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## ANTIDIARRHEAL ACTIVITY OF ETANOLIC EXTRACT OF BAY LEAVES (*SYZGIUM POLYANTHUM* [WIGHT.] WALP.)

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### ABSTRACT

Indonesia people have been used of Bay Leaves (*S. polyanthum* [Wight.] Walp.) to treat of antidiarrhea, cholesterol, hypertension, gastritis, and diabetes mellitus. Extract of Bay Leaves can be inhibit growth of *Escherichia coli*, *Vibrio Cholera*, and *Salmonella* sp. It's the bacterium that produce an enterotoxin so as to cause diarrhoea. Bioassay of antidiarrheal activity by using transit intestinal method and mice (*Mus musculus*) as an animal model induced by castor oil. The mice were grouped into five groups. Group I is received Na-CMC as control, group II, III, and IV are received of Ethanolic Extract of Bay Leaves (EEBL) with concentration 10%, 20%, and 30%, respectively and group V as positive control by received Loperamide-HCl. The charcoal used to marker as indicators. The result of the test indicate that EEBL have antidiarrheal activity to animal model. Refers to analysis of variant  $p=0.05$ , showing that concentration 30% has an activity non significant with loperamide-HCl as positive control.

**Keyword:** Antidiarrheal, Bay leaves, *S. polyanthum* [Wight.] Walp.

### INTRODUCTION

Diarrhea is one of the main problems in the case of child mortality to 19% and responsible for the deaths of children under five in developing countries<sup>1</sup> such as Indonesia. Refers to WHO estimation for the year 1998, there were about 7.1 million deaths due to diarrhea. Diarrhea is a symptom of clinical and gastrointestinal disorders characterized by increased frequency of defecation, usually accompanied by changes in the shape and consistency<sup>2,3</sup>.

Diarrhea is the most given dangerous symptom specially to children. Research to find active extracts antidiarrheal essential to achieving the treatment of diarrhea<sup>4</sup>. Treating diarrhea using medicinal plant extracts have been widely used empirically<sup>5</sup>. Bay Leaves (*S. polyanthum* [wight.] Walp.) is one of the medicinal plant and believed efficacious for treating of diarrhea<sup>5,6</sup>. Local name is Daun Salam.

Based on the description, to prove the antidiarrheal activity of ethanolic extract of bay leaves (EEBL) should be research in vivo preclinical studies. The results of this preclinical study is expected to be a reference to scientific evidence and material development EEBL as antidiarrheal medication.

### MATERIALS AND METHOD

**Material:** Mice as animal model (150-200 g) were purchase from Pharmacology Laboratory, Faculty of Pharmacy UMI, Bay Leaves (*S. polyanthum* [wight.] Walp.), aquabidistilled (Ikhapharmindo Putramas, Jakarta), ethanol 70%, Loperamide-HCl, Na-CMC, Arabic gum suspension 20% and tinged with 5% charcohal as a marker. vortex (CAT.M. Zippear GmbH. Etzenbach, W. Germany), castor oil.

#### Preparation and determine sample test

Sample of Bay leaves was collected from Manyampa Village, District of Ujung Loe, Bulukumba Regency, Indonesia. It was determined by Mrs Aksam Roskiana Ahmad in Botany Division, Pharmacognosy and Phitochemistry Laboratory Faculty of Pharmacy Universitas Muslim Indonesia Makassar Indonesia. The Bay Leaves is dried, grinded, and Then powdered to making light of extraction process<sup>7</sup>.

#### Extraction

Extraction using modified method from Fang<sup>8</sup>. Bay leaves of 500 g extracted by maceration method using ethanol solution 70% and allowed to stand for 5 days, then remaceration to obtain more extract. The Liquid extract are collected and evaporated with rotary vacuum evaporator (rotavapor IKA® HB 10 Basic) to getting thick extract. Extract concentration for assay is 10%, 20% and 30% in Na-CMC suspension<sup>9</sup>.

#### Preparation and Grouping Animal Test<sup>9,10</sup>

Animals model were purchase from pharmacology laboratory Faculty of Pharmacy UMI. It fasted about 18 hours, but drinking still receive. Animals were randomly divided into five groups and each group of three animal models. Group I is a control, group II, III, IV as a test group and group V as a positive controls.

#### Testing of EEBL to Animals Models<sup>9,10</sup>

Animals models were administering on orally. At time  $t=0$ , group I was given Na-CMC, Groups II, III and IV were each received EEBL a concentration of 10%, 20% and 30%, group V was given Loperamide-HCl suspension, respectively by volume 1.0 ml/100 gbw, 1 h before administration of castor oil. At time  $t = 45$  min after administration of castor oil, all groups were given marker charcoal suspension 0.1 mL/100 g bw. At time  $t = 65$  min, all groups were sacrificed by dislocation of cervical. Intestine removed then measured the total length of the intestine and creep distance marker charcoal. Then, calculated the ratio of the distance normal to the length of the colon charcoal marker entirely from pylorus to caecum<sup>10</sup>.

#### Statistical Analysis

Statistical analysis was used one way anova (ANOVA). The results of Analysis of Variant obtained from experiment groups ( $p=0.05$ ) and multiple comparisons of groups which cause different were calculated according to least significant different (LSD) ( $p=0,05$ ) as post hock test<sup>9</sup>.

Table 1. Ratio of marker charcoal

No	Treatment	Ratio	Percent Inhibition %
1	Na-CMC	0.671	0.00
2	EEBL 10%	0.252	62.44
3	EEBL 20%	0.142	78.84
4	EEBL 30%	0.080	88.08
5	Loperamid-HCl	0.069	89.72

EEBL = Ethanolic extract of Bay Leaves

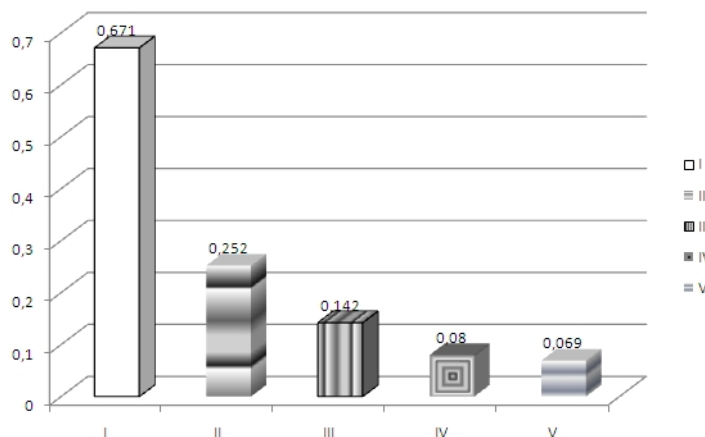


Figure 1. Ratio of flow charcoal marker in each groups Na-CMC, EEBL dan loperamid-HCl

## RESULTS

In each group of mice, ratio of charcoal is measured with metric tools. Charcoal as marker indicated with black colours and then flow of line up charcoal equal entire size of colon from phundus to phylorus (Table 1). Value of Ration are shown that all group extract different compared to control Na-CMC (Fig.1).

## DISCUSSION

Bay leaves (*S. polyanthum* [Wight.] Walp.) is a herbal widespred in Southeast Asia such as Indonesia country. It is used to flavoring agent and a native herbal medicine in Indonesia people to treat cholesterol, diarrhea, reduce of serum glucosa, LDL, and increase of HDL<sup>11,6</sup> antibacteria, antifungi, and inducing apoptosis<sup>9</sup>, rheumatic, gastritis and hypertension, antioxidant activity<sup>12,13</sup>.

In case diarrhea is same characterized by secretory cause of hypermotility. The use of castor oil induced diarrhea is logical because it is cause to increase of intestinals peristaltic. Castor oil is hydrolized by lipase enzyme to glycerol and ricinoleat acid. Ricinoleat acid is cause of diarrhea.

Antidiarrhea is measured with transit intestinal method. Transit intestinal method used marker charcoal as indicator<sup>14</sup> whereas flow of marker into overall intestinal from phundus to phylorus as determined. The delay of intestinal transit has been used to investigated bioactivity and effect as antidiarrhea no particular etiologic agent<sup>15</sup>.

The result showing that control of Na-CMC have hight ratio 0.671. EEBL 10%; 20%; 30% is 0.252, 0.142, 0.080, respectively and loperamide-HCl 0.069 (Table 1). Percent inhibition diarrhea of animal model induced carcoal oil measured to control of NaCMC. Percent inhibition of EEBL 10%, 20%, 30% and loperamide-HCl is 63%, 78%, 88%, and 89% respectively. Statistic analisys of ratio to all animals with ANOVA showing that significant different confidens level 95%. Analisis of multiple comparison according post hoc tests with LSD showing that all extract is significant to

compare with Na-CMC. At 10% and 20% is non significant, 10%, 30% and Loperamide-HCl is significant, and between 20%, 30% and Loperamide-HCl is non significant.

BL extract contains of phenolic, polyphenol e.g. tannin and flavonoid<sup>12,14,2</sup>, 10-epigazanioplide, gazaniolid, spirafolide, costunolid, reinosin, santamarin<sup>8</sup>, volatile compounds e.g. citral<sup>16,17</sup>. Bay leaves have active to *Streptococcus* sp<sup>6</sup>, *S. enterica* dan *E.coli*<sup>1,18</sup>. The extract activity of bay leaves may be related to the phenolic compounds present in the extracts. The phenolic such as tannin is potent as antidiarrhea afficity.

The conclusion that EEBL have effect to diarrhea. This study can be supporting to develop of phytopharmaca. According to Ministry of health in Indonesia country, Bay leaves is one of the several herb to must be deelop as phytopharmaca.

## CONCLUSION

Bay leaves extract in 30% have best antidiarrhea activity to animal models.

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