C (ppm)	HMBC (ppm)
1	5,15 (1H, *s*)	131,4	-
2	2,09 (2H, *m*)	26,3	C-1, C-3, C-4
3	1,56 and 2,12 (2H, *m*)	33,9	C-1, C-2, C-5, C-14
4	2,32 (1H, *m*)	46,7	-
5	-	214,2	-
6	2,38 (1H, *dd*, *J*= 16,69; 2,29 Hz and 2,68 (1H, *m*)	44,1	C-5, C-7, C-8, C-11
7	2,84 (1H, *ddd*, *J =8,8*; 8,8; 2,40 Hz)	53,5	C-6, C-8, C-11, C-13
8	-	211,0	-
9	2,94; 3,05 (2H, *d*, *J* = 11 Hz)	55,8	C-1, C-8
10	-	129,7	-
11	1,86 (1H, *m*)	29,9	C-7, C-13
12	0,94 (3H, *d*, *J* = 6,69 Hz)	19,7	C-7, C-11, C-13
13	0,87 (3H, *d*, *J* = 6,60 Hz)	21,0	C-7, C-11, C-12, C-14
14	0,97 (3H, *d*, *J* = 6,97 Hz)	18,4	C-3, C-4, C-5
15	1,65 (3H, *s*)	16,4	C-1, C-9, C-10

HMBC analysis showed a correlation between H-7 (δ_H 2.83 ppm) and C-6, C-11, C-13, and H-11 (δ_H 1.86 ppm) with C-12, C-13 indicating the position of the isopropyl fragment, and the correlation of H-1 (δ_H 5.15 ppm) with C-9 and C-2 was identified as the position of the alkene group. The heteronuclear multiple bond correlation (HMBC) of **compound 1** is shown in Figure 2. According to Zhang *et al.* (2018), this compound also exists in a C-C conformation possessing a *trans* relationship (*E*) between the C-4 methyl and C-7 isopropyl side chains. Moreover, this conformation is characteristic of the *syn*-arrangement of the C-5 carbonil and C-1 methine (Inayama *et al*., 1985). Based on the ^{1}H-NMR, ^{13}C-NMR, HSQC, and HMBC spectra, compound 1 was predicted to be compound *(E)-7-isopropyl-4,10-dimethylcyclodec-10-ene-5,8-dione* (*curdione*). A comparison of the proton and carbon data of compound 1 with the literature is shown in Table 2 (Fang *et al*.,2019).

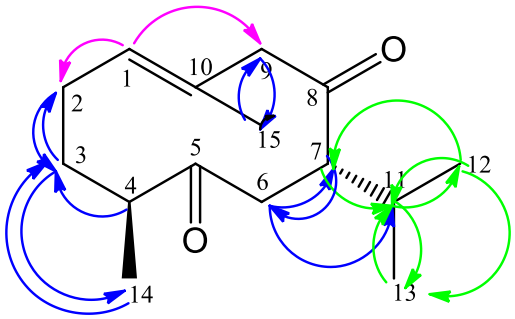

Figure 2. HMBC correlation in **compound 1**

Table 2. Comparison of ^{1}H-NMR and ^{13}C-NMR data of **compound 1** (*curdione*) with literature (Fang *et al*., 2019)

Number of C	Compound 1		Literature	
	δ_H (ppm)	**δC (ppm)**	**δ_H (ppm)**	**δC (ppm)**
1	5,15 (1H, *m*)	131,5	5,163 (1H, *m*)	131,5
2	2,08 (2H, *m*)	26,3	2,11 (2H, *m*)	26,5
3	1,56 ; 2,12 (2H, *m*)	33,9	1,58 ; 2,12 (2H, *m)*	34,1
4	2,32 (1H, *m*)	46,7	2,34 (1H, *m*)	46,8
5	-	214,2	-	214,2
6	2,37 (1H, *dd*, *J*= 16,6 ; 2,2 Hz) ; 2,68 (1H, *m*)	44,2	2,40 (1H, *dd*, *J*= 16,6 ; 2,2 Hz) ; 2,71 (1H, *m*)	44,3
7	2,83 (1H, *ddd, J = 9*,2 ; 9,2 ; 2,4 Hz)	53,5	2,85 (1H, *ddd, J* = 8,8 ; 8,8 ; 2,2 Hz)	53,6
8	-	210,9	-	210,9
9	2,92 ; 3,05 (2H, *d, J*=12,0 Hz)	55,8	2,94 ; 3,07 (2H, *d,J*=10,7 Hz)	55,9
10	-	129,8	-	129,8
11	1,86 (1H, *m*)	29,9	1,88 (1H, *m*)	30,0
12	0,93 (3H, *d*, 6,7 Hz)	19,7	0,95 (3H, *d*, 6,7 Hz)	19,9
13	0,87 (3H, *d*, 6,6 Hz)	21,0	0,88 (3H, *d*, 6,6 Hz)	21,2
14	0,96 (3H, *d*, 6,9 Hz)	18,4	0,98 (3H, *d*, 7,0 Hz)	18,6
15	1,64 *(*3H, *s*)	16,5	1,657 (3H, *s*)	16,7

Compound 2

The UV-Vis spectrum of **compound 2** showed a maximum wavelength of 219 nm. Absorbance was determined to be 0.546. This indicates that compound 2 did not have a conjugated system in its structure (Atmoko *et al*., 2018). The IR spectrum showed absorption bands with a wave number 2972-2882 cm^{-1} (C-H sp^3), 1707 cm^{-1} (C=O), and 1054-1086 cm^{-1} (C-O).

Analysis of ^{13}C-NMR spectrum on **compound 2** (Table 3) completed data from C-DEPT 90 NMR and C-DEPT 135 NMR consisted of two carbonyl signals (C=O) at δ_C 213 .0 and 208.9 ppm, one quaternary carbon signal at δ_C 58.9 ppm, four methine (CH) carbon signals at δ_C 64.1; 55.9; 44.4; and 30.2 ppm, four signals carbon methylene (CH_2) at δ_C 50.8; 42.6; 29.5; and 24.7 ppm as well as four methyl carbon (CH_3) signals at δ_C 21.0; 20.5; 18.9; and 16.1 ppm. Based on these data, there are two carbons that bind O: quaternary δ_C 58.9 ppm and methine 64.1 ppm. The presence of 15 carbon signals is a characteristic of the sesquiterpenoid class (Zhang *et al*., 2022).

Analysis of the ^{1}H-NMR spectrum of **compound 2** (Table 3) in $CDCl_3$ at 600 MHz showed the presence of four methine (CH) proton signals, namely at δ_H 2.89 (1H, dd, *J* = 10.0; 4.0 Hz), 2.68 (1H, m), and 2.82 (1H, d, *J* = 4.0 Hz) ppm. Moreover, it showed a forming isopropyl fragment with methine at δ_H 1.72 (1H, m) ppm which is adjacent with two methyl carbon signals at δ_H 0.98 (3H, d, *J* = 6.6 Hz) ppm and 0.88 (3H, d, *J* = 6.6 Hz) ppm. The other six methine proton signals were identified at δ_H 2.79 (1H, d, *J* = 4.0 Hz), 3.08 (1H, m), 2.71 (1H, d, *J* = 11.3 Hz), 2.24 (1H, d, *J* = 11.3 Hz), 2.08 (1H, m), and 1.35 (1H, m) ppm, which were derived from the protons of the three non-identical methylene groups, as confirmed by the HSQC data (Table 3). In addition, the proton signal of the methylene (CH_2) group appeared at δ_H 1.27 (2H, s) ppm. Furthermore, there were two other proton

signals from the methyl group (CH_3), which are shown at δ_H 1.18 (3H, s) and 1.08 (3H, d, *J*= 6.6 Hz).

Table 3. Data NMR of **compound 2**

Number of C	δ_H (ppm) HSQC	δ_C (ppm)	HMBC (ppm)
1	2,89 (1H, *dd*, *J* = 10,0; 4,0 Hz)	64,1	C-2
2	2,08 (1H, *m*); 1,35 (1H, *m*)	24,7	-
3	1,27 (2H, *s*)	29,5	-
4	2,82 (1H, *d*, *J* = 4,0 Hz)	44,4	-
5	-	213,0	-
6	2,79 (1H, *d*, *J* = 4,0 Hz) and 3,08 (1H, *m*)	42,6	C-5
7	2,68 (1H, *m*)	55,9	C-8
8	-	208,9	-
9	2,71 (1H, *d*, *J* = 11,3 Hz) and 2,24 (1H, *d*, *J* = 11,3 Hz)	50,8	C-8
10	-	58,9	-
11	1,72 (1H, *m*)	30,2	-
12	0,98 (3H, *d*, *J* = 6,6 Hz)	20,5	C-7, C-11, C-13
13	0,88 (3H, *d*, *J* = 6,6 Hz)	21,0	C-7, C-11, C-12
14	1,08 (3H, *d*, *J* = 6,6 Hz)	18,9	C-4, C-5
15	1,18 (3H, *s*)	16,1	C-1, C-8, C-9

The HMBC analysis showed a correlation of H-6 (δ_H 2,79 and 3,08 ppm) with C-5 and H-7 (δ_H 2,68 ppm) and H-9 (2,71 and 2,24 ppm) with C-8, indicating the position of the two ketone groups. In addition, the correlations of H-12 (δ_H 0,98 ppm) with C-7, C-11, C-13, and H-13 (δ_H 0,88 ppm) with C-7, C-11, and C-12 indicated the position of the isopropyl fragment. The heteronuclear multiple bond correlation (HMBC)of **compound 2** is shown in Figure 3. According to Hariyama *et al*. (1991) the ten-membered ring of this compound, *trans*-fused with the oxirane group at C-1 and C-10, exists in a chair-boat conformation. The C-4 methyl and C-7 isopropyl groups are also on the opposite sides (*R, R*) of the average molecular plane (Hariyama *et al*., 1991). Based on the ^{1}H-NMR, ^{13}C-NMR, HSQC, and HMBC spectra, **compound 2** was predicted to be *(1R,10R)-Epoxy-(-)-1,10-dihydrocurdione*, comparison of the proton and carbon data of **compound 2** with those in the literature is presented in Table 4 (Hariyama *et al*., 1991).

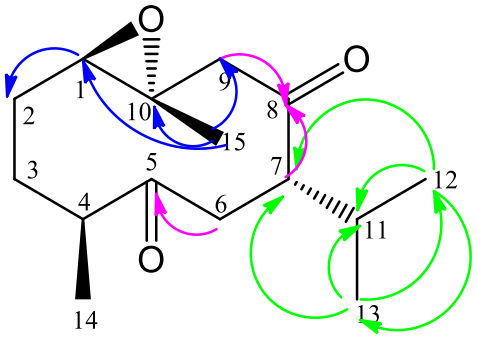

Figure 3. HMBC correlation in **compound 2**

Tabel 4. Comparison of ^{1}H-NMR and ^{13}C-NMR data of **compound 2** (*hydrocurdione*) with literature (Hariyama., 1991)

Number of C	Compound 2		Literature	
	δH (ppm)	**δC (ppm)**	**δH (ppm)**	**δC (ppm)**
1	2,89 (*dd*, *J*= 10,0; 4,0)	64,1	2,89 (*dd*, *J*= *9,9; 3,8*)	64,1
2	2,08 (*m*); 1,35 (*m*)	24,7	2,07 (*m*); 1,69 (*m*)	24,8
3	1,27 (*s*)	29,5	1,34 (*m*)	29,6
4	2,82 (*d*, 4,0)	44,4	2,83 (*m*)	44,5
5	-	213,0	-	213,1
6	2,79 (*d*, *J*= 4,0); 3,08 (*m*)	42,6	2,81 (*dd*, *J*= 18,3; 4,0); 3,08 (*dd*, *J*= 18,3; 11,9)	42,5
7	2,68 (*m*)	55,9	2,66 (*dd*, *J*= 11,9; 4,0)	56,0
8	-	208,9	-	208,9
9	2,71 (*d*, *J*= 11,3); 2,24 (*d*, *J*=11,3)	50,8	2,69 (*d*, *J*= 11,2)	50,9
10	-	58,9	-	58,9
11	1,72 (*m*)	30,2	1,73 (*m*)	30,1
12	0,98 (*d*, *J*= 6,6)	20,5	0,97 (*d*, *J*= 6,6)	20,6
13	0,88 (*d*, *J*= 6,6)	21,0	0,87 (*d*, *J*= 6,6)	21,1
14	1,08 (*d*, *J*= 6,6)	18,9	1,07 (*d*. *J*= 6,8)	18,9
15	1,18 (*s*)	16,1	1,17 (*s*)	16,1

Based on this study, wesuccessfully isolated five pure compounds, including *curdione* and *hydrocurdione*. The other three pure compounds have also been compared to other literature, such as *1-hexadecene* which was previously identified in *Chrozophora tinctoria* (L.) using GC-MS analysis (Sher *et al*., 2022). *Ethyl tetradecanoate* was found in the ethyl acetate extracts of *Citrus maxima* L. and *Commiphora swynnertonii*using GC-MS and NMR analyses (Zulbayu *et* al., 2021 and Credo *et al*., 2022). Moreover, *zedoalactone B* whose guaiane-type sesquiterpenes had been isolated by performing IR and NMR analysis from *Curcuma kwangsiensis, Curcuma xanthorrhiza,* and *Curcuma zedoaroides* which has been widely used as antiinflammation (Park *et al*., 2017, Tungcharoen *et al*., 2018, and Liao *et al.*, 2020).

Conclusions

Two new pure compounds from the rhizome of *Curcuma aeruginosa* Roxb. originating in East Java, Indonesia has been successfully isolated as *(E)-7-isopropyl-4,10-dimethylcyclodec-10-ene-5,8-dione* (*curdione*) from *n*-hexane

extract. The other compounds also identified as *1-hexadecene*, *ethyl tetradecanoate*, and *(1R,10R)-Epoxy-(-)-1,10 dihydrocurdione* from dichloromethane extract as well as *zedoalactone B* from ethyl acetate extract. The isolated compounds belongs to the sesquiterpenoid and terpene groups.

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Conflict of Interest

The authors have declared that no competing interests exist.

Data Availability Statement

All relevant data are within the paper and its Supporting Information files.

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Author Contributions

AFN and MNS conducted the experiment, YT and APW conducted the calculations, ANK and NSA wrote and revised the manuscript. All authors agreed to the final version of this manuscript.

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