Phytochemical and Antioxidant Activity of *Blumea balsamifera* and *Cordyline fruticosa* Based on Ethnopharmacology Knowledge of Muara Tae Tribe, East Kalimantan

Nur Maulida Sari¹, Farida Aryani^{2,*}, Wartomo¹, Muhammad Fikri Hernandi¹, Erna Rositah³, Joko Prayitno¹

¹Department of Forest Product Processing; ²Department of Plantation Products Technology; ³Department of Forest Management, Samarinda State Agriculture Polytechnic, Kampus Gunung Panjang Jalan Samratulangi, Samarinda 75131, East Kalimantan, Indonesia.

Corresponding author*

faridaaryani@politanisamarinda.ac.id

Manuscript received: 15 January, 2023. Revision accepted: 14 March, 2023. Published: 23 March, 2023.

Abstract

Plant use as traditional medicine is still widely practiced in Indonesia. Muara Tae tribe people, West Kutai regency are one of the regions that still rely on *Blumea balsamifera* and *Cordyline fruticosa* plants as traditional medicine. This study aims to determine the potential of *Blumea balsamifera* and *Cordyline fruticosa* leaves as medicinal plants with phytochemicals and antioxidants. Phytochemical analysis was tested using Harborne and Kokate methods. Antioxidant activity was evaluated by DPPH radical scavenging assay with slight modification. The results of the phytochemical analysis showed that the extracts of n-hexane, ethyl acetate, and ethanol from the leaves of *Blumea balsamifera* and *Cordyline fruticosa* contained alkaloids, tannins, and triterpenoids. Antioxidant activity of *Blumea balsamifera* leaves extract showed that the n-hexane extract display an ability to inhibit DPPH free radical by 50% at 100 ppm concentration, while ethyl acetate and ethanol extracts display an ability were 23.68 µg/mL and 17.59 µg/mL. Antioxidant activity of *Cordyline fruticosa* leaves sequentially were 73.72 µg/mL and 20.17 µg/mL. Based on the results, *Blumea balsamifera* and *Cordyline fruticosa* leaves sequentially were 73.72 µg/mL and 20.17 µg/mL. Based on the results, *Blumea balsamifera* and *Cordyline fruticosa* leaves sequential to develop as natural antioxidants.

Keywords: Blumea balsamifera; Cordyline fruticosa; DPPH; Ethnopharmacology; Phytochemical.

INTRODUCTION

Indonesia has unique flora and fauna that complement its, cultural and ethnic diversity. These ethnic groups occupy certain areas of the country (Sreekeesoon & Mahomoodally, 2014). Each ethnic group has its own lifestyle and traditions, including foods, herbs, and spices. It is also very capable of using and preserving organic and ecological diversity. Medicinal plants are used by the local community to treat illnesses, mainly due to health restrictions. The original knowledge of the use of medicinal herbs was probably passed down from generation to generation. This approach has so far succeeded in keeping knowledge alive (Yaseen et al., 2015). Therefore, local knowledge of medicinal plants has always been a source of research in testing the effects of plants and developing new therapeutic means (Bolson et al., 2015).

Indonesia has the second highest biodiversity in the world after the Amazon forests and a population of, people from more than 300 nationalities (Elfahmi et al., 2014). Most of the studies focused on the prevention of diseases or specific medicinal herbs in Indonesia (SILALAHI et al., 2014). Although Indonesian researchers have studied traditional knowledge about the use of plants as medicine, most studies have not been published in international journals.

About 30,000 medicinal plants grow and develop in Indonesia, which covers 90% of medicinal plants in the Asian region (Syamsiah et al., 2016). The use of traditional medicine is increasing with population growth, healthy lifestyles, and degenerative diseases (Triratnawati, 2016). Also up to 21,4% of the Indonesian population used traditional medicine to self-treat health problems. Herbal medicine and massage are the most common treatments offered by traditional healers in Indonesia (Peltzer & Pengpid, 2019). The consumption of traditional medicines in Indonesia increased by 5,4% per year. Efficiency, lower cost, easy availability, and dissatisfaction with traditional treatment methods are some of the reasons for choosing traditional treatment (Riptanti et al., 2018). Several studies in recent years about medicinal plants use, informed that plants were the best sources of natural antioxidants such as phenolics, alkaloids, and flavonoids compound (Zhao et al., 2018). An antioxidant agent from the plants was a huge resource to scavenging free radicals naturally. Medicinal plants also prevent oxidative stress, maintain health, and disease and delay aging processes (NGUYEN et al., 2018). Oxidative stress of human lipids is known to have various human health problems such as cardiovascular disease, cancer, diabetes, and others (Truong et al., 2019). Generally, antioxidants are divided into 2 types natural and synthetic antioxidants. Natural antioxidants are widely used as free radical inhibitors, while synthetic antioxidants had a negative effect with long-term used (Stoia & Oancea, 2022).

Muara Tae was local tribe exist in West Kutai, East Kalimantan, Indonesia. The Muara Tae community known as a sub-tribe of Dayak used medicinal plants as alternative drugs for healthcare treatment and diseases such as fever, diabetes, external wound, stomachache, and others. Several studies about potential plant use by Dayak people informed the plant used as natural antifungal, antioxidant, and antibacterial agents by Bentian Tribe (Kusuma et al., 2016). *Blumea balsamifera* and *Cordyline fruticose* were the plant's belief as medicinal plants by Muara Tae's people. These plants had only limited attempts to explore the biological properties of plants regards their uses as medicine by the local people. The present study aims to explore the potential of *Blumea balsamifera* and *Cordyline fruticosa* leaves extract for its antioxidant activity from the Muara Tae tribe in Indonesia (Figure 1).

MATERIALS AND METHODS

Plant collection

The leaves of *Blumea balsamifera* and *Cordyline fruticosa* were collected from Muara Tae Village, West Kutai, East Kalimantan, Indonesia. The samples were washed thoroughly with water to remove the extemporaneous and dried for about 3 days in the laboratory with air conditioning (A.C.) set for 20-25°C. The samples were kept in A.C. room to keep the moisture content stable and milled with a blender. The powdered samples were prepared for further analysis.



Figure 1. Morphology of Blumea balsamifera and Cordyline fruticosa Plants (Photo source: personal doc.).

Procedures

Maceration

About 30 gr powdered samples of *Blumea balsamifera* and *Cordyline fruticosa* were extracts with n-hexane, ethyl acetate, and ethanol solvent at room temperature with continuous shaking on a shaker for 48 hours. Following filtration of the suspension through Whatman paper No.2 (Maidstone, UK), the crude extracts of *Blumea balsamifera* and *Cordyline fruticosa* were evaporated in a rotary evaporator at 38-40 °C and put in a vacuum over near dryness to yield the plant extract.

Preliminary Phytochemical Analysis

The n-hexane, ethyl acetate, and ethanol extracts of *Blumea balsamifera* and *Cordyline fruticosa* leaves were subjected to preliminary screening of phytochemical such as alkaloids, flavonoids, tannin, steroids, triterpenoids, carbohydrate, and saponin using some following standard procedures (Harborne, 1998; Kokate, 2001).

Alkaloids determination: 5 mL of the n-hexane, ethyl acetate, and ethanol extracts of *Blumea balsamifera*

and *Cordyline fruticosa* leaves were added to 2 mL *Hydrochloride Acid*, then 1 mL of Dragendorff solution was added. The color changes in the solution indicated the presence of alkaloids (Kokate, 2001).

Flavonoids Determination: About 1 mL of the nhexane, ethyl acetate, and ethanol extracts of *Blumea balsamifera* and *Cordyline fruticosa* leaves were drops of 1% *Sodium Hydroxide*. The presence of yellow color at extracts solution and were colorless after the addition of 1% *Hydrochloride Acid* indicated the presence of flavonoids (Kokate, 2001).

Tannin Determination: 10 mL of the n-hexane, ethyl acetate, and ethanol extracts of *Blumea balsamifera* and *Cordyline fruticosa* leaves were added 1 % *Lead II Acetate*. The yellow precipitate reaction in the solution indicated the presence of tannin (Kokate, 2001).

Steroids Determination: 1 mL of the n-hexane, ethyl acetate, and ethanol extracts of *Blumea balsamifera* and *Cordyline fruticosa* leaves dropped about 10 *Acetic Acid Anhydride* and 2 drops of *Sulfuric Acid*, sequentially. The green or blue color changes in the solution indicated the presence of steroids (Harborne, 1998).

Triterpenoids Determination: 1 mL of the nhexane, ethyl acetate, and ethanol extracts of *Blumea balsamifera* and *Cordyline fruticosa* leaves dropped about 10 *Acetic Acid Anhydride* and 2 drops of *Sulfuric Acid*, sequentially. The red or purple color changes in the solution indicated the presence of steroids (Harborne, 1998).

Carbohydrate Determination: About 1 drop of Molisch solution were added to 1 mL of the n-hexane, ethyl acetate and ethanol extracts of *Blumea balsamifera* and *Cordyline fruticosa* leaves. Then 1 mL of *Sulfuric Acid* was added thourgh the tube glass wall, slowly. The formation of purple ring between 2 layers indicated the presence of carbohydrates (Harborne, 1998).

Saponins Determination: 10 mL of hot distilled water was added to 1 mL of the n-hexane, ethyl acetate, and ethanol extracts of *Blumea balsamifera* and *Cordyline fruticosa* leaves. The solution was then cooled and shaken vigorously (10 seconds). A stable froth upon standing for 10 minutes after adding 1 drops of *Hydrochloride Acid* 2N indicated the presence of saponins (Harborne, 1998).

Antioxidant Assay

Test of antioxidants using 5 concentration samples was grouped into 100 ppm, 50 ppm, 25 ppm, 12.5 ppm, and 6.25 ppm times of dilution, respectively. Further, 3 mg of *Ascorbic acid* was weighed, then dissolved in 1000 μ L of ethanol solvent and regarded as a positive control. While the ethanol solvent was used as a negative control. About 33 μ L sample was mixed in a glass tube with 467 μ L of ethanol added, and 500 μ L of 2,2-diphenyl-1-picryhydrazyl (DPPH) radical scavenging activity (Shimizu et al., 2001). The mixing of the sample was stopped while the volume reached 1000 μ L (1 mL). Samples were incubated for 20 minutes with minimum

light and A.C. set for 27-30 °C. The antioxidant activity was determined by decolorization of DPPH with a wavelength of 517 nm using a Spectrophotometer. Measurement was performed in the triplicate examination. The percentage of DPPH free radical was calculated using the following equation:

% DPPH radical scavenging activity =
$$\frac{\Delta control - \Delta sample}{\Delta control} \times 100$$
 (1)

The actual decrease in absorbance caused by the test was compared with positive controls. The values of IC_{50} (concentration giving 50% of inhibition) were calculated by a dose-inhibition curve over a linear range of by plotting the extract concentration and the corresponding washout effect (Tuldjanah et al., 2021).

RESULTS AND DISCUSSION

Plant extracts

Blumea balsamifera and Cordyline fruticosa leave were extracted using 3 different solvents n-hexane, ethyl acetate, and ethanol. Maceration is done with yielded 2.18-4.60% extracts on the basis of sample dry weight from Blumea balsamifera leaves, while Cordyline fruticosa leaves yielded 1.09-5.20% extracts (Table 1).

 Table 1. Yield of Blumea balsamifera and Cordyline fruticosa Leaves

 Extract.

Plants	Solvent	Extract yield (%)
Blumea balsamifera	n-hexane	2.18
	ethyl acetate	3.18
	ethanol	4.60
Cordyline fruticosa	n-hexane	1.09
	ethyl acetate	3.03
	ethanol	5.20

The results showed the ethanol extracts of *Blumea* balsamifera and *Cordyline fruticosa* produce an extractive content more concentrated than the n-hexane and ethyl acetate extracts.

Phytochemical screening

The n-hexane, ethyl acetate, and ethanol leaf extracts of Blumea balsamifera and Cordyline fruticosa were analyzed as the secondary metabolites. Plants known to contain multiple phytochemical molecules such as terpenoids, lignins, phenolics, vitamins, tannins, and others metabolites as antioxidant agents. Several studies showed many phytocompounds possess antidiabetic, anti-inflammatory antimicrobial, and activities (Campbell-Tofte et al., 2012). The pharmacological activities of secondary plant metabolites are known as large compounds as a source of medicinal agents (Muthukrishnan & Manogaran, 2018). Phytochemical analysis of the n-hexane, ethyl acetate, and ethanol leaves extracts of Blumea balsamifera and Cordyline fruticosa is presented in Table 2.

Compounds	Blumea balsamifera			Cordyline fruticosa		
	N-hexane	Ethyl Acetate	Ethanol	N-hexane	Ethyl Acetate	Ethanol
Alkaloids	+	-	+	+	-	+
Flavonoids	-	-	-	-	-	-
Tannins	-	+	+	-	-	+
Steroids	-	-	-	-	-	-
Triterpenoids	+	-	-	+	-	-
Carbohydrate	-	-	-	-	-	-
Saponins	-	-	-	-	-	-

Table 2. Phytochemical Analysis of Blumea balsamifera and Cordyline fruticosa Leaves Extract.

The phytochemical screening of *Blumea balsamifera* revealed the presence of alkaloids and triterpenoids in the n-hexane extract, and tannins in the ethyl acetate extract, while the ethanol extract contained alkaloids and tannins. The n-hexane extract of *Cordyline fruticosa* revealed the presence of alkaloids and triterpenoids, while the ethanol extract contained alkaloids and tannins.

The bioactivity potency of plants was indicated based on the secondary metabolites compounds. The presence compounds of tannins, flavonoids, and alkaloids are known as antitumor, antibacterial, antivirus also had antioxidant activity, antimicrobial and anticancer (Taşkın & Taşkın, 2017). Phenolic known as the largest compounds were found in plants and it's an important compound for free radical scavenging, also contains the hydroxyl group compound. Flavonoid, phenol, and tannin compound are known as the largest group of phenolic compounds and also had the abilities as antioxidant agents (Sen et al., 2013).

Antioxidant activity

Free radical DPPH scavenging of the n-hexane, ethyl acetate, and ethanol leaves extracts of *Blumea balsamifera* and *Cordyline fruticosa* was determined by its hydrogen donating abilities. The inhibition of *Blumea balsamifera* n-hexane leaves extracts displayed the ability to inhibit free radical DPPH formation by 50% at 100 ppm concentration, while the ethyl acetate and ethanol extracts by 77% and 81% at 50 ppm concentration, respectively. The antioxidant activity of the n-hexane, ethyl acetate, and ethanol leaf extracts of *Blumea balsamifera* is presented in Figure 2.

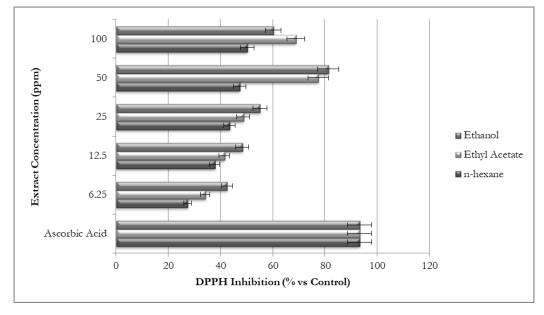


Figure 2. Antioxidant Activity of Blumea balsamifera leaves extracts against DPPH.

Several studies about *Blumea balsamifera* plants informed the plant extracts contained steroids, alkaloids, phenolic and saponins compounds used as traditional medicine for eczema, rheumatic, menstrual pain relief, flu, fever, asthma, diabetes cough, and diarrhea (Pang et al., 2014). Its also has biological activity as an antiinflammatory, anticancer, and antioxidant and also functions as an antimicrobial (Nessa et al., 2010).

The inhibition of *Cordyline fruticosa* n-hexane and ethyl acetate leaf extract displayed the ability to inhibit free radical DPPH formation by 45% and 56% at 100 ppm concentration, while the ethanol extracts by 76% at 50 ppm concentration. Antioxidant activity of the n-

hexane, ethyl acetate, and ethanol leaves extracts of *Cordyline fruticosa* is presented in Figure 3.

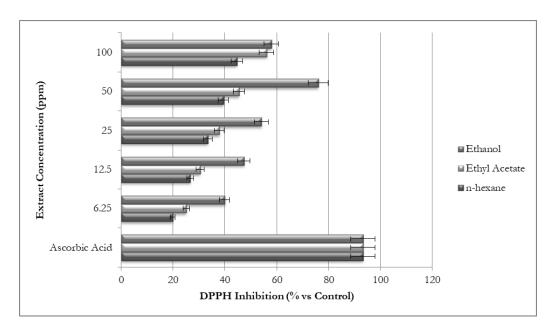


Figure 3. Antioxidant Activity of Cordyline fruticosa leaves extracts against DPPH.

Several studies about *Cordyline fruticosa* leaves plants, informed the plant contained saponins, tannins, flavonoids, polyphenol, and alkaloids compound which has biological activity as antioxidant, anti-inflammatory, antibacterial, anti-allergic, anticancer, and antivirus (Dyary et al., 2014). This plant is also known to accelerate the wound healing process, the function of tannins compound as astringents uses shrink skin pores and bleeding minor stopped.

The antioxidant activity of *Blumea balsamifera* and *Cordyline fruticosa* leaves extracts indicated plants had a source to inhibit free radical DPPH. Refers to the results, the IC₅₀ of the ethyl acetate and ethanol leaves extracts of *Blumea balsamifera* and *Cordyline fruticosa* extracts were determined as shown in Table 3.

Table 3. The IC_{50} of *Blumea balsamifera* and *Cordyline fruticosa* Leaves Extract.

Blumea balsamifera		Cordyline fruticosa		
Ethyl Acetate	Ethanol	Ethyl Acetate	Ethanol	
23.68±19.69	17.59±18.71	73.72±14.42	20.17±17.59	

The antioxidant activity of IC_{50} had a classification of the antioxidant activities compound based on the acquisition of the IC_{50} value as shown as Table 4 (Analianasari et al., 2022).

Table 4. The Antioxidant Activity of IC₅₀ Classification.

Inhibition Value (ppm)	Category
<50	Extreme
50-100	Strong
100-150	Moderate
150-200	Weak
>200	Fragile

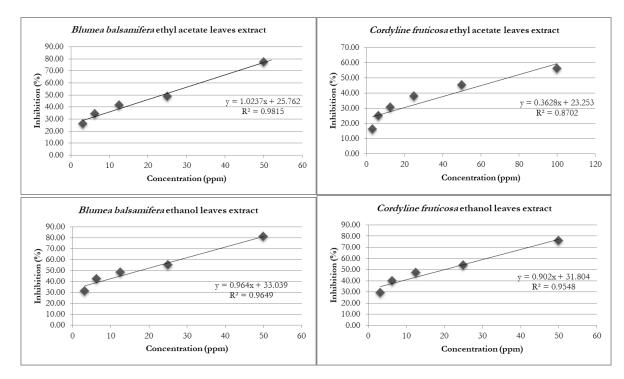


Figure 4. Antioxidant Activity of Cordyline fruticosa leaves extracts against DPPH.

Based on Figure 4, the linear equation value for *Blumea balsamifera* ethyl acetate extract y = 1.0237x+25.762, the calculation of the IC₅₀ value for *Blumea balsamifera* ethyl acetate extract obtained the following values: y = 1.0237x+25.762 (for y = 50), then the x value is 23.68 ppm. Furthermore, based on the calculation of the regression curve in the ethanol extract of *Blumea balsamifera*, the regression equation y = 0.964x + 33.039 (assuming the value of y = 50) is obtained, and the x value is 17.59 ppm. The IC₅₀ value of the *Blumea balsamifera* ethyl acetate and ethanol extracts had extreme IC₅₀ antioxidant activity (<50 ppm).

Several studies about *Blumea balsamifera*, especially the leaves part informed fresh and dry leaves had antioxidant activity with boiling water method, antidiabetic activity with hydro-ethanol extract, antitumor activity with essential oil extract and also anticancer activity of ethyl acetate extract fraction (Widhiantara & Jawi, 2021).

The linear equation value for *Cordyline fruticosa* ethyl acetate extract y = 0.3628x + 23.253 (assuming the value of y = 50), then the x value is 73.72 ppm. The IC₅₀ value of *Cordyline fruticosa* ethyl acetate extract is strong (50-100 ppm). While the regression of *Cordyline fruticosa* ethanol extract equation y = 0.902x + 31.804 (with the value of y = 50) is obtained, the x value is 20.17 ppm. The IC₅₀ value of *Cordyline fruticosa* ethanol extract had extreme IC₅₀ antioxidant activity (<50 ppm).

The bioactive compounds of *Cordyline fruticosa* methanol leaf extract have been isolated. The active compound had antitumor activity, antibacterial activity, and cytotoxic activity (Assylfa et al., 2022).

CONCLUSIONS

An ethnobotanically-selected medicinal plant, *Blumea* balsamifera, and *Cordyline* fruticosa has been investigated for their antioxidant activities. The results informed that the leaves extract of ethyl acetate and ethanol of the plant showed good antioxidant properties by the extreme-good category of IC₅₀. Further investigation is needed to find the responsible compound in the *Blumea* balsamifera and *Cordyline* fruticosa plants and explored the possibility of bioproduction for the active compounds, also developed for healthcare in the future.

Acknowledgments: The research was funded by the Ministry of Education, Culture, Research, and Technology through Penelitian Dosen Pemula Scheme (Junior Lecturer Research Grants) 2022 under contract number 012/PL21.G/PG/2022. Acknowledgments were conveyed to the Muara Tae Village, West Kutai, East Kalimantan, Indonesia who have supported the research by sharing ethnopharmacological information and plant specimens.

Authors' Contributions: In this research, Nur Maulida Sari designed the research, supervised all processes, and wrote the manuscript. Wartomo was observing and taking research samples. Muhammad Fikri Hernandi controlled the samples and preparation of materials. Joko Prayitno supervised the research data analysis and the manuscript. Erna Rositah controlled research data analysis and manuscript writing. Farida Aryani

supervised all research data analysis and manuscript writing.

Competing Interests: There is no conflict of interest in this research.

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