TRANSFORMATION OF EUGENOL AND SAFROLE INTO HYDROXYCHAVICOL

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ABSTRACT

Hydroxychavicol is found in betel leaf at low concentration and is reported to have antibacterial, anti-inflammatory, antioxidant, anticancer, and antimutagenic activities. This study aimed to synthesize hydroxychavicol from eugenol and safrole. Isolation of eugenol from clove oil by alkaline extraction method gave 71% yield, while the isolation of safrole from lawang oil by alkaline extraction method, followed by purification using preparative thin layer chromatography, gave 7% yield. Eugenol demethylation and safrole demethylenation with $AlCl_3$ reagent were successfully produced hydroxychavicol. The yields were 28% and 24%, respectively. Mechanisms of the synthesis are proposed in this article.

Keywords: *demethylation; demethylenation; eugenol; hydroxychavicol; safrole*

ABSTRAK

Hidroksikavikol dijumpai dalam daun sirih dengan kadar yang rendah dan diketahui memiliki aktivitas antibakteri, antiradang, antioksidan, antikanker, dan antimutagen. Penelitian ini bertujuan menyintesis senyawa tersebut dari eugenol dan safrol. Isolasi eugenol dari minyak cengkih dengan metode ekstraksi basa menghasilkan rendemen 71%, sedangkan isolasi safrol dari minyak lawang dengan metode ekstraksi basa dilanjutkan dengan pemurnian menggunakan kromatografi lapis tipis preparatif menghasilkan rendemen 7%. Proses demetilasi isolat eugenol dan demetilenasi isolat safrol dengan pereaksi $AlCl_3$ berhasil mendapatkan produk hidroksikavikol. Rendemen yang diperoleh berturut-turut 28% dan 24%. Mekanisme reaksi dalam sintesis diajukan dalam artikel ini.

Kata Kunci: *demetilasi; demetilenasi; eugenol; hidroksikavikol; safrol*

INTRODUCTION

Indonesia is the largest producer and consumer of cloves in the world, above the production of Madagascar and Zanzibar. It is widely known that eugenol is the main component in oil extracted from clove flower. Eugenol utilization in industry is generally limited to the production of flavor. Eugenol has functional groups that can be chemically modified so that, in principle, eugenol is a useful starting material for the synthesis of many more useful compounds, one of which is hydroxychavicol. Hydroxychavicol has antibacterial [1-2], antifungal [3], antioxidant [2], and anti-inflammatory activities [2,4], as well as cytotoxic effects [5]. Hydroxychavicol can be isolated from betel leaf. However, extraction of betel leaves only produce hydroxychavicol not more than 0.26% [6]. When compared with 80% levels of eugenol in clove oil, it can produce a lot more of eugenol to be transformed into hydroxychavicol.

Hydroxychavicol can also be synthesized from safrole. Formerly, safrole is widely used for beverage flavor enhancer, but it was later known carcinogenic and since 1960, its use has been banned in the United States [7]. Safrole is also used in the illegal production of 3,4-methylenedioxymetamphetamine (MDMA); thus, it is considered a precursor compound.

This study aims to synthesize hydroxychavicol from eugenol and safrole. Eugenol was chosen as the starting material due to its abundance and to increase the antimutagenicity. On the other hand, safrole was chosen to alleviate the carcinogenic properties. The basic principle of this transformation is demethylation, meaning removal of methyl group ($-CH_3$) from eugenol, and demethylenation, which is replacement of methylenedioxy group ($-OCH_2O-$) on safrole to form 1,2-diol groups (Fig. 1). Lewis acid is a commonly used reagent for demethylation, such as BBr_3 [8] and $AlCl_3$ [9], as well as for demethylenation, such as $BF_3{\cdot}O(C_2H_5)_2$ [10] and $AlCl_3$ [11-12]. Considerations used in determining reagent are availability and cost

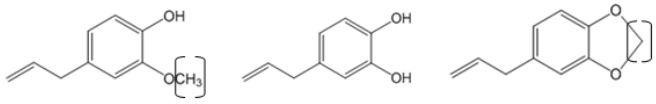

Fig 1. Structures of eugenol, hydroxychavicol, and safrole

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factors. In this study, anhydrous aluminum chloride ($AlCl_3$) was used. Until now, this reagent is still commonly used as demethylation reagent for quite simple aryl methyl ethers [13-14] as well as more complex compounds such as (*E*)-3,5-dihydroxy-4-isopropylstilbene [15] and quinine [16].

EXPERIMENTAL SECTION

Materials

Materials used included commercial cloves oil and *lawang* oil obtained from CV Kemika Jaya Bogor, eugenol standard (purity >99%, Sigma-Aldrich), anhydrous $AlCl_3$, dimethyl sulfide (DMS p.a.), CH_2Cl_2, silica gel 60 GF_{254} (Merck) for preparative thin layer chromatography (PTLC), silica gel 60 G (0.040–0.063 mesh, Merck) for flash column chromatography, and N_2 gas.

Instrumentation

Basic instruments used were thin layer chromatography plates (TLC, silica gel 60 F_{254}) and flash column chromatography (FCC). The spectra of ultraviolet-visible (UV-Vis) were recorded with a Shimadzu UV-1601 spectrometer at the Common Laboratory, Bogor Agricultural University. Fourier transform infrared spectra (FTIR) were recorded on KBr pellets using Shimadzu FTIR-8201PC spectrometer. Gas chromatograph-mass spectrometer (GCMS) was equipped with electron impact detector and consisted of a gas chromatograph GC-17A (Shimadzu) coupled with a mass spectrometer MS QP 5050A capillary column DB-5 ms (J & W) (silica, 30 m × 250 μm × 0.25 μm); column temperature 50 °C (*t* = 0 min) to 290 °C at a rate of 15 °C/min; helium carrier gas at a constant pressure 7.6411 psi; with Wiley 7N database (2008)] at the Forensic Laboratory, Police Headquarters, Jakarta. ^{1}H-NMR spectra obtained with a JEOL ECA 500 spectrometer working at a frequency of 500 MHz.

Procedure

Steps of this study consisted of isolation of eugenol from clove oil, isolation of safrole from *lawang* oil, demethylation of the standard (pure) eugenol and the isolated eugenol, and demethylenation of the isolated safrole. The isolated eugenol and safrole were characterized by TLC and GCMS. The isolated eugenol was also characterized by UV-Vis spectrometer and FTIR spectrophotometer. The products of demethylenation and demethylation were characterized by TLC based on R_f values, and the product of demethylated eugenol was further characterized by ^{1}H-NMR.

Isolation of eugenol (modified [17])

Clove oil (10 mL, density 1.0021 g/mL) in *n*-hexane (20 mL) was mixed with 20 mL NaOH 2 M, stirred for 15 min using a magnetic stirrer, and transferred into a separating funnel. The water layer was collected, and the organic layer was re-extracted with 20 mL NaOH 2 M and stirred for 30 min. After settling about 10 min, the water layer was collected and the organic layer was monitored using TLC to ensure no remaining eugenol. All water layers were combined and neutralized with dropwise H_2SO_4 15% to pH 6. Subsequently, it was extracted with CH_2Cl_2 (2×15 mL), dried with anhydrous Na_2SO_4, and concentrated by rotary evaporator.

Isolation of safrole

Lawang oil (10 mL, density 0.9776 g/mL) in CH_2Cl_2 (20 mL) was added to 50 mL NaOH 1 M, stirred for 10 min using a magnetic stirrer, and transferred to a separation funnel. The water layer were separated, the organic layer was extracted twice more with 50 mL NaOH 1 M, for 20 and 30 min, respectively, and tested with 5% $FeCl_3$, giving negative results. The organic layer was then dried with anhydrous Na_2SO_4, and concentrated. The isolated safrole was re-purified by preparative thin layer chromatography (PTLC) using mixture of *n*-hexane-EtOAc (8:2).

Demethylation of eugenol (modified [9])

Dimethyl sulfide (2.5 mL) was added dropwise to a suspension of anhydrous $AlCl_3$ (0.330 g, 2.5 mmol) in CH_2Cl_2 (5 mL) at 0 °C while stirring until $AlCl_3$ was completely dissolved. Then the solution of eugenol (0.164 g, 1.0 mmol) in CH_2Cl_2 (3 mL) was added in a period of 10 min at the same temperature. The mixture was allowed to stand at room temperature and stirred for 24 h. All dissolution process and the reaction were carried out in a nitrogen gas atmosphere. Afterward, 15 mL cold HCl 1 N was added and the mixture was extracted with CH_2Cl_2 (2×15 mL). The organic layer was dried with anhydrous Na_2SO_4, filtered, and concentrated. The crude product was purified using FCC with *n*-hexane-EtOAc (8:2) eluent.

Demethylenation of safrole (modified [12])

Safrole solution (0.24 g, 1.5 mmol) in CH_2Cl_2 (7.0 mL) was added slowly to a cold suspension of $AlCl_3$ (0.68 g, 5.1 mmol) in CH_2Cl_2 (5.0 mL) at 0 °C. The resulting mixture was stirred for 2 hours at -10 °C, added with 10 mL of cold water, and stirred again for 18 h at room temperature. All the dissolution and reaction were carried out under nitrogen atmosphere.

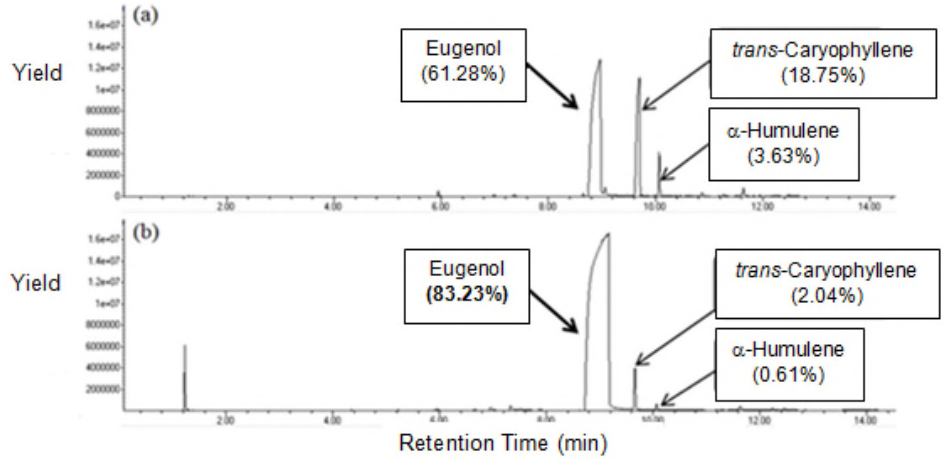


Fig 2. GCMS chromatogram of clove oil (a) and the isolated eugenol (b)

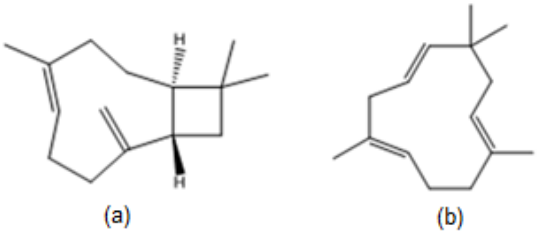


Fig 3. Structures of *trans*-caryophyllene (a) and *α*-humulene (b)

Afterward, the mixture was poured into a saturated $NaHCO_3$ solution (100 mL) and extracted with ethyl acetate (2×50 mL). The organic layer was washed again with 100 mL saturated $NaHCO_3$ solution and dried with anhydrous Na_2SO_4, filtered, and concentrated. The crude product was purified using FCC with *n*-hexane-EtOAc (8:2) eluent.

RESULT AND DISCUSSION

The Isolated Eugenol

Based on the analysis of GCMS (Fig. 2a), eugenol is the highest component of clove oil sample (61%), with 2 main impurities: *trans*-caryophyllene (19%), and α-humulene (4%) (Fig. 3). Similar results are also reported by Jirovetz et al. [18] with different composition, namely 77%, 17%, and 2%, respectively.

Eugenol was isolated from the clove oil using NaOH extraction. Being phenolic in nature (pK_a = 10.3), eugenol reacted with bases to form salts of Na-eugenolate, which was water-soluble. This salt was separated from impurities components that were soluble in nonpolar *n*-hexane, as also reported elsewhere [17]. Stirring increased kinetic energy of the reacting molecules so that intermolecular collisions increased and accelerated the rate of reaction. The reaction was exothermic. The optimum reaction product was achieved at room temperature.

The water layer containing Na-eugenolate was then neutralized with acid to reform the eugenol. Low concentration (15%) of H_2SO_4 was dripped slowly from the burette until the pH 6, which is marked by the color of the solution changes from yellow to white. There was no addition reaction to the double bond by the dilute H_2SO_4 as expected from Markovnikov reaction. The eugenol formed was extracted with dichloromethane. The remaining water was removed by adding anhydrous Na_2SO_4 in order not to interfere with the demethylation reaction.

The alkaline extraction gave eugenol isolates with an average yield of 71%, which was 83% pure as confirmed by GCMS analysis (Fig. 2b). The yield is nearly similar to that reported by Sudarma et al. [19], who isolated eugenol using column chromatography and gradient elution with *n*-hexane-dichloromethane. Fig. 2b clearly shows that the impurities are greatly reduced: *trans*-caryophyllene decreased from 18.75% to 2.04% and α-humulene from 3.63% to 0.61%. Some of nonpolar components such as α-pinene, α-limonene, α-cubebene, β-cubebene, α-amorphene, and α-farnesene are successfully removed and are undetectable at GCMS chromatograms. There is trace of dichloromethane (1.56%).

Monitoring by TLC using the best eluent (*n*-hexane-EtOAc 8:2) to isolate eugenol showed an R_f ~0.67, which was the same as the standard eugenol. The isolated eugenol was also confirmed by the UV-Vis spectrum showing the maximum wavelength, λ_{max}, at 281 nm. The result was similar to the value reported by Bihari et al. [20].

FTIR spectrum of the isolated eugenol showed similar absorption peaks as reported by Mohammed and Al-Bayati [17]. Broad absorption band at 3525 cm^{-1}

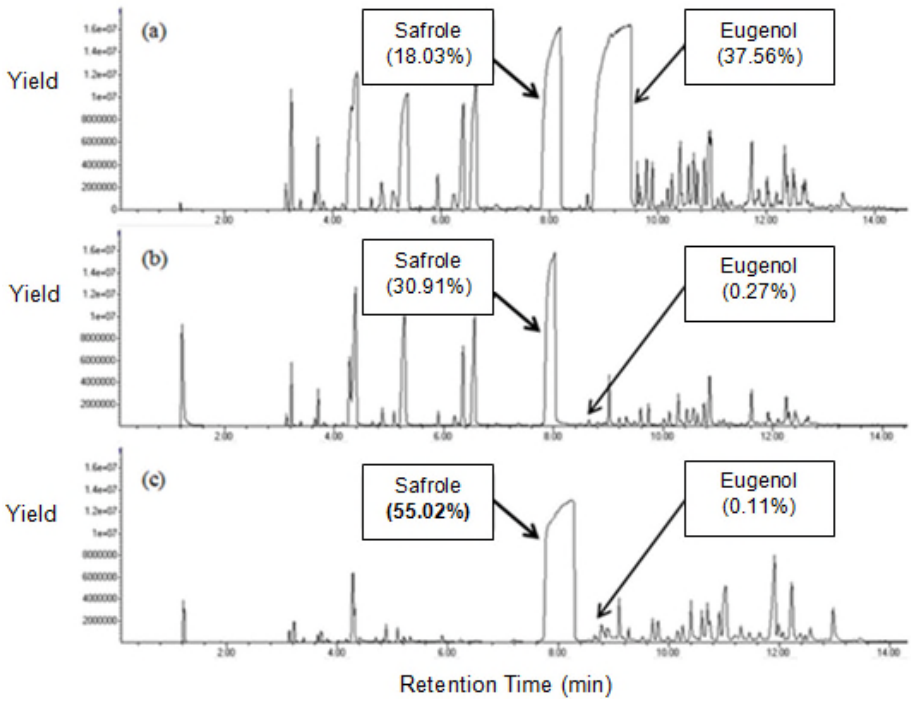


Fig 4. GCMS chromatogram of *lawang* oil (a), the isolated safrole from alkaline extraction (b), and purified isolated safrole using PTLC (c)

was due to O–H stretching vibrations. Sharp absorptions at 2842 and 2939 cm^{-1} were from C–H stretching vibrations of the methyl group (C-sp^3) and a double bond (C-sp^2), respectively. Absorptions at wavenumbers 1515 and 1611 cm^{-1} were originating from stretching vibrations of aromatic C=C. The double bond (C=C) was indicated by absorption at 1638 cm^{-1}. Absorption at 1035 cm^{-1} was due to C-O stretching vibrations of the methoxy group.

The Isolated Safrole

GCMS analysis of the oil samples shows only 18% safrole content, the second largest component after eugenol (38%) (Fig. 4a). In addition, there are also eucalyptol (6.96%), linalool (5.28%), α-terpineol (4.01%), and α-pinene (1.23%).

Safrole was first isolated by crystallization, based on the freezing point of pure safrole that is 11 °C. With a purity of about 20%, the freezing safrole by a linear equation should be -12 °C [21]. To reach this temperature, a mixture of dry ice and acetone was employed. The crystals formed were collected and analyzed by GCMS. Surprisingly, eugenol content was not decreasing; however, this process could reduce the impurities, i.e., α-humulene, α-murolene, δ-cadinene, caryophyllene oxide, α-cadinol, muurolol, and azunol.

Alkaline extraction was again used to separate eugenol from safrole. Adding 50 mL NaOH 1 M and stirring for 30 min was able to remove eugenol, as indicated by negative test with 5% $FeCl_3$, showing the absence of phenolic compound. The TLC assay using *n*-hexane-EtOAc (8:2) eluent also confirmed a single spot at R_f ~0.84. The use of alkaline extraction gave an average yield of 38% crude safrole, with 31% purity as showed by GCMS chromatogram (Fig. 4b). This procedure was able to eliminate impurities, i.e. β-ocymene, piperitol, chavicol, δ-elemene, *trans*-caryophyllene, β-celinene, δ-cadinene, and muurolol. Eugenol content was much reduced from 38% to 0.27%.

The crude safrole was further purified using PTLC plate with the same eluent. The yield at this stage was 19% (or 6.6% of the total *lawang* oil) and the purity was 55% according to GCMS chromatogram (Fig. 4c). In addition to the increased level of safrole, the refining process removed more impurities, namely phelandrene, phenyl alcohol, 1-terpineol, and 4-terpineol, and greatly reduced eugenol, α-pinene, eucalyptol, linalool, and α-terpineol.

Hydroxychavicol from Demethylation of Eugenol

Demethylation or replacement of the methyl group with hydrogen on eugenol to form hydroxychavicol was carried out using $AlCl_3$. The reaction was performed in a homogeneous system by first dissolving eugenol in CH_2Cl_2 and $AlCl_3$ in CH_2Cl_2-DMS. $AlCl_3$ is a Lewis acid that can accept lone pair of O-methoxy in the eugenol

Table 1. Yield of transformation from eugenol into hydroxychavicol

No.	CH_2Cl_2 grade	$AlCl_3$ (mmol)	Eugenol (mmol)	Hydroxychavicol (mmol)	Yield (%)
1◊	Technical	2.55	1.0069*	0.1738	17.26
2♦	Technical	2.51	1.0009*	0.2550	25.48
3♦	Technical	4.02	1.0051*	0.2331	23.19
4♦	p.a	2.56	0.9954*	0.1079	10.84
5♦	p.a	2.63	0.9882*	0.5600	56.67
6♦	p.a	2.51	0.9978*	0.3090	30.97
7♦	p.a	2.51	0.9912*	0.3190	32.18
8♦	p.a	2.70	1.0193+	0.2803	27.50
9♦	p.a	2.58	1.0705+	0.3030	28.30

Remark: ◊ (addition of 10 mL cold HCl);
♦ (addition of excessive cold HCl);
* (standard eugenol, 99.00% purity);
+ (isolated eugenol, 83.23% purity)

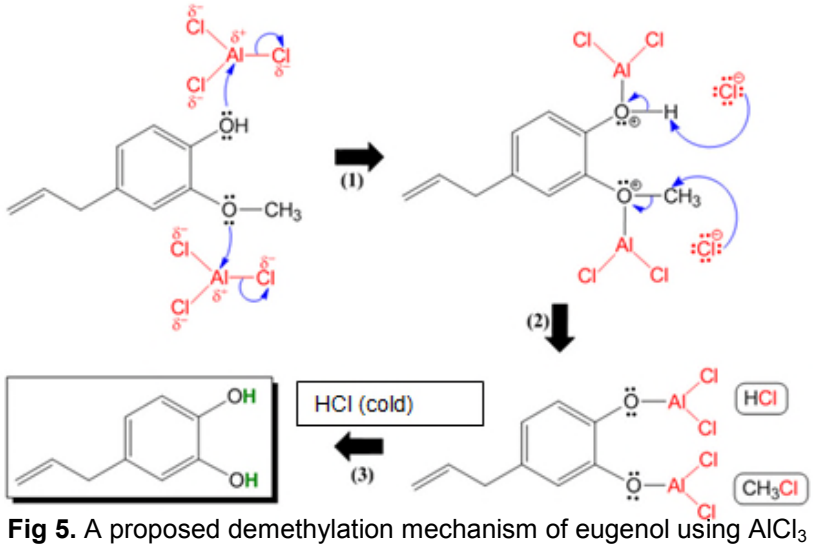


Fig 5. A proposed demethylation mechanism of eugenol using $AlCl_3$

molecule. The O-hydroxy atom can donate its electron pair as well, although the ability is slightly smaller than O-methoxy atom that receives electron release from the methyl group. Therefore, the reaction was carried out with excess $AlCl_3$ (2.5 equivalents). A proposed demethylation mechanism is as follows (Fig. 5). Electron pairs on O-methoxy atom attack the central Al metal and the Cl group will depart as a good leaving group **(1)**. Chlorine ion is also nucleophilic in nature and will attack the methyl group, which has partial positive charged, giving a chloromethane byproduct and a complex of Ar-O-$AlCl_2$ **(2)**. During this reaction, N_2 atmosphere is able to avoid interaction with moisture from the atmosphere, which can change $AlCl_3$ into unreactive $Al(OH)_3$. Upon the completion of demethylation step, the complex is decomposed using cold HCl 0.1 N **(3)**.

S_N2 reaction in the demethylation step went slowly. This was because the electron pair on O-methoxy atom may resonate into the aromatic rings thus lowering the chances of nucleophilic attacking the Al atom. In addition, the weak nucleophilic chloride ions seemed to be slow in attacking the methyl group. Although it was slow, the reaction was kept at a low temperature (0 °C) to avoid the addition reactions and unwanted isomerization of the double bond in the allyl group. Therefore, the reaction was run for 24 hours. Decomposition with cold 0.1 N HCl took place in two-layer system. It seemed to reduce the possibility of addition reaction to the double bond in hydroxychavicol as HCl will be more distributed in the water layer, while hydroxychavicol will be more distributed in the organic layer.

Table 1 shows that demethylation of pure eugenol with the limited addition of 0.1 M HCl (10 mL) and the use of technical CH_2Cl_2 solvent produces a low yield (entry 1). Increasing $AlCl_3$ reagent to 4 equivalents has no significant effect on the yield (entry 3). Adding excess HCl and use of pure solvent directly increase the yield up to 32% (entry 6 and 7). Demethylation of the isolated eugenol gives lower yield (entry 8 and 9) due to the lower purity of eugenol (83%). There is an indication that impurities interfere the interaction between $AlCl_3$ and eugenol.

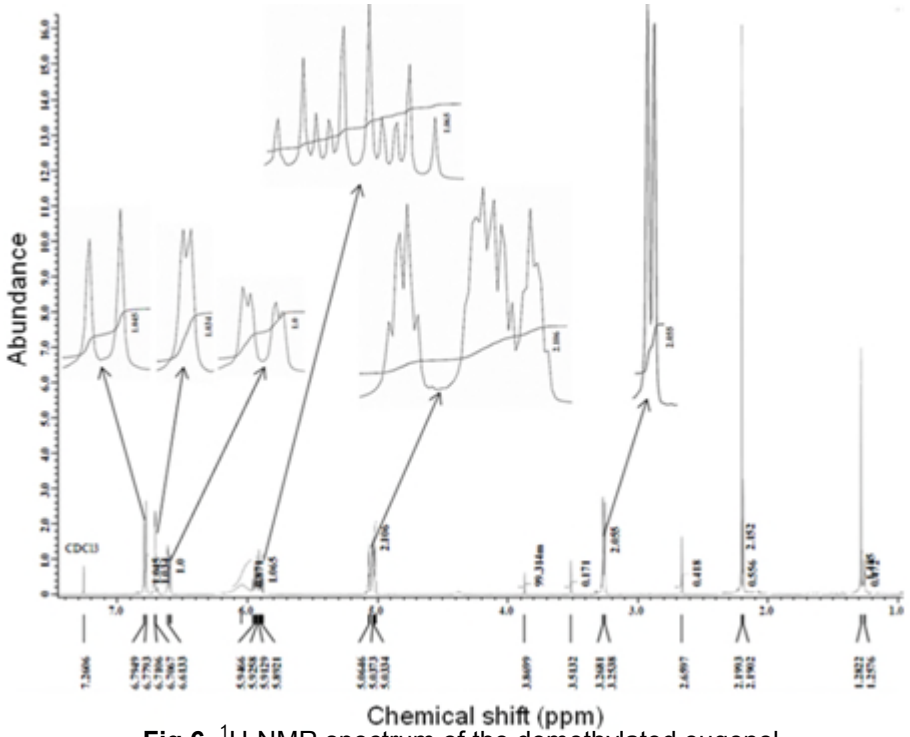
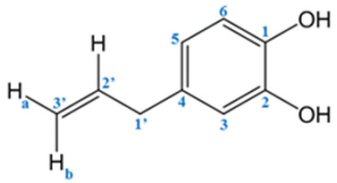


Fig 6. ^{1}H-NMR spectrum of the demethylated eugenol

Table 2. Position of ^{1}H-NMR signals of hydroxychavicol in $CDCl_3$ solvent



H Atom	Σ H	δ_H 500 MHz (ppm) (multiplicity, *J* (Hz))
1/2-OH	1	6.04 (*s*)
3	1	6.71 (*d*, 2.0)
5	1	6.60 (*dd*, 7.8, 2.0)
6	1	6.79 (*d*, 7.8)
1’	2	3.26 (*d*, 6.5)
2’	1	5.92 (*ddt*, 16.9, 10.4, 6.5)
3’a	1	5.03 (*m*)
3’b	1	5.05 (*ddt*, 16.9, 3.2, 3.2)

In general, the yield obtained are smaller than the reported demethylation of 2-acetyl-5-methoxy-1,2,3,4-tetrahydronaphtalene by Vera and Banerjee [8] using BBr_3 in CH_2Cl_2, which is 46%. This is due to BBr_3 that has better Br leaving group as compared to Cl leaving group in $AlCl_3$. The demethylation yield is also lower than that using $AlCl_3$ in DMS [9] with an analogous compound (ostenol), which is 62%. In this case, the number and the position of substituents on the aromatic ring greatly affect the demethylation on aryl methyl ether (Ar-O-CH_3). Demethylation of eugenol has also been investigated by Kraft and Eichenberger [22] using lithium chloride (LiCl) in DMF, giving 50% yield. Several ways have been reported to increase the yield of demethylation using $AlCl_3$, such as (1) adding thiourea to increase the nucleophilicity of $AlCl_3$ [14], the same function as Et_3SH reported by Gopalakrishnan et al. [9]; (2) increasing the equivalent of $AlCl_3$ [15-16], and (3) using microwave heating instead of the conventional one [15].

Monitoring by TLC using *n*-hexane-EtOAc (8:2) eluent of the crude product produces 2 spots, at R_f ~ 0.25 and R_f ~ 0.53, respectively. The first spot is the demethylation product and the second is the residual eugenol. Purification of these compounds was successful using FCC with the same eluent.

^{1}H-NMR spectrum of the demethylated eugenol is shown in Fig. 6 and the analysis is summarized in Table 2. There are 8 signals similar to hydroxychavicol spectrum as reported by Villegas et al. [10]. There are also 5 insignificant signals (1.28, 2.20, 2.66, 3.51, and 3.87 ppm). The use of CH_2Cl_2 p.a reduces impurity signals to 3 signals (1.28, 1.85, and 3.88 ppm).

Wide signal in 6.04 ppm indicates the presence of a hydroxyl proton. Aromatic protons give three signals in the 6.60, 6.71, and 6.79 ppm with coupling constants of J_{ortho} = 7.8 Hz and J_{meta} = 2.0 Hz. The analysis of coupling constants shows that the first signal comes from protons in *ortho* position to the allyl substituent (C5), the second signal comes from the protons in *ortho* to the allyl and hydroxyl substituents (C6), and a third signal from the protons in *meta* position with the

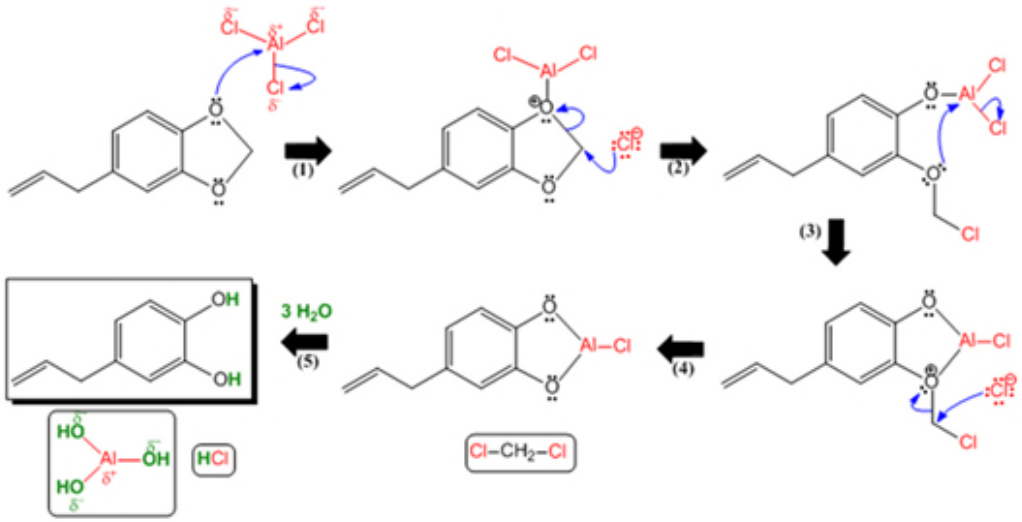


Fig 7. A proposed safrole demethylenation mechanism using $AlCl_3$

Table 3. Yield of safrole transformation into hydroxychavicol

No.	$AlCl_3$ (mmol)	Safrole (mmol)	Hydroxychavicol (mmol)	Yield (%)
1	5.26	0.8488*	0.1691	19.93
2	5.09	0.8180*	0.2264	27.92

Remarks: * (isolated safrole, 55.02% purity)

respect of the allyl substituent (C3). The positions of these three signals are relatively upfield due to the influence electron donation from the hydroxyl group.

In 3.26 ppm there are two proton signals with integration of 2, which come from methylene groups (C1'). The signal is relatively downfield than it should (~2.00 ppm) due to withdrawing electrons of the aromatic ring and the vinylic clusters. The vinylic proton gives 3 signals in 5.03, 5.05, and 5.92 ppm with coupling constants J_{trans} = 16.9 Hz, J_{cis} = 10.4 Hz, and J_{gem} = 3.2 Hz, respectively. The first and the second signals come from 2 overlapped geminal protons (C3'a and C3'b), therefore, only the C3'b coupling constants that can be calculated. The third signal originates from vinylic proton, next to the methylene group with relatively downfield chemical shift due to additional anisotropic effect of the aromatic ring. Determination of the splitting pattern of the proton signals and coupling constants in aromatic and allylic protons can be accurately interpreted using a tree diagram. Demethylation process proved to be successful as the ^{1}H-NMR of the methoxy methyl signal is absent at 3.86 ppm with an integration area 3; this signal belongs to eugenol [23].

Hydroxychavicol Derived from Demethylenated Safrole

Hydroxychavicol can also be synthesized from safrole demethylenation, by replacing the methylene ($-CH_2-$) with 2 hydrogen atoms. In general, the principle of safrole demethylenation does not much different from eugenol demethylation, as well as the reagents used. Demethylenation in this study used 6 equivalents $AlCl_3$. This was done to break the 2 sigma C-O bonds in the safrole molecule. A proposed demethylenation mechanism is provided in Fig. 7.

Demethylenation on safrole begins with nucleophilic attack of the lone pair of oxygen atom in the safrole to chloride atom in $AlCl_3$ and the chlorine leave as a good leaving group **(1)**. Chloride ion which is a weak nucleophile attacks the methylene carbon that has partial positive charge and opens the ring **(2)**. Furthermore, the lone pair of the oxygen in *meta* position to the allyl substituent re-attack the Al **(3)**. The released chloride ion attacks the methylene carbon that has high partial positive charge due to the pulling effect by the two electronegative atoms. The chloride is released as dichloromethane molecule. This attack resulted in formation of a five-membered ring O-Al-O **(4)**. Upon the completion of demethylenation step, the complex is decomposed with cold water **(5)**.

S_N2 reaction in safrole demethylenation step also went relatively slow. However, the process is much faster as compared with the eugenol demethylation because it involves a ring-opening reaction in the safrole molecule.

Table 3 shows that demethylenation of the isolated safrole produce gives 20% and 28% yield, or 24% in average. The result is lower than that reported by Villegas et al. [10], i.e. 55%, using a reagent $BF_3{\cdot}O(C_2H_5)_2$ in anhydrous 1,4-dioxane. This is due to the low content of safrole (55%); indicating that the impurities directly lead to side reactions with $AlCl_3$ and disrupt its interaction with safrole. Demethylenation conducted by Catalan et al. [12] gives 57% yield.

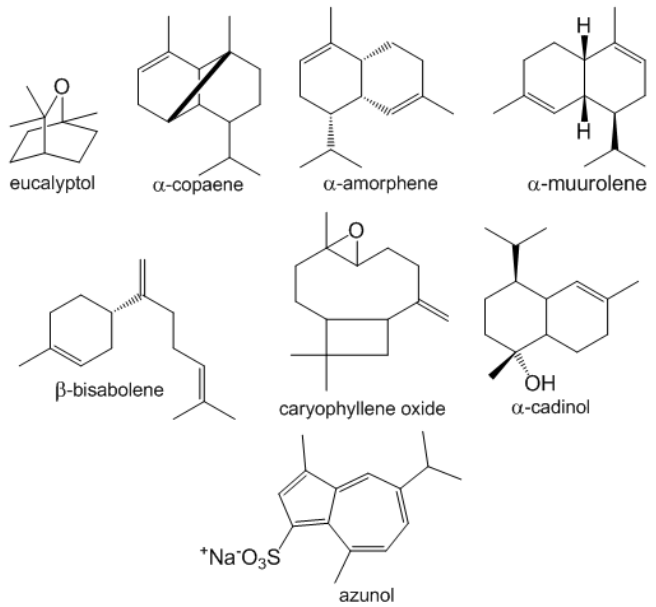


Fig 8. Chemical structures of the impurities found in purified *lawang* oil

However, prior the demethylation, a nitro group (NO_2) is attached to deactivate the aromatic rings in the C5 atom of safrole molecule, forming a nitrosafrole. According to Villegas et al. [10], the formation of a catechol will be facilitated in the presence of electron-withdrawing group ($-NO_2$) in the molecule.

Based on the GC-MS chromatogram, there are 7 main impurities (relative area > 1%) in the purified safrole isolated from *lawang* oil, namely caryophyllene oxide (6.05%), eucalyptol (4.02%), α-muurolene (3.11%), α-amorphene (2.30%), α-cadinol (2.20%), α-copaene (2.05%), β-bisabolene (1.53%), and azunol (1.28%) (Fig. 8). The main functional group in these compounds is alkene, which is practically inert to $AlCl_3$. However, epoxide group (in caryophyllene oxide), the ether group (in eucalyptol), and hydroxy group (in α-cadinol) are likely to react with $AlCl_3$ and can reduce the effective amount of the Lewis acid used for demethylenation. This effect account partially for the very low yield obtained. Increasing the amount of $AlCl_3$ to more than 2.5 equivalents is expected to increase the yield.

Monitoring by TLC using *n*-hexane-EtOAc (8:2) eluent of the crude product gave 3 spots at R_f ~ 0.21, R_f ~ 0.40, and R_f ~ 0.87, respectively. The first spot was hydroxychavicol, based on the R_f as previously reported. Therefore, ^{1}H-NMR analysis was not necessarily done. The second spot (R_f ~ 0.40) was not identified. The third spot belonged to the residual safrole (R_f ~ 0.84). FCC purification using the same eluent successfully separated these three constituents.

CONCLUSION

Isolation of eugenol from clove oil by alkaline extraction gave 71% yield, while the isolation of safrole with the same method followed by purification using PTLC gave 7% yield. Demethylation of the isolated eugenol and demethylenation of the isolated safrole using $AlCl_3$ has secured hydroxychavicol products as characterized by ^{1}H-NMR analysis. The hydroxychavicol obtained from eugenol demethylation was 28%, while that from safrole demethylenation was 24%.

Isolation of safrole still needs to be optimized to improve the purity. Effective demethylation and demethylenation reagents still need to be found to obtain higher yield.

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