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IDENTIFICATION OF SECONDARY METABOLITES AND ANTIDIARRHEAL ACTIVITY OF PIRDOT LEAVES ETHANOL EXTRACT (*Saurauia vulcani* Korth.) FROM WEST PAKPAK, NORTH SUMATERA PROVINCE, INDONESIA

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ABSTRACT

Pirdot (*Saurauia vulcani* Korth) is used by the people of West Pakpak in North Sumatera as medicinal ingredients. This study aims to identify secondary metabolite and to know the antidiarrheal activity of ethanol extract of pirdot leaves which tested on mice male which is induced by oleum ricini. The dosage variation of ethanol extract is 50 mg/kg body weight, 100 mg/kg body weight, and 200 mg/kg body weight. Loperamide was used as positive control with 2 mg/kg body weight dose, and 0.5% CMC Na as negative control. Determination of antidiarrheal activity from the frequency of diarrhea, duration of diarrhea, and the consistency of the mice's feces which induced with oleum ricini by comparing the % inhibition from parameters. The results obtained showed that the extract dose 100 mg/kg body weight better than other variation of dose but not better than loperamide 2 mg/kg body weight as antidiarrheal activity. Based on this research that the extract of pirdot leaves can be use as antidiarrheal with the right dose.

Keywords: Pirdot leaves, Oleum ricini, Loperamide, and Antidiarrheal.

INTRODUCTION

Diarrhea is closely related to impaired of physical development, cognitive, susceptible to disease and cause of death especially in children under the age of 5 years old. This event generally occurs in developing countries such as Africa and Southeast Asia. Diarrhea is also referred to as gastroenteritis.³ Diarrhea is characterized by an increase the frequency of bowel movements, soft-liquid stool conditions, and abdominal pain discomfort.^{1,2} Treatments is generally done by giving synthetic drugs such as loperamide, diphenoxylate, and antibiotics that give some side effects and drug resistance.^{2,4} The overcoming the side effects caused of, it's important to find a potential new drugs that can be obtained from natural ingredients. Traditionally treatment using natural ingredients from plants is considered safer from the side effects caused.⁵ Therefore, research into the discovery of active compounds from potential plants is considered to be important to study.

The pirdot plant (*Saurauia vulcani* Korth.) Is a type of plant, that is rich in potential bioactive compounds. Components of bioactive compounds found in pirdot leaves (*Saurauia vulcani* Korth.) Include alkaloids, flavonoids, saponins, triterpenoids and tannins.⁶ Empirical information of pirdot leaves has been traditionally used as a diabetes drug from boiled water.⁷ In vivo through pharmacological tests of extract extracts pirdot leaf ethanol has been proven to be an

antidiabetic mellitus-induced antidiabetic mellitus in mice,⁸ and wound healing activity in hyperglycemic mice induced by Streptozotocin (STZ)⁹ and as an antioxidant.¹⁰ Other potentials of the bioactive compounds of pirdot leaves (*Saurauia vulcani* Korth.) as antidiarrheal have not been report. Based on the description above, researchers are interested in testing the activity of pirdot leaves ethanol extract as an antidiarrheal which is tested on adult white male mice induced by oleum ricini.

EXPERIMENTAL

Preparation of the plant

Fresh sample of Pirdot leaves taken and collected from the District of Salak (West Pakpak, North Sumatera Province, Indonesia) and authenticated by Herbarium Medanense, Faculty of Mathematics and Natural Science - University of Sumatera Utara (No. 2380/MEDA/2018). Clean the sample, dried in drying cabinet at 40 °C, powdered and stored at the Pharmacognosy Laboratory, Sekolah Tinggi Ilmu Kesehatan Medan.

Preparation and Process of the Extract Plant

500 g pirdot leaves powder extracted with ethanol 96% with maceration method 5 days arrow at temperature room and occasionally stirred. After 5 days, filtered with Whatman paper No 1. Macerated the residu with the same way twice. Evaporated the filtrate with evaporator rotary vacuum at 55 °C to get crude extract.¹¹ Mixed the sticky extract then dried it in the oven at 40 °C. put the dried powder in a bottle and stored in desiccator at laboratory at the temperature room.


Phytochemical screening

The extract of pirdot leaves that soluble in ethanol analyzed of phytochemical compounds with standard methods.^{12,13,14}

Preparation the mice

The animal that used for the research are adult male mice and health (± 25 g) from House of Animal STIKes Senior Medan, Indonesia. Mice are place in standart cage at temperature rom and fed it with standart pellets and freely water. This research get permission and done this according to ethical norms from National Health Research Ethics Committees (No. 0403/KEPH-FMIPA/2019). The mice are fasted for 18 hour not fed but given free access to water.

Antidiarrheal activity testing of Pirdot Leaves Ethanol Extrac (PLEE)

The mice grouped into 5  at consist to:

Group 1: PLEE dose 50 mg/kg body weight

Group 2: PLEE dose 100 mg/kg body weight

Group 3: PLEE dose 200 mg/kg body weight

Group 4: Negative Control 0,5% CMC Na

Group 5: Positive Control Loperamide 2 mg/kg body weight

The mice induced with 0,5 mL oleum ricini to get diarrhea by oral.¹⁵ After 2 hour of induction, then given the preparation test as much as the fit dosage that determined before, then do the observation per 30 minutes. Observation that have done included diarrhea frequently, duration of diarrhea, and consistency of the feces. The percentage of diarrhea determined by:¹⁶

$$\% \text{inhibisi diarrhea} = \left(\frac{X_{\text{Control}} - X_{\text{Sample}}}{X_{\text{Control}}} \right) \times 100$$

Statistic analysis

Data that obtained analyzed with ANOVA and followed by Post hoc Tuckey using SPSS 16 version. All the data is stated as mean \pm SEM and n = 5 with level of trust 95% ($\alpha < 0,05$).

RESULTS AND DISCUSSION

Crude extract ethanol of pirdot leaves that obtained means 136.24 ± 0.05 g. Result of phytochemical screening against ethanol extract of pirdot leaves shows the difference phytochemical compounds (Table-1).

Table-1: Phytochemical screening of PLEE

Secondary Metabolites	Reagent	Ethanol Extract
Alkaloids	Mayer	Positive (+)
	Dragendroff	Positive (+)
	Wagner	Positive (+)
Flavonoids	Shinoda test	Positive (+)
Saponins	Foaming Test	Positive (+)
Tannins	FeCl ₃ 1%	Positive (+)
Triterpenoids	Liebermann Bouchard	Negative (-)
Glycosides	Liebermann's Test	Negative (-)
Anthraquinones	NH ₄ OH test	Positive (+)

Antidiarrheal Activity Test

Antidiarrhea parameters testing from ethanol extract of pirdot leaves which diarrhea frequency, duration of diarrhea, and consistency of diarrhea. Antidiarrheal activity was measured by in vivo method by using mice male (*Mus musculus*) as test animal. *Mus musculus* induced with 0,5 mL oleum ricini by oral so the mice get diarrhea (gastrointestinal).^{16,17}

Tabel-2: PLEE formulation againts antidiarrheal Activity Parametere that tested on mice after 2 hours Induction of Oleum Ricini

Treatment	Antidiarrheal Activity			
	Frequency of diarrhea	% inhibition	duration of diarrhea	% inhibition
0.5% CMC Na	14.0 ± 0.27	0.00	$272.4 \pm 0.11^{**}$	0.00
PLEE 50 mg/kg bw	$11.0 \pm 0.38^*$	21.43	$230.2 \pm 0.15^{**}$	15.5
PLEE 100 mg/kg bw	$9.2 \pm 0.46^*$	34.29	$149.1 \pm 0.20^{**}$	45.3
PLEE 200 mg/kg bw	$10.4 \pm 0.38^*$	25.71	$203.2 \pm 0.15^{**}$	25.40
Loperamide 2 mg/kg bw	$8.4 \pm 0.37^{**}$	40.00	$120.2 \pm 0.22^{**}$	55.88

Description: the result is stated as average \pm SEM, n = 5; * $P < 0,05$, ** $P < 0,01$

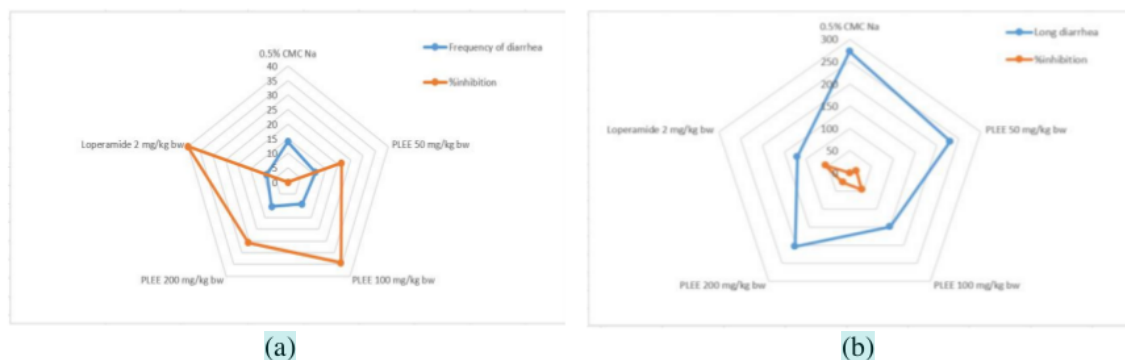


Fig-1: Antidiarrheal activity of PLEE; (a) diarrhea frequency, and (b) duration of diarrhea

Based on Table-2 and Fig.-1a, ethanol extract of pirdot leaves shows activity against diarrhea frequency. The variation dose of PLEE showed that activity could significantly inhibit the frequency of diarrhea 21.43-34.29% ($P < 0.05$) and loperamide drugs were able to inhibit diarrhea by 40% with significance ($P < 0.01$). Same as with the old parameters of diarrhea shows activity as antidiarrheal. The variation dose of PLEE was able to inhibit 15.5-45.3% with significance ($P < 0.05$) and loperamide drugs 55.88% with significance ($P < 0.01$) (Fig.-1b). Observation the consistency of diarrhea feces for variations at the dose of PLEE was observed in the form of mice's feces. Observation of feces form was carried out after 2 hours to induction of oleum ricini per 30 minutes observation. Form of the feces is divided into three categories including liquid feces 3, soft feces 2, and normal feces 1. Diarrhea stool is categorized in the soft and liquid feces form. The observations are shown in Fig-2.

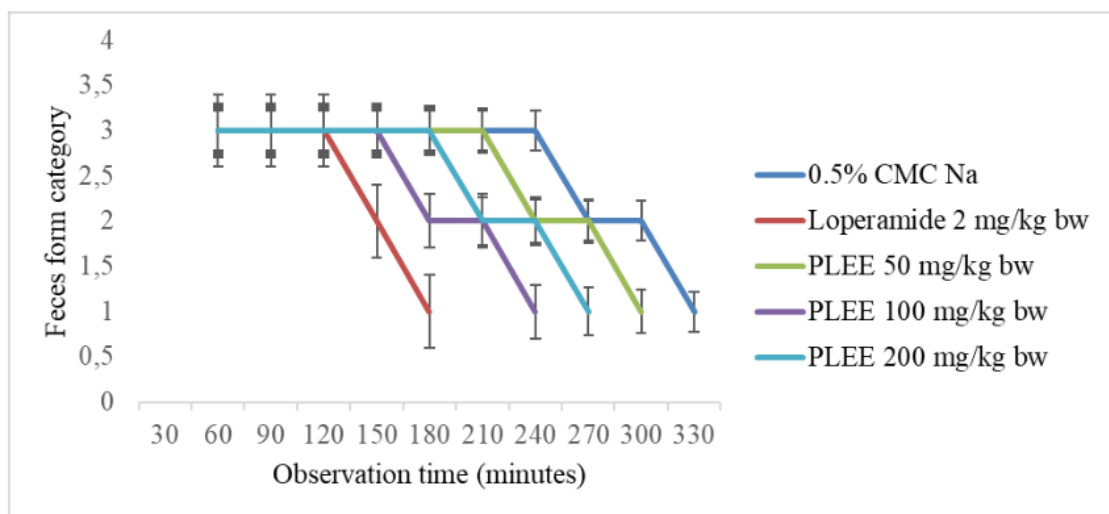


Fig-2. Antidiarrheal activity of PLEE of consistency of diarrhea feces

The best dose PLEE as antidiarrheal at 100 mg/kg bw. The effect increasing of extract concentration is not directly proportional to increase in antidiarrheal activity, it can be seen between the concentration of 100 mg/kg bw with 200 mg/kg bw the concentration of 200 mg/kg bw decreases antidiarrheal activity. This is because of possibility at a concentration of 100 mg/kg bw has occupied all parts of the receptor. Besides that it is possible that the active compound contained in the extract has an "agonist-antagonist" effect. For example, tannin active is compounds with a anthraquinone have opposite work effects, tanins effectively inhibit secretion of chloride induced by forskoli and toxins while anthraquinone acts as a laxansia.¹⁸

CONCLUSION

Ethanol extract of pirdot leaves (*Saurauia vulcani* Korth) shows activity as antidiarrheal that tested to male mice. Increase concentration ethanol extract of pirdot leaves not directly proportional with antidiarrheal activity. The best activity ethanol extract of pirdot leaves at 100 mg/kg body weight.

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