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Research Article

ANTIOXIDANT ACTIVITY OF ALKALOID FRACTIONS OF *LITSEA CUBEBA* LOUR. FRUITS

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ABSTRACT

Objective: The oxidation induced by free radicals results in many degenerative disease. The purpose of this study is to determine antioxidant activities of alkaloid fractions of *Litsea cubeba* (Lour.) fruit.

Methods: *L. cubeba* Lour. was extracted by maceration. Ethanol extract was fractionated with liquid-liquid extraction using n-hexane and chloroform at pH 3, 7, 9, and 11 to obtained alkaloid fractions. Antioxidant activity for extract and fractions was determined with 1,1-diphenyl-2-picrylhydrazyl.

Results: The IC_{50} of extract and fractions was 219.43±0.43, 242.97±0.93, 92.38±0.17, 40.84±0.04, 103.83±3.29, and 103.75±0.42 µg/mL, respectively.

Conclusion: The results reveal that alkaloid fractions of *L. cubeba* fruits have very strong antioxidant potential. Our further study is to isolate the alkaloid compounds.

Keywords: Antioxidant, *Litsea cubeba*, Fruits, Alkaloid, Fractions.

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INTRODUCTION

Oxidation is an important process (normally) in living organisms. Free radicals are producing from metabolism pathway process or environmental sources which interact with biological system. Reactive species are molecules which have an electronic unstability and most reactive. Reactive oxygen species are the biggest sources of a primary catalyst which initiate the process of oxidation *in vivo* and *in vitro* and produce oxidative stress. Oxidative stress products when reactive forms of oxygen are produced faster than they could be safely neutralized with antioxidant mechanisms and/or from a decrease in antioxidant defense. The uncontrolled production of oxygen free radicals and the unrateable system of antioxidant capability in protection results in the cause of many diseases, such as cancer, diabetes, heart diseases, Alzheimer's, and aging [1-6].

Attarasa *Litsea cubeba* (Lour.) is a plant from Lauraceae family which contains much essential oils which used as antidepressant, anti-inflammation, antioxidant, pesticide, antimicrobial, anticancer, and neuropharmacology [3]. Methanol extract from attarasa fruits showed to be active on cervix cancer (HeLa cell lines) which causes apoptosis through activation of caspase 3/7 [3,4]. There are more than 40 isoquinoline alkaloids that contained in *Litsea* genus which are active as antibacterial agents against *Staphylococcus aureus* [5]. The heartwoods of *L. cubeba* contained a high level of phenolic and flavonoid and found to be active as antioxidant [6]. The aim of this study was to determine the antioxidant activities of alkaloids fraction of *L. cubeba* Lour. fruits.

METHODS

Plant and chemicals material

Fresh fruits of *L. cubeba* (Lour.) were collected from Balige subdistrict, Sumatera Utara province, Indonesia. *L. cubeba* (Lour.) was identified in Herbarium Medanense, Faculty of Mathematics and Natural Products, University of Sumatera Utara, and the voucher specimen was deposited in herbarium. Chemicals used were distilled water, 1,1-diphenyl-2-picrylhydrazyl (DPPH) (Sigma), and methanol (Merck).

Preparation of extract and fractionation

The air-dried and powdered fruits of *L. cubeba* (Lour.) (1 kg) were repeatedly extracted by cold maceration with ethanol 96% (3×3 d, 7.5 L) at room temperature with occasional stirring. The filtrate was collected and then evaporated under reduced pressure to give a viscous extract and then freeze-dried to dry [7-9]. Viscous extract was fractionated with n-hexane and continued with chloroform at pH 3, 7, 9, and 11 [10].

Free radical scavenging activity test

The DPPH assay was carried out according to the previous study with some modifications [11]. About 0.2 mM solution of DPPH• in methanol was prepared, and 100 µl of this solution was added to various concentrations of fractions at the concentrations of 25, 50, 100, and 200 µg/ml. After 60 minutes, absorbance was measured at 516 nm and the percentage of inhibition was calculated by comparing the absorbance values of the control and test samples [2,5].

$$\text{Percentage of inhibition} = \frac{\text{Abscontrol} - \text{Abstest}}{\text{Abscontrol}} \times 100\%$$

Statistical analysis

Data were expressed as mean±standard deviation which was analyzed using the SPSS 21 software.

RESULTS AND DISCUSSIONS

Antiradical activity

Antiradical activity of the plant was measured in terms of hydrogen-donating ability using DPPH which is a stable, nitrogen-centered free radical and produces deep purple color in methanol solution, and antioxidants either transfer an electron or a hydrogen atom to DPPH, thus neutralizing its free radical character [12]. Antioxidant assay with DPPH which is based on the ability of DPPH, a stable free radical, to decolorize in the presence of antioxidants, is a direct and reliable method for determining radical scavenging action [13] and has been largely used as a quick, reliable, and reproducible at *in vitro* antioxidant activity assay [14]. The reducing capacity of compounds could serve as a marker of potential antioxidant activity [15-18]. Alkaloids are compound which contains OH and NH functional group, and they could be donating their hydrogen to DPPH [19]. IC_{50} for each fraction is shown in Table 1.

Table 1: IC_{50} value of alkaloid fractions of *L. cubeba* fruit with DPPH assay

Treatment	IC_{50} (µg/mL)
n-hexane Fraction	219.43±0.43
Chloroform Fraction pH 3	242.97±0.93
Chloroform Fraction pH 7	92.38±0.17
Chloroform Fraction pH 9	40.84±0.04
Chloroform Fraction pH 11	103.83±3.29
Water Fraction	103.75±0.42

CONCLUSION

The result of this study showed that alkaloid fractions of *L. cubeba* fruit possess antioxidant activity.

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