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194 <https://oamjms.eu/index.php/mjms/index> Scientific Foundation SPIROSKI, Skopje, Republic of Macedonia Open Access Macedonian Journal of Medical Sciences. 2022 Jan 03; 10(T8):194-199. <https://doi.org/10.3889/oamjms.2022.9458> eISSN: 1857-9655 Category: T8 -"APHNI: Health Improvement Strategies Post Pandemic Covid-19" Section: Pharmacology Screening and Antidiarrheal Activity Testing of Sembung Rambat (Mikania micrantha) Leaves Arif Ardianto 1 , Dena Munarsih 1 , Iin Nur Rahayu 1 , Muhammad Muzhil Aslam 1 , M.

Fahrul Aditya 2 , Daru Estiningsih 1* , Annisa Fatmawati 1 , Pujo Hari Saputro 2 1Department of Pharmacy, Faculty of Health Sciences, Alma Ata University, Kasihan, Indonesia; 2Department of Informatics, Faculty of Computer, Alma Ata University, Kasihan, Indonesia Abstract BACKGROUND: prevalence diarrhea Indonesia exceedingly In it an increment morbidity mortality in Furthermore, year than billion suffer from diarrhea, and 3.2 million experience death from this disease. Based on empirical evidence in Pelita Jaya, Pesisir Barat of Lampung, Sembung Rambat (Mikania micrantha) leaves are commonly used to treat diarrhea, rheumatism, and bleeding wounds.

In addition, alkaloids are one of the compounds contained in sembung rambat leaves that can reduce or inhibit the disposal of waste substances from the body or have antidiarrheal properties. AIM: The research aims to prove the potential effect of sembung rambat (M. micrantha) leaf extract as an antidiarrheal agent by screening and conducting a pharmacological test on male Swiss mice. METHODS: The phytochemical qualitative analysis was employed to screen the M. micrantha leaf extract. Moreover, the antidiarrheal effect was tested using the intestinal transit method of diarrhea in mice induced by Oleum Ricini.

RESULTS: phytochemical results that are secondary of tannins, and contained the The of marker on intestine length after = min that ethanol of rambat M. micrantha) contains antidiarrheal activity with a dose variation of 100 mg/kg BW, 150 mg/kg BW, and 200 mg/kg BW. CONCLUSIONS: extract sembung (M. micrantha) at doses 100 mg/kg 150 mg/kg and mg/kg possesses activities. The effective is mg/kg as it performed the closest ratio to the positive control group. Edited by: Sinisa Stojanoski Citation: Ardianto A, Munarsih D, Rahayu IN, Aslam MM, Aditya MF, Estiningsih D, Fatmawati A, Saputro PH.

Screening and Antidiarrheal Activity Testing of Sembung Rambat (Mikania micrantha) Leaves. Open Access Maced J Med Sci. 2022 Jan 03; 10(T8):194-199. <https://doi.org/10.3889/oamjms.2022.9458> Keywords: Antidiarrheal; Sembung rambat (Mikania micrantha); Intestinal transit *Correspondence: Daru Estiningsih, Department of Pharmacy, Faculty of Health Sciences, Alma Ata University, Kasihan, Indonesia. E-mail: daru_estiningsih@almaata.ac.id Received : 13-Oct-2021 Revised: 21-Nov-2021 Accepted: 02-Dec-2021 Copyright: © 2022 Arif Ardianto, Dena Munarsih, Iin Nur Rahayu, Muhammad Muzhil Aslam, M.

Fahrul Aditya, Daru Estiningsih, Annisa Fatmawati, Pujo Hari Saputro Funding: This study was supported by the Ministry of Education, Culture, Research, and Technology of the Directorate General of Higher Education for the 2021 Student Creativity Program grant Competing Interests: The authors have declared that no competing interests exist Open Access: This is an open-access article distributed under the terms of the Creative Commons Attribution- NonCommercial 4.0 International License (CC BY-NC 4.0) Introduction Diarrhea one the causes infant mortality in the world with a fairly high child mortality rate. The thing also in pathophysiology with etiologic [1]. is one the public problems Indonesia. can seen the morbidity mortality particularly toddlers.

was that than billion suffered diarrhea, the toll of in reaches million year. could diarrhea about times year and more than 80% of them died at the age of <2 [2]. According the in diarrhea the second cause life reduction 1.97 years, just below the lower respiratory tract infections (2.09 years). addition, 2016 drinking water, sanitation, an environment were main responsible the of million people including more than 470,000 infant deaths caused diarrhea, the and world organizations to deal with it [3]. Moreover, major of in children five malnutrition.

billion experience every Malnourished and living HIV at for and even In past, people severely dehydrated could in However, another of nowadays a bacterial which lead death. is symptom infection the tract by bacterial, and infections. infection can be spread on contaminated food and drink or it can be passed from person to person [4]. According the Health in the of in was at (9%) the of year, 73,188 (11.5%) at the age of

1–4 years, 182,338 (6.2%) at the age of 5–14 years, and 165,644 (6.7%) in children aged 15–24 years [5]. Based the survey by Diarrhea of Ministry Health 2000 2010, was tendency diarrhea to In the rate diarrheal was population, Open Access Maced J Med Sci. 2022 Jan 03; 10(T8):194-199.

195 2003 rose 374/1,000 in it to 423/1,000 and 2010 reached population. events diarrhea also still found, with high Fatality Rate In there outbreaks 69 districts a of cases 239 (CFR 2.94%). In 2009, there were extraordinary events in sub-districts a of cases 100 (CFR while 2010, was diarrhea occurred 33 with a of patients 73 (CFR and is second cause death children 5 years. there be innovative action to develop antidiarrheal drugs [6]. Based the efforts be to address morbidity mortality by in One the that be is development the of medicines are generally to minimal effects. it is that development these can later consumed by community from to Observation in Jaya Kec.

Selatan Pesisir Lampung, where the vines (*Mikania micrantha*) are used by the community as traditional antidiarrheal medicine. The found facts in Pelita village, Selatan Pesisir Barat, *M. micrantha* used the as antidiarrheal Sembung leaf *M. micrantha*) a of possessing great as alternative that be However, plant rarely because grows the of rubber oil plantations are by as a with efficacy. that it based the results of phytochemical analysis, sembung rambat leaf contains substances the of secondary such alkaloids, flavonoids, steroids, tannins, and terpenoids [7].

Furthermore, on empirical evidence, study conducted prove hypothesis sembung (*M. micrantha*) extract be as by out phytochemical to secondary with effects pharmacological on male mice. animal was out to the activity *M. micrantha*. A quantitative analysis was also conducted to figure out the content of alkaloid compounds found in *M. micrantha* which is regarded to be efficacious as antidiarrheals as well to the dose can be used directly by the public. The results that rambat contain triterpenoids, steroids [7], of is alkaloid with antidiarrheal, anti-microbial, anti-malarial properties.

are most secondary metabolites nitrogen which often found plant animal Almost alkaloid compounds from especially More than 20% of angiosperm species contain alkaloids [8]. Methods Tools and materials Tools Maceration glassware, masks, medical rotary mouse weighing for animals, feeding tube, 50 ml beakers, mice instruments, pipettes, flask, volumetric flask were used. The tools used were obtained from pharmacology and laboratory of Alma Ata University, Yogyakarta. Ingredients Ethanol Aquadest, 10% FeCl₃, reagent, norit marker, (loperamide), preparation mikania leaf extract), CMC 0.5%, Oleum The used were obtained from CV. Chem-Mix Primary. Research location This study was conducted at the Pharmacology Laboratory the

Laboratory the Pharmacy Study Alma University, Yogyakarta.

Sample This used Swiss weighing approximately 20–35 g with an average age of 8 weeks. The mice were divided into five groups and each group consisted four obtained the animal development unit, UGM. Determining the plant The of rambat were from area, Java. Determination plants to the of the species avoid in usage the material that will be studied. Making ethanol extract of sembung rambat (*M. micrantha*) leaves Simplicia extracted employing maceration [9] 96% The 196 <https://oamjms.eu/index.php/mjms/index> was obtained from the evaporated solvent using a rotary evaporator, was on water The extraction was carried out by 2× remuneration. Phytochemical test The screening carried out determine groups compounds contained the extract sembung leaves.

phytochemical was out conventional of tannins, flavonoids, and saponins. Antidiarrheal test using the intestinal transit method The activity the extract of rambat was using intestinal transit method in mice-induced *Oleum Ricini*. Before being used test male mice adapted the environment 1 week then for 18–24 h the The of the is no intake affect antidiarrheal process. the were to the administration, they divided five groups four each. division the was: Group I control (2) II control (-), Group III a I a of mg/kg (4) Group IV test at dose 150 mg/kg and (5) Group V as the third test with a dose of 200 mg/kg BW. All groups were induced with *Oleum Ricini* suspension at a dose of 0.2

ml/20 g mice. This treatment has met ethical clearance the of approval the number KE/AA/VII/10578/EC/2021. At = min being with *Ricini*, group given treatment, (1) Group I given suspension a dose 16 mg/70 BW control (2) II was CMC suspension as (-), (3) III given suspension curd at dose 100 mg/kg (4) IV given a of propagation at dose 150 mg/kg and Group V given suspension extract curdling at a dose of 200 mg/kg BW. At = min the treatment, groups given norit per-oral as a marker, continued at t = 30 min after administration of norit. The mice's neck dislocation was until mice Afterward, mice's was dissected.

mice's intestines taken, the of intestine through the marker and total length the were The was to the of track by the to total length. The obtained were statistically analysis variance (ANOVA) The obtained statistically analyzed determine there significant differences between each group [11]. Results Plant determination results The was out the Biology Laboratory of the Faculty of Applied Science and Technology, Ahmad Dahlan University, Yogyakarta. The results that plant was *M. micrantha* Kunth, Flora of Java [12].

Determination code • 1b–2b–3b–4b–12b–13b–14b–17b–18b–19b– Asteraceae • Mikania

• 1b–2a–3a *M. micrantha* Kunth. Synonyms *Conyza balsamifera* (L.) DC. Extraction results Sembung leaf using 96% solvent a of The of yield obtained the calculation the of thick (g) by weight the powder multiplied 100%. extraction a of The percentage of the yield was obtained by calculating the thick weight dividing the of simplicia (g), multiplying 100%. extraction results are shown in Table 1.

Table 1: Results of ethanol extract of sembung rambat leaves Materials and Yield Amount Weight of Simplicia powder 478 g The volume of 96% ethanol 6 L Thick extract weight 55 g Yield 11.50% Phytochemical screening results of Simplicia and extracts The results of the phytochemical screening test are in Figure 1. The in Table 2 the phytochemical using methods or reagents determine content secondary metabolites of alkaloids, tannins, flavonoids, and saponins. The results of phytochemical screening showed that rambat (*M.*

micrantha) were positive for alkaloids, flavonoids, tannins, and saponins. Open Access Maced J Med Sci. 2022 Jan 03; 10(T8):194-199. 197 Antidiarrheal activity test The testing carried using intestinal method, divided test into groups. this the control (+) the of suspension the of mg/70 BW an ratio 0.170 0.064, than negative group with CMC-Na The average value of the ratio is 0.322 ± 0.971 . Tests of antidiarrheal activity carried out in five treatment groups obtained results, as presented in Table 3 .

Table 2: Results of phytochemical screening of Simplicia and extracts Phytochemical Test Reagent Discoloration Results Alkaloids HCl 2 N+reagent Wagner A brown precipitate is formed + Tannins FeCl₃ Brownish green to blue Black + Flavonoids NaOH 10% Brownish green to green Yellowish + Saponins Aquadest+Shake Strong Foam formed + (+): Detected, (-): Not detected. The between length the passed the to overall of intestine each tested shown Figure 2 , in test I, Group II, test III have smaller than negative group. Test Group III has an average ratio close to the positive control group.

Based the graph in 2, the control (+) 0.17, negative control has 0.322 test has ratio 0.313, test has ratio 0.261, test has ratio 0.213 The (+) group the ratio, was proportional the negative (-) control group. Discussion The of has determined to that Simplicia in study sembung leaf *M. micrantha*) from Pangandaran Jawa The of sembung leaf 96% solvent because on polarity 96% can attract isolate metabolites alkaloids, tannins, and most which antidiarrheal thick resulted the extraction of active compounds from plant Simplicia or Simplicia a table Then, the was and thick was obtained [13]. Before the extract sembung rambat phytochemical was out determine secondary content in rambat.

examination alkaloids carried by 1 ml sembung leaf extract adding N and 's There a

reaction alkaloids a precipitate formed. examination was out taking ml sembung leaf extract and adding FeCl₃ reagent. A positive reaction for tannins occurred because a black or dark blue color was formed Examination flavonoids performed by 1 ml sembung leaf and adding a few drops of 10% NaOH. The reaction showed positive flavonoids with a specific color change. Saponin examination was carried out by taking 1 ml of sembung rambat extract, distilled and it. The positive reaction of saponins was indicated in the formation of long-lasting foam [10].

The of dose for test based the studies Figure 1: Phytochemical screening results Figure 2: The average ratio of marker length to overall intestinal length Table 3:

Pharmacological test results data Group Code Length of markers (cm) Length of intestine (cm) Ratio p-value Control(+) K(+) 1 11.5 58 0.198 K(+) 2 9 63.5 0.141 0.655* K(+) 3 9.5 61 0.155 K(+) 4 10 54 0.185 Control (-) K(-) 1 18 60 0.300 K(-) 2 19.5 54 0.361 0.325* K(-) 3 18.5 62 0.298 K(-) 4 18 55 0.327 Test I U (1) 1 18 57.5 0.313 U (1) 2 19.5 59 0.330 0.406* U (1) 3 18 59 0.305 U (1) 4 18 59.5 0.302 Test II U (2) 1 15 56 0.267 U (2) 2 14 56 0.250 0.625* U (2) 3 15.5 55 0.281 U (2) 4 14 57 0.245 Test III U (3) 1 12 53 0.226 U (3) 2 13 62 0.209 0.627* U (3) 3 12 61 0.196 U (3) 4 12 54 0.222 *Data are normally distributed (p > 0.05). 198 <https://oamjms.eu/index.php/mjms/index> five of consisting 20 mg/kg 40 mg/kg 80 mg/kg 100 mg/kg and 150 mg/kg From dose the of mg/kg and mg/kg began have an effect.

this used three of consisting 100 mg/kg 150 mg/kg BW, and 200 mg/kg BW. In addition, loperamide was given to the positive control as comparison determine the rambat extract an activity not. negative group given CMC-Na 0.5% to determine whether the solvent had an antidiarrheal This was to that the antidiarrheal was – it from effect sembung leaf and not the effect of the solvent. Furthermore, mechanism loperamide as antidiarrheal slow motility the gastrointestinal tract through the circular or longitudinal muscles the and results interactions with receptors the [15]. can intestinal time flow from intestine to the colon.

It can also normalize the balance of or of in intestinal mucous membrane [16]. The transit to the antidiarrheal was out measuring ratio of the length of the intestine that the marker passed at specific to overall of intestine. A small ratio indicates antidiarrheal activity. According to the research loperamide as control an effect, the average value of the ratio of the positive control group was smaller, which is 0.667 compared to the negative control group 0.708. addition, ratio the Leaf Ethanol (Melia azedarach Linn) group smaller the control The of reducing peristalsis and will food subsequently, smaller ratio measurement is obtained.

The of study analyzed statistically to determine whether there were differences in group.

Statistical and Solution ANOVA was because than groups involved Before ANOVA the underwent normality The of groups more than which that data normally distributed. results the ANOVA indicate a of (<0.05); it be concluded there a difference each group. test show differences the antidiarrheal activity of various doses of the ethanol extract sembung (M. leaves against male Swiss mice. For research, analyses could carried on secondary using methods as Liquid graph y. addition, is to herbal using rambat (M. micrantha) extract the can consume by public. M.

micrantha as antidiarrheal the to combined with flour a in manufacture antidiarrheal In previous feed glucomannan had good in the of Escherichia coli bacteria cause diarrhea but had little effect on stimulating the growth of Bifidobacteria and Lactobacilli [18]. Conclusions The results the extract of rambat contain tannins, flavonoids, saponins, which of secondary have properties. Ethanol of rambat M. micrantha) leaves various of mg/kg 150 mg/kg BW; and 200 mg/kg BW has antidiarrheal activities. The most dose 200 mg/kg as has ratio close to the positive control group. Acknowledgment This was by Ministry of Culture, and of the General Higher for 2021 Creativity grant. addition, we thank Ata for exceeding adequate research facilities.

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