

# Phytochemical Screening of *Etilingera elatior* (Torch ginger) Cultivated on Different Dosage of Biochar

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## Abstract

**Objective:** This study was carried out with an objective to investigate the impact of biochar on phytochemical composition in plant especially *Etilingera elatior* cultivated on different dosage of biochar. **Methods:** *Etilingera elatior* was cultivated on the pot with 20 cm diameter and 35 cm height. 3 replicates for pots of *Etilingera elatior* was cultivated and label as 0%, 5% and 20%. The ordinary farm soil without biochar (0%) as control, fertilized soil with biochar; 5% and 20%. In a net house and watered twice daily. The gas chromatography mass spectrometry analysis was performed by using a non-polar BPX-5 capillary column with an initial temperature of 50°C hold for five minutes and then increased to 300°C at a rate of 5.0°C per minutes and hold 10 minutes. The biochar samples were analysed using an ATR-FTIR equipped with diamond crystal, controlled by OMNIC software (Thermo Nicolet Analytical Instruments, Madison, WI). A flat tip powder press was used to achieve even distribution and contact. **Results:** The result showed significantly increased in the phytochemical composition with increase in the biochar concentration. At 0% phytol (13%), Hexadecanoic acid (9.76%), Neophytadiene (6.51%), coumarin (5.65%), precocene (5.27%) and caryophyllene (4.59%). At 5% are Dihydrocucurbitacin (13.69%), Niacinamide (11.02%),  $\alpha$ -Limonene (10.01%), Phyrachen (9.23%), Phytol (7.24%) and Neophytadiene (5.75%) and at 20% Linoleic acid (39.98%), 2-pinen-4-ol (12.32%), Hexatriacontyl pentafluoropropionate (6.89%), Benzofuran (5.12%), Acethophenon (4.41%) and furfural (4.03%). **Conclusion:** Application of biochar on soil can increase nutrient availability and enhance the development of phytochemical composition in plants. Without biochar, the chemical composition *Etilingera elatior* extract was slightly low. At 5% and 20% biochar, some compounds are increasing and new were obtained compared to 0% biochar. Most of the compounds known to be secondary metabolite which are rich in medicinal values. **Thus, biochar could be used to increase the quantity and quality of phytochemicals in plant especially medicinal plants.**

Keywords: phytochemical, biochar, ethanolic extract, sod

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## Introduction

Biochar (BC) is a carbon-rich material produced by pyrolysis. In recent decade, it is widely described as a soil booster, improving soil and invariably plant quality. The main reason for the positive impact on soil properties, plant and microbial ecosystem is a direct Biochar influence on soil physical-chemical properties, as well as the nutrients available contents, and its ability to release nutrient slowly into soil solution [1]

Pyrolysis and composting technologies had introduced biochar into global agriculture attention due to its benefits [2,3,4]. The elemental composition of Biochar consists of carbon, nitrogen, hydrogen, potassium, and magnesium all of which can serve as major nutrients in plant growth. thereby improving soil physicochemical and biological properties. Biochar can positively or negatively affect the soil microbial growth to alter the agricultural environment [5]. It is a very stable, carbon-based material obtained from the pyrolysis of biomass under anaerobic [6].

*Etilingera elatior* metabolites, possess various pharmacological activities. The plant is also used as vegetable to prevent cardiovascular disease, diabetes and cancer. [7] Several studies had been evaluated in the activities of plant extract such as antibacterial and antifungal, [8] as well as the antioxidant activities [8,9]. Long-term oxidative stress contributes to pathogenesis of chronic diseases such as diabetes, kidney disease and cancer diseases. There was also clinical potency of plant as food supplement on diabetes patients [10,11].

Torch ginger (*Etilingera elatior*) belongs to the family of *Zingiberaceae* it is normally grown in Asia and found common in Samarahan, Sarawak Malaysia. It was generally used as herb [12]. It was also reported to contained enormous metabolite as well as antioxidant properties [13]. *Etilingera elatiorextract* expressed various pharmacological properties; antioxidant, antimicrobial, antifungal, tyrosinase inhibition, cytotoxic and has hepatoprotective activities[14] In this study, it was hypothesized that the use of biochar to *Etilingera elatior* with different dosages might affect the type of secondary metabolite produced from this medicinal plant. Therefore, this present study aimed to evaluate the effect of biochar at two dosages [15] and 20% of fresh mixture weight) on the type of secondary metabolite extracted from this medicinal plant *Etilingera elatior*.

## Materials and Method

### Cultivation of *Etilingera elatior*

*Etilingera elatior* was grown in the pot with 20 cm diameter and 35 cm height. 3 replicates for pots of *Etilingera elatior* was cultivated and label as 0%, 5% and 20%. The soil used was obtained from farm with little or non-fertile soil (0%) fertilized soil with biochar; 5% and 20%. The pots are placed in the netting house and the growth of *Etilingera elatior* was monitored. Watering process was done twice a day. The biochar used in this study were purchased from Black Owl Biochar Products. The dosage of biochar used was calculated over the weight of soil. The soil treatments are as follows:

- a) Non-fertilized soil (0%), b) non-fertilized + biochar (5% and 20%)

## Sample extraction

Phytochemical was extracted according to [16,17,18] with some adjustments. 100 gm of the dry leaf powder of *Etlingera elatior* were weighted, transferred to a flask, soaked with ethanol until the powder was fully immersed and incubated overnight. The extracts were then filtered through Whatman filter paper No.1 along with 2 gm sodium sulfate anhydrous to remove the traces of water in the filtrate. Before filtering, the filter paper along with sodium sulphate was wetted with 95% ethanol. The filtrates were then air dried and subjected to gas chromatography-mass spectrometry analysis.

## Fourier Transform Infrared (FTIR)

The biochar samples were analysed using an ATR-FTIR equipped with diamond crystal, controlled by OMNIC software (Thermo Nicolet Analytical Instruments, Madison, WI). A flat tip powder press was used to achieve even distribution and contact. All spectra were collected at 12 scans with a resolution of 4 cm<sup>-1</sup> in the range 4000–670 cm<sup>-1</sup>. The spectrum of each sample was recorded against a fresh background spectrum recorded from the bare ATR crystal. The ATR crystal was cleaned with ethanol.

## Gas Chromatography Mass Spectrometry (GCMS) analysis

The gas chromatography mass spectrometry analysis was performed by using a non-polar BPX-5 capillary column with an initial temperature of 50°C hold for five minutes and then increased to 300°C at a rate of 5.0°C per minutes and hold 10 minutes. The temperature of the injector and detector were set at 320°C respectively. 1µl of the fractions was diluted in 100µl hexane was introduced into the gas chromatography. The gas used as the carrier was Helium. Interpretation of mass-spectrum was conducted using the database of National Institute Standard and Technology (NIST17). The spectrum of the secondary metabolites components was compared with the spectrum of known components stored in the NIST library. The name, molecular mass and structure of the components of the test materials were ascertained.

## Results and discussions

The Fourier Transform Infrared spectra shown in Figure 1. The spectra with pronounced bands at 1591 cm<sup>-1</sup> corresponding to carboxyl group of protein compounds. Broad bands at 1700 cm<sup>-1</sup> due to stretching of C-O bond in polysaccharide. However, polysaccharide would have degraded during the process of biochar, whilst the phosphate content in biochar increase. The phosphate band can be observed at 1029 cm<sup>-1</sup>. These findings confirm that the biochar can be used to improve the nutrient and metabolites in plants such as *Etlingera elatior*. This also agrees with the report of [19] partial detachment of the functional group leads to the formation of unpaired negative charges such as carboxyl and hydroxyl group that have the ability to attract positive charge thus releasing hydrogen and oxygen contributing to increase carbon content which are some important properties of biochars remediation of metal contaminated soil.

The gas chromatography's chromatograms of *Etlingera elatior* obtained from ethanolic extract are shown in Figure 2. The chromatograms demonstrate different peaks for different dosage of biochar (Figure 2(a)-(c)). Different pattern of chromatogram was observed on different dosage of biochar applied. The major compounds with their percentage area (PA%) are summarize in Figure 3. The results revealed that phytol (13%), Hexadecanoic acid

(9.76%), Neophytadiene (6.51%), coumarin (5.65%), precocene (5.27%) and caryophyllene (4.59%) were among the major compounds identified from *Etilingera elatior* ethanol extract on 0% biochar (Figure 3(a)). Increasing biochar dosage to 5%, the identified compounds shows that, the 6 major compounds are Dihydrocucurbitacin (13.69%), Niacinamide (11.02%),  $\alpha$ -Limonene (10.01%), Phyrahen (9.23%), Phytol (7.24%) and Neophytadiene (5.75%) shown in Figure 3(b). In 20% biochar the major compounds of *Etilingera elatior* are Linoleic acid (39.98%), 2-pinen-4-ol (12.32%), Hexatriacontyl pentafluoropropionate (6.89%), Benzofuran (5.12%), Acethophenon (4.41%) and furfural (4.03%) shown in Figure 3(c). Most of the compounds known to be secondary metabolite which are rich in medicinal values.

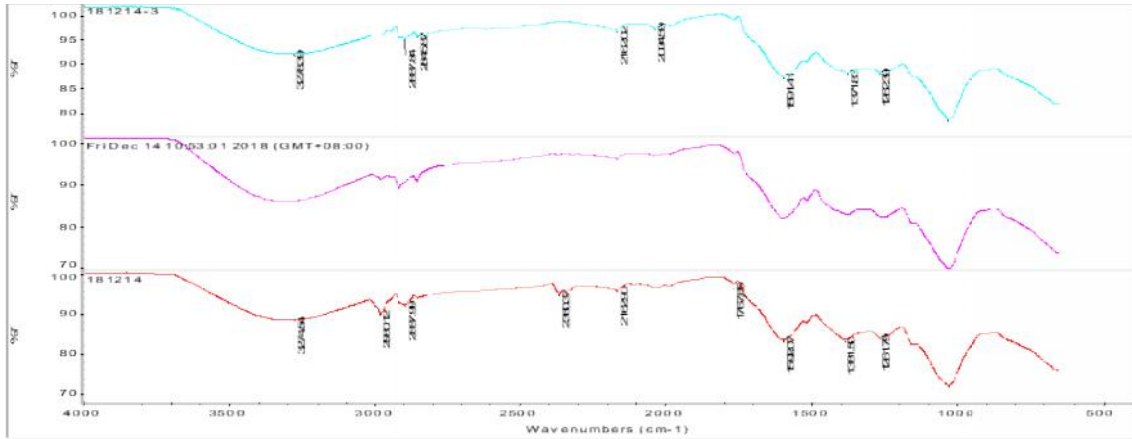
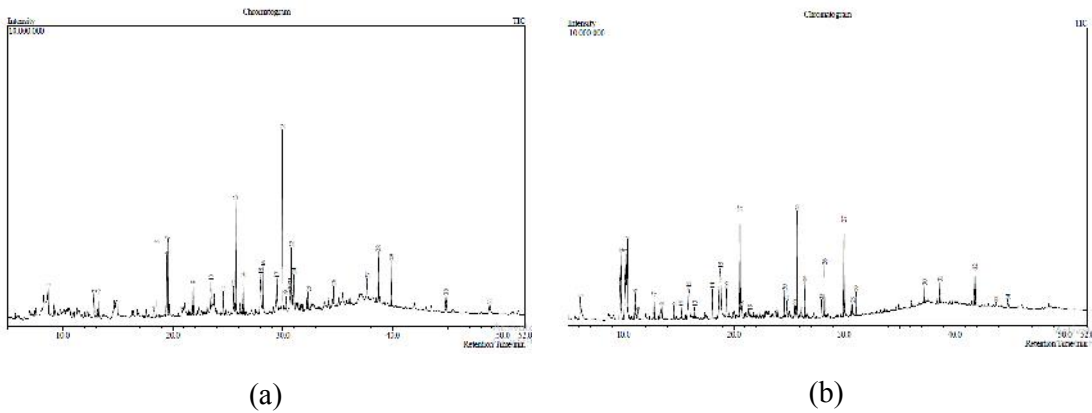
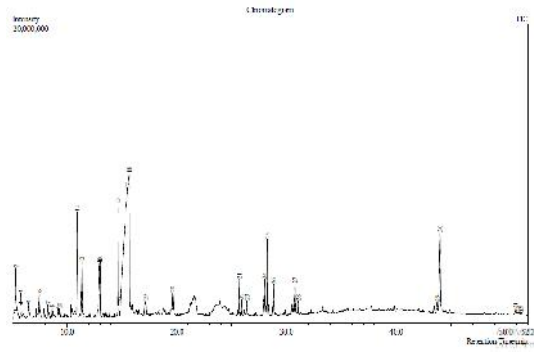


Figure 1. Stack spectra of biochar.



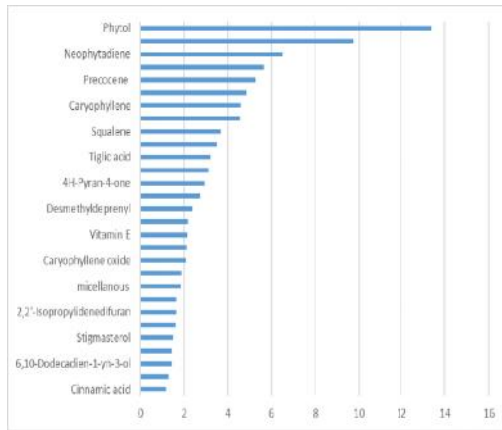
(a)

(b)

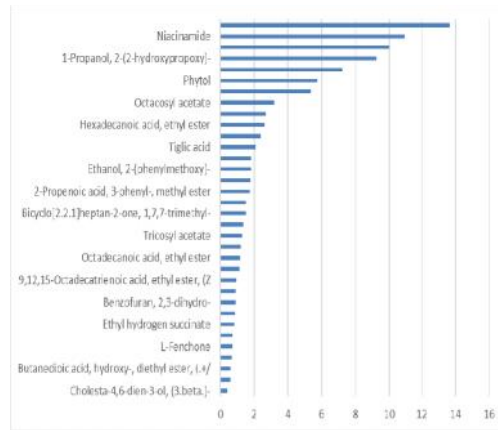


(c)

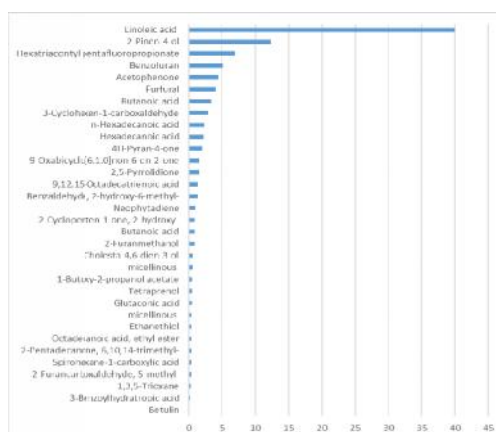
Figure 2. Chromatogram of phytochemical from *Etilingera elatior* at different dosage of biochar; (a) 0% biochar, (b) 5% biochar and (c) 20% biochar.



(a)



(b)



(c)

Figure 3. Peak area percentage of compounds extracted from *Etilingera elatior* at different dosage of biochar; (a) 0% biochar, (b) 5% biochar and (c) 20% biochar.

### Conclusion

Application of biochar on soil can increase nutrient availability and enhance the development of phytochemical composition in plants. Without biochar, the chemical composition *Etilingera elatior* extract was low. At 5% and 20% biochar, some compounds are increasing and new compounds are develop compared to 0% biochar. This suggest that the biochar not only able to increase the growth rate of plants but also the nutrients of the plants.

**Ethic : NA**

**Consent : NA**

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### Conflicts of Interest

The authors declare that there is no conflict of interests regarding the publication of this paper.

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