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 obtain alkaloid fractions. Antioxidant activity for extract and fractions was determined with 1,1-diphenyl-2-picrylhydrazyl (DPPH) (Sigma), and methanol (Merck).

**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 produced 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 unreactable system of antioxidant capability in protection results in the cause of many diseases, such as cancer, 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 alkaloid 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].

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 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.
CONCLUSION

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

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