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Potential Combination of Kapok Leaf Extract (*Ceiba pentandra* G.) and Turmeric Extract (*Curcuma domestica* Va) as an Anticancer Compound

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Abstract. Combination of kapok leaf extract (*Ceiba pertandra* G.) and turmeric extract (*Curcuma domestica* Va.) was carried out to determine the potential of extracts in treating cancer with BSLT and murine cells P388. Cancer is a disease that is very feared because it's difficult to cure, and even rarely causes death. The sample was extracted with methanol, the extract was mixed so that the mixture extract from the two samples was obtained. The results showed that in the BSLT test the mixed extract had a bioactivity against shrimp larvae with an LC₅₀ value of 142.946 ppm. While in Leukemia P388 cell testing showed that the combination of mixed extracts had a cytotoxic effect on Leukemia P388 cancer cells with inhibitory concentration values of 54.34 ppm. This shows that the kapok leaf extract (*Ceiba pentandra* G.) and combination of turmeric extract (*Curcuma domestica* Va.) has potential and can be developed as an anticancer agent because it has an IC₅₀ value that can inhibit murine P388 cell growth and LC₅₀ value which can kill shrimp larvae *Artemia salina* L.

Keywords: Kapok Leaves, turmeric, cancer, BSLT, murine P388 cell

Abstrak. Kombinasi ekstrak daun kapuk (*Ceiba pertandra* G.) dan ekstrak kunyit (*Curcuma domestica* Va.) telah diilakukan untuk mengetahui potensi ekstrak dalam mengobati kanker dengan BSLT dan sel murine P388. Kanker merupakan penyakit yang sangat ditakuti karena sulit disembuhkan, bahkan jarang sekali menyebabkan kematian. Sampel diekstraksi dengan metanol, ekstrak dicampur sehingga diperoleh ekstrak campuran dari kedua sampel tersebut. Hasil penelitian menunjukkan bahwa pada uji BSLT ekstrak campuran memiliki aktivitas bioaktivitas terhadap larva udang dengan nilai LC50 142,946 ppm, sedangkan pada pengujian sel leukemia P388 menunjukkan bahwa kombinasi ekstrak campuran memiliki efek sitotoksik terhadap sel kanker Leukemia P388 dengan nilai konsentrasi penghambatan 54,34 ppm. Hal ini menunjukkan bahwa ekstrak daun kapuk (*Ceiba pentandr*a G.) dan kombinasi ekstrak kunyit (*Curcuma domestica* Va.) berpotensi dan dapat dikembangkan sebagai agen antikanker karena memiliki nilai IC50 yang dapat menghambat pertumbuhan sel murine P388 dan nilai LC50 yang dapat membunuh larva udang *Artemia salina* L.

Kata Kunci: Daun kapuk, kunyit, kanker, BSLT, sel murine P388

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INTRODUCTION

Cancer is the second largest cause of death after cardiovascular disorders. Cancer or carcinoma is a disease caused by damage to the basic regulatory mechanism of cell behavior, specifically the mechanism of cell growth and change regulated by genes. New body tissue cells grow abnormally due to genetic mutations of cells, invade surrounding tissue, and metastasis (spread) to the body. The main cause of cancer is unknown, but it is suspected that the cause is genetic factors. The research of cancer concluded that 70 - 90% of cancers in humans are caused by

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environmental factors, such as food, alcohol consumption, air pollution, water, chemicals in the workplace, radiation and ultraviolet light (Miranti *et al.*, 2014).

The discovery of drugs derived from plants continues and provides important direction to various pharmacological targets including cancer, malaria, metabolic diseases, cardiovascular disease and neurologic disorders (Balunas & Kinghorn, 2005). Several studies have been carried out to find out various types of secondary metabolites found in several parts of kapok plants (Ceiba pentandra G.) including the leaves (Pratiwi, 2014). Methanol extract in leaves has high angiogenesis activity (Nam et al., 2003). Curcumin has potential in cancer treatment. Curcumin can inhibit breast development by inhibiting estrogen receptor activation (ER) by estrogen and is also able to inhibit the development of colon cancer cells (Liu & Ho, 2018). This study aims to determine the potential of anti-cancer compound activity in a combination of turmeric extract (Curcuma domestica Va.) and leaf extract of kapok plants (Ceiba pentandra Gaertn.).

MATERIAL AND METHODS

Materials

The materials used in this study were include turmeric and kapok leaves, ethanol, shrimp larvae, murine P388 cells, Fetal Bovine Serum (FBS), and Dimetil Sulfoksida (DMSO).

Procedure

Sample preparation

100 gram samples of turmeric and kapok leaves are taken around Kendari City, Southeast Sulawesi.

Preparation of kapok leaves and turmeric.

The kapok leaves that have been taken, washed to remove the dirt in the sample, then

dried and mashed using a blender to obtain fine powder. Meanwhile, turmeric samples are cut into small pieces, dried, then mashed using a blender to obtain fine powder. Each sample is stored in a beaker or container and ready to be extracted.

Extraction of kapok leaves and turmeric.

Extraction using ethanol 75% solvent, extraction carried out by maceration. Each vessel is added with 2500 mL ethanol, left for 2x24 hours. Then the residuals and extras are separated. Maceration was carried out 3 times with ethanol solvent.

Toxicity test with BSLT method

Toxicity tests were carried out on the eggs of A. salina which had hatched which was called nauplius. The shrimp larvae are ready to be tested by Brine Shrimp Lethality Test (BSLT), which is 48-hour-old larvae. The concentration of the test in this study for the mixture of kapok leaf extract and turmeric extract was 1000 ppm, 500 ppm, 100 ppm, 50 ppm, 25 ppm and 12.5 ppm. This study also made a concentration of 0 ppm as a control, without the addition of extract. Larvae are left to 24 hours, the number of dead larvae is calculated with the help of a magnifying glass (Setiawan et al., 2018). The parameters used were the number of dead larvae of 50% of the total test larvae. Then the LC50 value is calculated by entering the probit number (50% mortality of test larvae).

Activity test of combination of kapok leaf extract and turmeric in preventing cancer cells

Testing of cytotoxic activity against murine P388 cells with MTT medium (Ernawati et al., 2014). Murine P388 cells in RPMI 1640 medium (with a concentration of> 106 cells / mL) from culture flask were inserted in a 15 mL

centrifuge tube and centrifuged at a speed of 1200-1300 rpm for 5 minutes at room temperature. The supernatant was separated by a sterile Pasteur pipette and the precipitate was added with 1 mL FBS and 100 µL DMSO mixed slowly and transferred in a 2 mL tube. Cells were then dissolved and centrifuged for separation. Subsequently the test material subculture was carried out.

RESULT AND DISCUSSION

Result of Toxicity Test with BSLT Method

Based on the data in **Table 1** shows that the extract from a combination of kapok leaf extract and turmeric has bioactivity to shrimp larvae which is indicated by the LC₅₀ (Lethal

Concentration 50%) value of 142.946 ppm. This value indicates that at concentrations above the extract can kill shrimp larvae up to 50% of the population. The smaller the LC_{50} value of a sample, the higher the bioactivity (Pasilala *et al.*, 2016).

According to Meyer *et al.* (1982) an extract is said to be toxic if it has an LC_{50} value <1,000 ppm and a pure compound is said to be toxic if it has an LC_{50} value <30 ppm. Therefore, it can be said that the mixture of ethanol extract from kapok leaves and turmeric leaves is toxic because the LC_{50} value <1,000 ppm and has the potential as an anti-cancer agent.

Table 1. The results of the BSLT combination are kapok leaf extract and turmeric at same concentration

Concentration (ppm)	Concentration Log	Total Test Larvae	Number of Dead Larvae	% mortality	Probit Value	LC ₅₀
0	0	10	0	0	0	
12.5	1.09	10	0	0	0	
25	1.39	10	4	40	4.75	
50	1.69	10	6	60	5.25	142.946
100	2	10	7	70	5.39	
500	2.69	10	8	80	5.84	
1000	3	10	9	90	6.28	

LC₅₀ = Lethal Concentration 50%.

Activity Test Result of Combination of Kapok Leaf Extract and Turmeric in Preventing Cancer Cells

The results of data analysis showed that IC₅₀ (Inhibition Concentration) combination of ethanol extract of kapok leaves and turmeric leaves against Leukemia P388 cancer cells showed that the combination of ethanol extract of kapok leaves and turmeric had cytotoxic effects on P388 Leukemia cancer cells with

inhibitory concentration values of 54.34 ppm (**Table 2**). These results indicate that the IC₅₀ combination of ethanol extract of kapok leaves and turmeric leaves are quite active extracts. According to (Winarno *et al.*, 2010), the greater the IC₅₀ value, the more the compounds are not cytotoxic or the anti-cancer potential decreases. Based on this, the combination of ethanol extract from kapok leaves and turmeric has the potential to be anti-cancer.

Type of Extract	Concentration (ppm)		IC ₅₀		
		1	2	3	。
The combination of kapok leaf extract and turmeric	1000	0.012	0.003	0.02	54.34
	500	0.051	0.156	0.387	
	100	0.211	0.653	0.679	
	50	0.662	0.625	0.794	
	25	0.72	0.636	0.65	
	12.5	0.619	0.527	0.68	
	0	0.649	0.525	0.564	

Table 2. Cytotoxic test results and IC₅₀ values combination of ethanol extract of kapok leaves and turmeric against murine cells P388

IC₅₀= Inhibition Concentration 50

CONCLUSION

Inhibition of murine cell growth P388 50% (IC₅₀) from a combination of turmeric extract (*Curcuma domestica* Va.) And kapok leaf extract (*Ceiba pentandra* G.) was 54.34 ppm and also the results of toxicity tests on Artemia salina L. larvae with LC₅₀ values 142.946 ppm. Based on these results it can be seen that the combination of turmeric extract (*Curcuma domestica* Va.) And kapok leaf extract (*Ceiba pentandra* G.) has potential and can be developed as an anticancer agent because it has an IC₅₀ value that can inhibit murine P388 cell growth and LC₅₀ values that can kill *Artemia salina* L. shrimp larvae.

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