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The Comparison of Three Different Methods on Extraction of Cigarette Butt as Natural Insecticide

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Abstract. Cigarette butt is potential to be used as an insecticide due to the present of alkaloid compounds in high yield, especially nicotine and eugenol. To maximize the extraction process of cigarette butt, the study related to the effect of extraction method to the effectivity of extraction process should be performed. The main purpose of this study was to identify the effect of extraction methods on the total yield and the nicotine and eugenol content of the extract of cigarette butt. In this study, maceration (**CB-1**), soxletation (**CB-2**), and microwave-assisted extraction (MAE) (**CB-3**) methods were conducted and compared the yield and the nicotine and eugenol content of the extracts by LC-MS. The results showed that **CB-3** has the highest yield (26,77%), which compared to **CB-1** and **CB-2**, which are 11.72% and 14.56%, respectively. Furthermore, the yield of extracted nicotine and eugenol from the samples were much higher in **CB-3**, which are 3.77% and 0.28%, respectively. To conclude, MAE method was more effective to extract the cigarette butt compared to maceration and soxletation method. This method was also reduced the time consuming and the volume of the solvent used in the extraction process which follow green chemistry rule. Thus, MAE method is potential to be used in the extraction process of cigarette butt in order to produce the natural insecticide.

Introduction

Nowadays, many researchers concern on the use of synthetic pesticides due to the negative impact of the usage in the agriculture (Ndakidemi et al., 2016; Oves et al., 2017). Some research found that the residue of synthetic pesticide in the plant and ground can affect the human health as well as the ecosystem, thus the use of synthetic pesticide should be reduced (Curl et al., 2020; Jallow et al., 2017; Mesnage & Séralini, 2018; Nicolopoulou-Stamati et al., 2016). One of the alternative solutions in the pest management is the use of other plants which can act as pesticide, which called biochemical pesticide or natural insecticide (Korunić et al., 2016; Mossa et al., 2018; Reddy & Antwi, 2016; Veer & Gopalakrishnan, 2016). One of the plants that has been proved to effectively decrease the presence of some pests is tobacco plant, *Nicotiana tabacum*, due to the high nicotine and eugenol content in its leaf (Ahmed & Karim Ahmed,

2018; Akyazi et al., 2018; Andjani et al., 2019; Telles et al., 2020).

In many countries, tobacco plant has been used in cigarette production as the main ingredient. According to WHO, the estimated prevalence of smoking among those aged 15 or more is 31% in 2017 (WHO, 2019). The smoking activity not only has a bad effect on the health of the smoker (active and passive) (Aboulmaouahib et al., 2018; Jiang et al., 2020; Karanasos et al., 2020; Lane et al., 2016; Sliwinska-Mosson & Milnerowicz, 2017), but also has negative impact for the environment due to the toxicity of the cigarette butt (Araújo & Costa, 2019; Torkashvand & Farzadkia, 2019). Cigarette butt contains similar composition to a dried tobacco leaf, such as nicotine, phenol, and eugenol (Syifa et al., 2020). Nicotine is an alkaloid which toxic to some organism thus can be use as natural insecticide (Villaverde et al., 2016). Some studies report that cigarette butt extract

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showed remarkable activity as insecticide (Fagerström, 2013; Murugan et al., 2018).

To use cigarette butt as insecticide, it has to be prepared by extraction process in the solvent. One of the important factors in extraction process is the extraction method, because it can affect the quality and quantity of the extract (Latifi et al., 2015). Maceration is one of extraction methods which is generally used to extract cigarette butt due to its easiness and not use much energy (Ćujić et al., 2016). However, this method generally gives a low yield and it takes quite long time in the extraction process, around 3-6 days. To optimize the extraction process, the use of the other methods is needed, which can extract the sample faster and in higher yield than maceration (Capriotti et al., 2014). Nowadays, many kinds of extraction methods have been developed which have some excellencies such as reducing the solvent amount, lowering the time consuming, or saving the energy (Da Porto et al., 2013). One of the new methods is Microwave-assisted extraction (MAE), which use the energy of microwave oven to extract the chemical component in the sample (Dahmoune et al., 2015; Herbert et al., 2006; Vinatoru et al., 2017).

This study has been conducted to compare three methods in extraction of cigarette butt, namely maceration, soxletation, and microwave assisted extraction (MAE) methods. All resulted extracts were calculated their yields, evaluate their phytochemical content, as well as analyze the chemical compositions by Liquid Chromatography - Mass Spectrometry (LC-MS). This report is an useful information in the use of cigarette butt as natural insecticide in agricultural field.

Experimental

Materials

The materials used in this study were absolute ethanol, aquadest, cigarette butt, filter paper Whatmann 40, TLC plat, Mayer reagent, Dragendorff reagent, Wagner reagent, and aluminium foil.

Equipment

The apparatus used in this study were Erlenmeyer, domestical microwave (Sharp Microwave Oven R21A1 22 L), maceration apparatus, soxletation apparatus, magnetic stirrer, analytical balance, evaporator, and LC-MS/MS QToF instrument, and glassware commonly used in laboratories.

Procedures

Cigarette butt sample was collected from some restaurants and hotels. The sample was prepared by separating its dried tobacco, then it was cleaned from other contaminant, and it was air-dried and grinded. Crude

extracts were prepared in ethanol absolute by maceration, soxletation, and microwave-assisted extraction methods.

Maceration. 20 grams of sample was placed in a dark flask, adding 200 mL of ethanol absolut as solvent. The mixture was stirred for 10 minutes and placed in dark place for 48 hours. The mixture was filtered off, and was re-macerated until it was exhausted. The filtrate was evaporated and air-dried (**CB-1**).

Soxletation. 20 grams of sample was placed in a thimble on the soxlet apparatus, and 200 mL of ethanol was poured in its round-bottom flask. The system was put in refluxing for 12 hours until the sample was exhausted. The solution was evaporated and air-dried (**CB-2**).

MAE. 20 grams of sample was placed in the round bottom flask, and 200 mL of ethanol absolut was poured. It was then placed in microwave apparatus which has been modified for extraction process. MAE was performed using the power of 100 watt for 8 minutes. The mixture was filtered off and the residue was extracted again two more times, until it was exhausted. The filtrate was evaporated and air-dried (**CB-3**).

All extracts (**CB-1**, **CB-2**, and **CB-3**) were evaluated their alkaloid, saponin, terpenoid/steroid, flavonoid, and tannin content according to the methods by Yadav et al. (2016), and analyzed their chemical content by LC-MS Shimadzu Mariner HP 5972.

Result and Discussion

The yield of extracts **CB-1**, **CB-2**, and **CB-3** are shown in Table 1. The results indicate that extraction methods affect the yield of the extraction process. Compared to maceration and soxletation methods, microwave-assisted extraction (MAE) method was the best extraction method which can result around two-fold weight of extract of both in maceration and soxletation process. Moreover, based on the time process, the MAE only need 3 x 8 minutes of extraction. It was very sort time compared to maceration (2 x 48 hours) and soxletation methods (12 hours). Other studies related to MAE also report that this method was not only higher the quantity of extract, but also consumed less solvent and time saving (Dahmoune et al., 2014, 2015).

Table 1. The Yield of Extraction

Sample	Weight of sample (g)	Weight of dried extract (g)	Yield (%)
CB-1	20.0058	2.3446	11.72
CB-2	20.0034	2.9125	14.56
CB-3	20.0048	5.3545	26.77

Table 2 shows the result of phytochemical screening of all extracts. All extracts contain alkaloid, terpenoid, flavonoid and tannin compounds, but did not contain saponin, which correlated with previous reports about the phytochemical content of tobacco leaf extract (Nunes & Miguel, 2017; Rawat & Mal, 2018; Shekins et al., 2016).

Table 2. Phytochemical Screening of Extract

Phytochemicals	CB-1	CB-2	CB-3
Alkaloid	+	+	+
Saponin	-	-	-
Terpenoid	+	+	+
Flavonoid	+	+	+
Tannin	+	+	+

Table 3. Chemical composition of all extract by LC-MS/MS

Compounds	CB-1 (%)	CB-2 (%)	CB-3 (%)
4-Hydroxy-3-butylphthalide	6,36	6,19	2,14
Ethyl-5-ethoxy-2-hydroxy benzoate	6,84	7,39	7,09
Eugenol	1,45	1,32	1,24
Nicotine	16,47	21,72	14,07
Stigmastan-3,6-dion	3,16	6,93	4,70

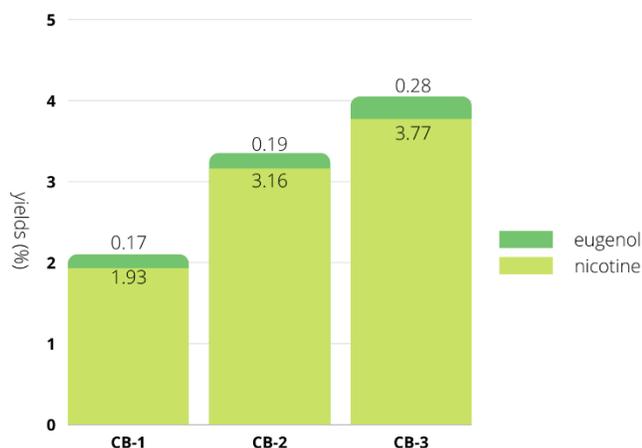


Figure 1. The yield of nicotine and eugenol from the samples.

The LC-MS/MS analysis of all extracts showed that all samples contain nicotine and eugenol as the major components which both have been reported to act as natural insecticides for some pests (Andjani et al., 2019; Xu et al., 2015). The percentage of five major compounds of all extracts were shown in Table 3.

Eventhough the **CB-3** has the lowest nicotine and eugenol contents, the yield of nicotine as well as eugenol from the sample were much higher in **CB-3**, as shown in Figure 1. Thus, based on the results, microwave-assisted extraction method was the best method for the extraction of cigarette butt compared to maceration and soxletation methods.

Conclusion

As conclusion, the MAE method performed the best result in the extraction of cigarette butt in term of the yield, which is 26.77%, compared to maceration and soxletation methods. The extracts contain nicotine and eugenol as the major components, which potent to be used as a natural insecticide. Thus, further study should be performed in order to confirm its effectivity as an insecticide for specific pests.

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