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ACTIVITY TEST OF ETHANOLIC EXTRACT FROM SEMBUNG'S ROOT (*BLUMEA BALSAMIFERA* [L.]/DC) ON INCREASING APPETITE OF WISTAR FEMALE MICE

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ABSTRACT

Objectives: Empirically, the root of Sembung can be used as an appetite enhancer/booster. Increasing appetite effect could be seen from increasing animal body weight (BW) and reducing leftover food every time observation. The aim of this study was to analysis of an ethanolic extract of Sembung's root on increasing appetite that exposure in Wistar female mice.

Methods: This research was laboratory experimental which used 25 female Swiss Wistar. Rats, then, were randomly divided into five groups (n=5), carboxymethyl cellulose 0.5%, normal control, ethanolic extract from sembung's root Dose I, II, III Dose of 600, 1200, 1800 mg/Kg Body Weight. Tests conducted for 14 days with two parameters: Weight gain and the remaining amount of feed. The second parameter is then calculated area under the curve (AUC) of total BW and food scraps. Data were analyzed using statistical test of one-way ANOVA.

Results: The results showed that ethanolic extract of root of *Blumea balsamifera* (L.) DC could increase that the AUC value of the average BW with average Dose I: 199.51±6.19, Dose II: 198.26±9.35, and Dose III: 196.81±7.58, negative control: 187.71±3.79 and normal control: 187.64±5.00. While the average AUC value of the remaining amount of food at first dose was 10.94±1.96 showed the lowest value. ANOVA test presented significant value (p<0.05) in reducing the amount of leftover food and increasing BW.

Conclusion: Exposuring of ethanolic extract of Sembung's root B. balsamifera (L.) DC indicated increasing appetite behavior of female Wistar rats.

Keywords: Blumea balsamifera (L.) DC, Appetite, Body weight.

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INTRODUCTION

Nutrient problem in both developing and developed countries consists of not only an essential nutrient deficiency but also appetite reduction [1]. Decreasingofappetiteisoneofthesymptomsfromseveraldiseases. If it occurs continually, it will interfere developing and growing of human body. Indirectly, through digestion of food, it supports energy for activity and brain [2].

To resolve this problem, there were several synthetic drugs but it necessary be considered that those had some side effects than traditional drugs [2]. One of the plants that can be treated to enhance appetite is Sembung (*Blumea balsamifera* [L.] DC) based on heritage prescription. Empirically, Sembung's root can be used to increase the appetite and contains essential oils that consist of cineole, borneol, limonene, dimethyl ether fluoroacetophenon, palmitic asam, myristin, sesquiterpene alcohol, dimethyl ether chloroacetophenone, pyrocatechin, glycoside, and linalool that have possibility to be used as appetite stimulator [3]. Aims of this study were to know the activity and effective dose of Sembung's root extract that can be used as an appetite enhancer with exposuring into Wistar mice.

METHODS

Sembung's root that was already cleaned and dried, ethanol 70%, Nacarboxymethyl cellulose (CMC) as a negative control, hydrochloride acid, FeCl₃, and magnesium metal. Animal test used were 25 female Swiss Wistar mice with an average weight between 150 and 200 g. Permission and approval for animal studies were obtained from Faculty of Medicine, Gadjah Mada University, Yogyakarta.

Identification of flavonoid

Powder of sample and extract was added with 5 mL ethanol, shaked then heated (10 minutes). Afterward, mixtures were filtered, added with Mg 0.2 g and 3 drops of HCl in every filtrate. Showing red color in ethanol layer indicated flavonoid compound [4].

Identification of tannin

The sample was added with 20 ml of water, boiled and filtered. Into the mixture, it was added several drops of FeCl_3 1%. Presenting greenish brown or livid color indicated tannin [5].

Identification of essential oils

About 70 g of simplicia from Sembung's root was destilated using water approximately for 4 hrs. Afterward, essential oil was separated. Water in essential oil was removed, and essential oil was tested using thin layer chromatography (TLC) [6].

Activity test of ethanolic extract of Sembung's root in female mice A number of 25 female mice were taken as animals test. Before administering, female mice were adapted in an environmental test during a week. Body weight (BW) of mice was observed and divided into five categories. There was Group I as negative control using Na-CMC, Group II as normal control (no administering), Groups III, IV, and C as treated animals. Treated animals were given doses of 600 mg/kg BW, 1200 mg/kg BW, and 1800 mg/kg BW of ethanolic extract of Sembung's root. Positive control and treated animal were given feed with amount 30 g/day for 14 days, then the BW of animals was observed.

Data analysis

Data were obtained from direct observation in female mice by register the BW and leftover food. After obtaining data (for 14 days), calculation of area under the curve (AUC) was conducted using equation (1). Data computation is done by calculating the value of AUC from the difference between the weight and the difference between the remaining amount of food for each individual treatment and data were analyzed using statistical test of one-way ANOVA.

$$\left[AUC\right]_{tn-1}^{tn} = \frac{Cn-1+Cn}{2}(tn+tn-1)$$
(1)

RESULTS

Identification of chemicals compound

Chemicals compound identification in Sembung's root in either powder or ethanolic extract was aimed to check the active compound contain in the sample that responsible as an appetite enhancer.

According to Fig. 1, identification of chemicals compound was specified for two chemicals contain, there were flavonoid and tannin. Flavonoid would show red color while tannin would be greenish brown or livid color. As it can be seen, the first Fig. 1a for flavonoid determination, it shows brownish red indicated no flavonoid there. In the other hand, Fig. 1b shows livid color and indicated the availability of tannin. Essential oil with TLC shows some spots.

The test of ethanolic extract of Sembung's root

Administering of ethanolic extract in female mice was observed with a deviation of BW and leftover food as main parameters. The results of administration can be seen in Table 1.

According to Tables 1 and 2, the difference in BW in each treatment group showed a weight gain on the Dose I of 600 mg/kg WB, Dose II 1200 mg/kg WB, and the Dose III 1800 mg/kg WB. The weight gain at a Dose I of 1600 mg/kg WB, then the Dose II of 1200 mg/kg WB, and the Dose III of 1800 mg/kg WB. AUC values for each treatment group showed a decrease at a Dose I of 600 mg I/kg WB, Dose II 1200 mg/kg WB, and the Dose III 1800 mg/kg WB.



Fig. 1: Identification of chemicals compound, (a) Flavonoid, (b) tannin, (c) Essential oil

DISCUSSION

The results of chemicals compound identification of both simplicia powder and extract showed the availability of tannin and essential oil. Essential oil contained in Sembung's root was used as a tonic which could be stimulated of appetite [3]. After treating during 14 days, the results (BW mice) were calculated the average of their weight, leftover food, and BW deviation. By observing the deviation of BW and decreasing of leftover food, it can be indicated improving appetite. It means, if Sembung's root has activity as appetite enhancer, it should be any increasing BW and decreasing leftover food.

This study showed the average value of AUC weight and the average value of AUC for residual feed the mice in each treatment group and the control group inversely. The fewer the remaining amount of feed, the greater BW of mice, after administration Sembung root extract essential oil [7]. There are several classes of essential oil based constituent components of essential oils of hydrocarbons, alcohols, phenols, phenol ethers, acid oxides palmitic, myristin, and ester [8].

According to Table 1, distinction AUC average showed between weight mice bodies in the negative control, normal control, and tested group. This probably Sembung's root had affect in increasing appetite of Wistar female mice. Average of enhancing BW mice shown that Dose I had the most increasing BW. It was caused by containing of palmitic acid, myristin, and linaloid in the essential oil that can induce appetite sense [9]. Dose I presented the biggest effect in increasing appetite. Probably, in the Dose I, the main compounds were extracted and more concentrated than other doses. Containing essential oil in Sembung's root had choleretic nature that accelerated bile secretion so that could speed up gastric draining, digesting and absorbing of fat in intestines. This mechanism could secret various hormones that regulated appetite enhancing [10].

Statistical results were an increase in BW and leftover food on Wistar female mice strain was significantly different between the control group and the test group which had significant value <0.05, which means the ethanol extract of the Sembung roots had the effect of increasing appetite.

CONCLUSION

In this research conducted was observed the appetite enhancing activity from Sembung's root by exposuring in Wistar female mice. According to the results taken, ethanolic extract of Sembung's root had activity as appetite enhancer with an effective dose of 600 mg/kg WB.

Table 1: Average of AUC of BW deviation in every group

Test group mg/kg BW	Average of increasing weight mice bodies (g)									
	Mice I	Mice II	Mice III	Mice IV	Mice V	Average AUC				
Dose I	190.36	200.18	205.00	197.00	205.00	199.51±6.19				
Dose II	186.07	196.25	193.71	208.21	207.04	198.26±9.35				
Dose III	204.32	205.43	189.64	190.36	194.29	196.81±7.58				
Negative control Normal control	190.36 192.50	183.21 187.14	183.93 186.79	190.54 191.79	190.54 180.00	187.71±3.79 187.64±5.00				

AUC: Area under the curve, BW: Body weight

Table 2: Average of AUC of leftover food in every group

Test group mg/kg BW	Average of increasing weight mice bodies (g)								
	Mice I	Mice II	Mice III	Mice IV	Mice V	Average AUC			
Dose I	13.10	8.34	10.78	9.86	12.61	10.94±1.96			
Dose II	16.53	11.45	11.70	10.91	10.89	12.29±2.39			
Dose III	10.39	14.06	11.62	10.19	12.08	11.67±1.56			
Negative control	21.39	21.70	23.08	22.94	15.60	20.94±3.08			
Normal control	19.51	19.20	20.81	18.57	18.46	19.31±0.96			

AUC: Area under the curve, BW: Body weight

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