FAKULTAS FARMASI
UNIVERSITAS MUHAMMADIYAH PURWOKERTO

PHARMACY: Jurnal Farmasi Indonesia
(*Pharmaceutical Journal of Indonesia*)

Journal homepage: https://jurnalnasional.ump.ac.id/index.php/PHARMACY

Received 30-08-2023 Accepted 17-05-2024 Available online 08-08-2024

Formulasi Sirup Ekstrak Etanol *Citrus amblycarpa* Ochse dan Aktivitas Antidiare Ekstrak dan Sirup pada Mencit yang Diinduksi *Castor Oil*

Syrup Formulation from Ethanol Extract of Citrus amblycarpa Ochse *and Antidiarrheal Activities of The Extracts and Syrup in Mice Induced by Castor Oil*

Dina Yuspita Sari*, Rini Fajaria, Ika Selvia, Jevi Asmitha Dewi, Ratna Widyasari
Program Studi DIII Farmasi, Akademi Farmasi Yarsi Pontianak, Pontianak 78237, Kalimantan Barat, Indonesia

Kata Kunci:
Antidiare, *Citrus amblycarpa* Ochse, metode proteksi, sirup

A B S T R A K

Kulit jeruk sambal (*Citrus amblycarpa* Ochse) mengandung tanin dan flavonoid yang diduga memiliki aktivitas sebagai antidiare. Penelitian ini bertujuan untuk mengetahui aktivitas antidiare ekstrak etanol kulit buah jeruk sambal (EELP), membuat formula sirup, serta menentukan aktivitas sirup ekstrak etanol kulit jeruk sambal (sirup EELP) terhadap mencit putih jantan. Simplisia kulit jeruk sambal dimaserasi menggunakan etanol 96%. Ekstrak etanol diformulasikan menjadi sirup dengan variasi konsentrasi ekstrak. Evaluasi sirup meliputi organoleptis, homogenitas, bobot jenis, viskositas, dan pH. Pengujian aktivitas antidiare terhadap ekstrak etanol dan sirup dari ekstrak etanol kulit buah jeruk sambal menggunakan metode proteksi dengan *oleum ricini* sebagai penginduksi diare. Larutan uji meliputi Na CMC (kontrol negatif), Loperamid HCl (kontrol positif), EELP dosis 0,042; 1,491; dan 2,94 g/kg BB, dan sirup EELP dengan konsentrasi 3,26; 4,26; dan 5,26%. Ketiga formula sirup menghasilkan warna hijau pekat, bau khas kulit jeruk sambal dan rasa manis sedikit pahit, homogen, pH 6, bobot jenis 1,2; dan viskositas 1,65 mPa s. Persentase antidiare ekstrak etanol kulit jeruk sambal dan sirup masing-masing sebesar 41,20; 65,11; dan 82,62%, serta 42,41; 47,08; dan 57,08%. EELP 1,492 dan EELP 2,940g/kg menunjukkan aktivitas berbeda nyata dengan kontrol negatif. Ketiga formula sirup EELP memiliki aktivitas sebagai antidiare yang ditunjukkan. Dapat disimpulkan bahwa ekstrak dan formula sirup dari ektrak etanol kulit jeruk sambal berpotensi sebagai antidiare.

Keywords:
Antidiarrheal, Citrus amblycarpa Ochse, *protection method, syrup*

A B S T R A C T

Limau citrus (Citrus amblycarpa Ochse) peel contains tannins and flavonoids that might exert antidiarrheal activity. This study aims to produce syrup formulation and evaluate the antidiarrheal activity of the ethanol extracts and their syrup in male mice. Limau citrus peels were macerated with 96% ethanol and then formulated into syrup preparations with extract concentration variations. The syrup was analyzed for organoleptic, homogeneity, specific gravity, viscosity, and pH. The in vivo antidiarrheal activity of the extracts and their syrup were examined in experimental models of castor oil-induced diarrhea. Rats were treated with Na CMC suspension (negative control), loperamide suspension (positive control), three doses of EELP (0.042, 1.491, and 2.94 g/kg BW), and three doses of EELP syrups (3.26, 4.26, and 5.26%). Syrups were deep green color, distinctive smell of limau citrus peel, sweet, slightly bitter taste; homogeneous; pH of 6; specific gravity of 1.2; and viscosity 1.65 mPa s. The three doses of EELP and the three EELP syrup formulas provided antidiarrheal activity with antidiarrheal percentages of 41.20, 65.11, and 82.62%, and 42.41, 47.08, and 57.08%, respectively. EELP 1.492g and 2,940g/kg showed a significant antimicrobial effect. All three EELP syrup formulas have significant antidiarrheal activity These findings indicate that extracts and syrup formulations display good anti-diarrheal activity.

1. Introduction

Diarrhea, an endemic disease, causes mortality in Indonesia. In 2020, 44.4% for all ages and 28.9% for children under five (Puspitarini et al., 2020). Diarrhea is a condition where a person defecates with a soft or liquid consistency; it can even be just water, and the frequency is more frequent than usual, three or more times in one day. Diarrhea is also a symptom of a digestive tract infection affected by bacteria such as *Salmonella*, and *Escherichia coli*, viruses, and parasites (Sharma et al., 2012). Plant extracts or purified bioactive compounds could be a potential alternative to prevent and treat diarrhea (Song et al., 2021).

Limau citrus (*Citrus amblycarpa* Ochse) is a type of plant from West Kalimantan that can be used as a traditional medicinal plant. Limau citrus juice was implemented as a health drink in West Kalimantan, Indonesia (Suri et al., 2022). Limau citrus contains antioxidant and antibacterial activity (el Fihry et al., 2022), anti-inflammatory activity, and enhanced metabolic disorders (Lota et al.,

Received 30-08-2023; Received in revised form 30 August 2023;
Accepted 17-05-2024; Available online 08-08-2024
E-mail address: dinayuspitasari7@gmail.com

2001). Based on scientific research, several citrus species possess antidiarrheal activity, incorporating limau citrus leaves at a dose of 0.2 and 0.3g/kg, grapefruit peel at a dose of 0.2g/kg (Pongoh et al., 2020), sweet orange peel at doses of 0.42 and 1.126g/200g (Juliana, 2017), and limau citrus peel at a dose of 0.02g/kg owned antisecretory and antimotility activities (Adeniyi et al., 2017).

Twenty-five compounds were isolated from citrus plants, ten displaying cytotoxic activity, encompassing flavones. Limau citrus peel incorporates phenolic compounds, polyphenols (Lee et al., 2022), flavonoids, limonoids, alkaloids, essential oils, pectin (Liu et al., 2021), and carotenoids (Suri et al., 2022). Sanches et al. (2022) discovered 24 phenolic components in peel from six citrus species (Sanches et al., 2022). Another study revealed that the total flavonoids and carotenoids uncovered in limau citrus peel were 41.0 and 0.737 mg g-1, respectively (Wang et al., 2022). The ethanol extract of limau citrus peel provides an antioxidant activity by employing the DPPH method with a total flavonoid content of 0.3324 mgQE/g with an inhibition of 89% (Barluado et al., 2016). Flavonoids possess antidiarrheal activity by regulating gastrointestinal motility and muscle contraction (Juliana, 2017) and curtailing fluid and electrolyte secretion. Decreased absorption of fluids in the intestine caused changes in the consistency of the feces (Khuluq and Marlina, 2021). Flavonoids and tannins denature proteins, formulating a protein tannate complex that coats the intestinal mucosa, making it more resistant and reducing the secretion (Sharma et al., 2012).

Antibacterial activity of the ethanolic extract of the limau citrus peel against *E. coli* at concentrations of 20% and 40% was unveiled by an inhibition zone of 7.2 mm (medium inhibition) (Husni et al., 2020; Amiliah et al., 2021) with a minimal inhibitory concentration (MIC) of 7.8 to 31.3 mg mL-1 (Lee and Najiah, 2009). The presence of tannins and flavonoids reduces intestinal motility and secretion during diarrhea (Adeniyi et al., 2017).

To increase the usefulness and economic value of limau citrus peel by making syrup formulas and testing their antidiarrheal activity. The syrup is a solution containing high concentrations of sucrose and other sugars. The general formula for syrup is the active substance, solvent, sweetener (sucrose and other sugars), cosolvent, preservative, and controller viscosity (Goeswin, 2008). This research aims to make syrup formulas to increase the usability and economic value of citrus peel limau citrus and test their antidiarrheal activity. Based on literature searches, research on the antidiarrheal activity of limau citrus peel, both in extract and extract form, still needs to be improved, especially when compared with other citrus species. The objective of the study is to investigate the syrup formulation of the ethanol extract of limau citrus peel and identify the antidiarrheal activity of the extract and its syrup in white male micer.

2. Materials and Tools

Materials and instruments

Limau citrus peel were purchased from Sungai Pangkalan, Bengkayang district, West Kalimantan, Indonesia, and identified in the Indonesia Institute of Science, Bogor (B-957/V/DI.05.07/12/2021). The materials used in this study were technical ethanol (96%), sucrose, propylenglycol, sodium benzoate, carboxymethyl cellulose sodium, metabisulfite sodium, and distilled water were purchased from Alkamid Co. (St. Tehran, Iran), filter paper (OneMed). The test animals were male mice (*Mus musculus* L.). The tools used in this study were analytical balances (Sartorius BL 210S), mouse balance scales (Ohaus 750SO), stirring rods, beaker glass, measuring cups, pipettes, mortar, stamper, maceration vessel, rotary evaporator (Dragon LAB RE-10 Pro), filter paper, stopwatch, oral sonde and 1 mL syringe (Onemed), gloves (Sensi), jars, mouse cages, desiccators, filter paper, Brookfield viscometers (Ntech), pycnometers, and pH meter.

Experiments

1. Extraction process and syrup formulation

This research was conducted at the Pharmacology Laboratory, Akademi Farmasi Yarsi Pontianak, from January to April 2022. The crude drugs (1591.49g) were extracted with 96% ethanol for 72 h at room temperature. After 72 h, the solution was filtered. The filtrate was concentrated in a rotary evaporator and the last trace was removed in a vacuum. The extract (EELP) was then separated into two parts (for the syrup formula (**Table I**) and to determine the antidiarrheal activity).

Na.CMC was developed with warm water to form mucilage, and the ethanol extract of limau citrus peel was diluted with propylene glycol. Sucrose, sodium benzoate, and sodium metabisulfite were dissolved in water. Sodium metabisulfite was gradually added to the mixture containing Na.CMC mucilage and then homogenized. Next, the sodium benzoate solution was homogenized. After that, limau citrus peel extract, which was dissolved in propylene glycol, was added to the solution, followed by a sucrose solution, and then homogenized. The preparation was filtered using filter paper and put into a glass bottle.

2. Evaluation of EELP syrup

The syrup formula was then calculated, encompassing organoleptic, homogeneity, specific gravity, viscosity, and pH test.

Organoleptic test

The organoleptic test used ten panelists. The examination includes color, smell, and taste (Sari et al., 2021).

pH test

The pH of syrup in each formula was measured by a pH meter. The pH meter was inserted into the syrup formulas. The experiment was performed in 10% syrup solution at 25°C (Hidayati et al., 2020; Sari et al., 2021).

Homogeneity test.

The homogeneity test is done by observing the particles visually. The syrup formulas were held on a glass plate and then observed visually for the presence of particles (Hidayati et al., 2020; Sari et al., 2021).

Viscosity test

Syrup formulas were tested for viscosity using a Brookfield viscometer. The samples were placed in the test chamber using the spindle No. 2 with a speed of 600 rpm (Hidayati et al., 2020; Sari et al., 2021).

Specific gravity test

Specific gravity was determined using a clean and dry pycnometer. At room temperature, an empty pycnometer (W1) is weighed, then filled with distilled water, and the outside of the pycnometer is wiped dry and then weighed (W2). The distilled water in the pycnometer is discarded and then dried, then the dry pycnometer is filled with the syrup preparation and then weighed (W3) (Sari et al., 2021).

$$Specific\ gravity\ (\rho) = \frac{W3-W1}{W2-W1} x\ 100\% \qquad (1)$$

Table I. Ethanol extract of limau citrus peel syrup formula

Materials	Concentration (%)			Uses
	FI	FII	FIII	
EELP	3.26	4.26	5.26	Active substance
Sucrose	67	67	67	Sweetening agent
Propylene glycol	10	10	10	Cosolvent
Sodium benzoate	0.1	0.1	0.1	Preservative
Sodium CMC	0.5	0.5	0.5	Viscosity troller
Sodiummetabisulfite	0.05	0.05	0.05	Antioxidant
Aquadest	ad 100	ad 100	ad 100	Solvent

3. Anatidiarrheal activity test of EELP and EELP syrup

The study was reviewed and approved by the ethics committee with reference number 22/II.I.AU/KET.ETIK/IV/2023. Healthy white mice (20-30g) were segregated into two experimental stages, five test groups each. The mice were treated to acclimatize for two weeks and fasted for 12 hours, and they were randomly allocated as three animals in each group. Treatment mice were screened for diarrhea by administering 0.75 mL castor oil. After one hour, animals in the different groups were treated as follows: Group I received 0.1% sodium CMC; Group II loperamide (3 mg/kg); group III EELP (0.042g/kg); group IV EELP (1,492g/kg); and group V EELP (2,940g/kg). Meanwhile, for syrup, group I received 0.1% sodium CMC; group II loperamide (3 mg/kg); group III EELP syrup (3.26%); group IV EELP syrup (4.26%); and group V EELP syrup (5.26%). The parameters observed were diarrhea onset, diarrhea frequency, feces consistency, diarrhea duration, water absorption diameter, and feces weight. Observations were performed every 30 minutes for 4 hours(Adeniyi et al., 2017). The activity of each group was expressed as percent inhibition (%) of diarrhea and was measured as follows:

$$\%\, Inhibition\ of\ defecation = \frac{A-B}{A} x\ 100\% \qquad (2)$$

where A indicates the mean number of defecations caused by castor oil and B indicates the mean number of defecations caused by loperamide, extracts, and syrup (Kola-Mustapha et al., 2019).

Data analysis

One-way ANOVA was employed to scrutinize the results at the 95% confidence interval.

3. Result And Discussion

A total of 7kg of limau citrus peel produced 1591.49g of crude drugs powder with a yield of 22.73%. The results unveiled that the three syrup formulas generated a dark green color, a characteristic smell of limau citrus peel, and a slightly bittersweet taste; homogeneous; contained a pH of 6, a specific gravity of 1.2, and a viscosity of 1.65 mPa s. The results obtained from the three formulas showed a thick green color influenced by a viscous extract that was greenish brown, a distinctive smell of limau citrus peel, and a slightly bittersweet taste. The taste generated is affected by the content of flavonoid glycoside compounds, which causes a bitter taste in citrus (Sanches et al., 2022).

The homogeneity of the preparation is affected by the solubility of each ingredient in the syrup formula. The solvent in syrup preparations uses water to ensure that all components of the ingredients in the syrup formula are dissolved in water. As the active substance, the ethanol extract of limau citrus peel is used, where the extract contains a mixture of compounds that are not yet pure. Propylene glycol was added to homogenize the preparation containing the extract. The syrup preparation's homogeneity was examined to ensure uniformity of dosage levels. Specific weight shows the ratio of the mass of a substance to the mass of the same volume of water at a specific temperature. In the syrup formula, some masses of active substances and additives affect the results of calculating the density of masses, where these substances cause an increase in the specific gravity of the three syrup formulas compared to water (Goeswin, 2008). The specific gravity produced from the three formulas has met the standard (>1.2). In the development of syrup formulations, data on the stability of the active substance and additives were required to attain the optimal pH. The pH obtained has met the pH standard for syrup (4-7). The viscosity of syrup preparations influences palatability. Thus, the results met good viscosity standards (1-3 mPa s) (Kola-Mustapha et al., 2019). The viscosity results show a low viscosity value where a liquid will flow more easily. The suitable viscosity of syrup preparations affects palatability or the ease of pouring syrup from packaged bottles so that it is easier to use (Goeswin, 2008).

Testing of antidiarrheal activity by employing the castor oil-induced method was affected by castor oil containing ricinoleic acid, which facilitates the release of endogenous prostaglandins in the gut, which caused arachidonic diarrhea, characterized by an increase in intestinal transit time and the loose feces production (Adeniyi et al., 2017). Most antidiarrheal drugs are used to reduce secretions and the propulsion of movements of the gastrointestinal smooth muscle (Alam et al., 2021). Test animals were prepared with fifteen white male mice (*Mus musculus* L.) weighing 20-30 grams. The experimental animals were habituated to the experimental environment one week before the study. Before the experiment, the test animals were fasted for 18 hours but were still given water ad libitum. Furthermore, the mice were divided into five experimental groups, each consisting of 3 mice Mice were placed in separate cages; each mouse was marked (Ayalew et al., 2022). In the antidiarrheal test, the parameters examined were onset, frequency, and duration of diarrhea, feces consistency, water absorption diameter, and feces weight (Sharma et al., 2012). The antidiarrheal parameter results, both in the extract and syrup test groups, can be observed in **Table 2**. Consistency is converted into the following scores: normal feces (0), slimy-soft feces (1), and liquid feces (3) (Adeniyi et al., 2017).

The onset of diarrhea is the time of appearance of diarrhea, which is determined by looking at the first time (minutes) the test animal has diarrhea after administration of the test solution. The sooner the onset of diarrhea, the weaker the antidiarrheal activity, and vice versa. The longer the onset of diarrhea, the stronger the antidiarrheal activity (permanent). Stool frequency was observed by counting the number of times diarrhea occurred in mice after treatment, every 30 minutes for 4 hours (Maesaroh and Marini, 2021). The smaller the value of the frequency of diarrhea, the stronger the antidiarrheal activity. The greater the frequency of diarrhea, the weaker the antidiarrheal activity (Anggraeni et al., 2020). Stool consistency is the form of feces excreted by the test animal during the observation time.

The parameter of stool consistency is determined by looking at the shape of the stool. Stool consistency is categorized into 3, namely normal, soft, and liquid. The consistency is said to be liquid when the stool is not normal, and there is much water in the stool (Susanti et al., 2017). Decreased absorption of fluids in the intestine causes a change in stool consistency from regular to more watery. The smaller the consistency value, the better the treatment group is given to improve stool consistency. Changing the consistency of the stool to become thinner indicates that the amount of fluid in the intestine is not being absorbed properly, so it will be carried away by stool that contains excess fluid (Abdillah et al., 2018). Fecal weight is the number of feces excreted by the test animal during the observation period.

The large amount of liquid in a stool can be caused by the rapid movement of the intestinal peristalsis, which causes the osmotic pressure in the intestinal cavity to increase. The smaller the fecal weight value obtained, the stronger the antidiarrheal activity (Ayalew et al., 2022). The duration of diarrhea is a parameter that is seen in how many minutes the liquid stool returns to normal (Toemon et al., 2019). The faster the change in the duration of diarrhea, the better the antidiarrheal effect. The slower the duration of diarrhea, the worse it is in reducing the duration of the stool (Abdillah et al., 2018; Ezeja et al., 2012). The water absorption diameter is a parameter that is determined by placing the feces on filter paper every 30 minutes after administration of oleum ricini and measuring the water absorption diameter on the filter paper. The total water absorption diameter was measured from the onset of diarrhea, indicated by liquid stools, until the cessation of diarrhea, characterized by normal stools (Song et al., 2021).

Table 2. Antidiarrheal parameter test results

Group	Diarrhea onset (min)	Diarrhea frequency	Feces consistency (score)	Diarrhea duration (min)	Water absorption diameter (cm)	Feces weight (g)
I (Sodium CMC)	23.33±13.31	17.66±1.52	11.66±2.51	216±0.00	14.58±1.92	1.03±0.30
II (Loperamide)	25.33±2.51	12.66±3.78	6.66±3.21	204±17.32	10.40±1.72	0.52±0.14
III (EELP 0.042 g/kg)	58.33±22.14	12.66±2.30	5.66±1.52	1512±34.64	7.37±1.11	0.62±0.05
IV (EELP 1.491 g/kg)	31.66±12.09	11.00±1.73	6.33±2.30	168±17.32	8.19±2.15	0.49±0.22
V (EELP 2.940 g/kg)	64.00±19.31	6.66±5.03	5.00±1.73	96±17.32	6.07±0.94	0.24±0.10
VI (EELP syrup 3.26%)	7.33±4.04	10.00±1.00	5.66±2.08	192.6±15.69	6.85±1.76	0.93±0.24
VII (EELP syrup 4.26%)	8.33±5.68	12.33±4.16	4.33±0.57	161.6±67.26	4.06±0.31	0.87±0.60
VIII (EELP syrup 5.26%)	9.66±5.68	11.66±2.08	4.66±3.21	180.3±85.33	5.22±4.03	0.73±0.14

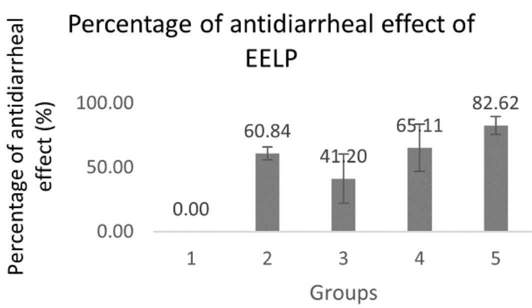

Figure 1. The diarrheal activities of ethanol extract of limau citrus peel. Group I = sodium CMC 0.1%, group II = loperamide), group III = EELP 0.042g/kg, group IV = EELP 1.491g/kg, and group V = EELP 2.940g/kg

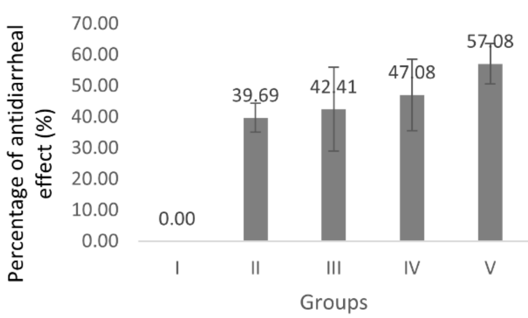

Figure 2. The diarrheal activities of syrup formulas. Group I = sodium CMC 0.1%, group II = loperamide), group III = EELP syrup 3.26%, group IV = EELP syrup 4.26%, and group V = EELP syrup 5.26%

When diarrhea occurs, the large amount of water excretion is caused by intestinal peristalsis that helps push excess fluid into the large intestine. In the colon, fluid absorption will occur, and if the fluid capacity exceeds the absorption capacity, the liquid will come out together so that much fluid will come out (Debora et al., 2016).

The antidiarrheal activities of ethanol extract of limau citrus peel and their syrup are presented in **Figures I - II**. The data obtained were investigated by employing the one-way ANOVA test ($p<0.05$), normally distributed and homogeneous ($p>0.05$). The significance value between groups in the two test groups (extract and syrup) was $p<0.05$, indicating that there was a difference in the average antidiarrheal effect. In identifying the differences between groups in each test group, the Least Significance Different (LSD) test was administered. Based on the LSD test in the extract test group, the positive control group, EELP 1.492g/kg, and EELP 2.940g/kg displayed significantly different results from the negative control ($p<0.05$). The EELP 0.042, 1.492, and 2,940g/kg were not significantly different from the positive control group ($p>0.05$) (**Figure 1**). Based on the LSD test in the syrup test group, the positive control group and the three syrup formulas uncovered significantly different results from the negative control ($p <0.05$), and the three syrup formulas were not significantly different from the positive control group ($p>0.05$) (**Figure 2**).

The flavonoid secondary metabolite compounds generated in the extract possess antidiarrheal activity by preventing the release of acetylcholine in the digestive tract, which causes reduced activation of nicotic acetylcholine receptors (especially Ach-M3), regulating gastrointestinal motility and muscle contraction. The cause of the difference in the decrease in diarrhea frequency can be due to the food processed in the digestive system of each mouse differently (Juliana, 2017). Decreased absorption of fluids in the intestine caused changes in the consistency of the feces. It is due to the rapid intestinal peristalsis that the osmotic pressure in the intestinal cavity is enhanced. The decrease in the duration of feces in each group was different due to differences in the ability to process food entering the digestive tract in each mouse (Khuluq and Marlina, 2021). The total diameter of water absorption was calculated from the onset of diarrhea indicated by liquid feces to the cessation of diarrhea identified by normal feces. The amount of water expenditure during diarrhea was precipitated by intestinal peristalsis, which helps to push excess fluid into the large intestine. In the colon, if the fluid capacity exceeds the absorption capacity, much fluid will occur (Juliana, 2017).

Phytochemical screening of extract limau citrus peel uncovered the presence of flavonoids and tannins (Widyasari et al., 2018), which possess antidiarrheal activity. They denatured proteins, formulating a protein tannate complex, which coats the intestinal mucosa, making it more resistant and reducing secretion (Sharma et al., 2012). Flavonoids also contain an antidiarrheal effect by restricting intestinal motility, thereby curtailing fluid and electrolyte secretion (Khuluq and Marlina, 2021).

4. Conclusion

It can be concluded that the results of this study showed that The EELP and EELP syrup had in vivo antidiarrheal activities. The ethanol extract of limau citrus peel (EELP) 0.042, 1.491, and 2.94 g/kg, and their syrup (EELP syrup) 3.26, 4.26, 5.26% possessed antidiarrheal activity with antidiarrheal activity percentages of 41.20, 65.11 and 82.62% and 42.41, 47.08, and 57.08%, respectively. The results might indicate that flavonoids and tannins are responsible for the effects. Furthermore, the findings of this study are encouraging and may be used as the basis for further studies in the area, including pharmaceutical technology. These results imply that the extracts and their syrup products occupy the potential to be alternative herbal medicines with antidiarrheal properties.

5. References

Abdillah R, Eriadi A, Khasanah YN. 2018. Aktivitas antidiare ekstrak etanol herba greges otot (Equisetum debile Roxb.) pada mencit putih jantan. *Jurnal Farmasi Higea*, 10, 33–40.

Adeniyi OS, Omale J, Omeje SC, Edino VO. 2017. Antidiarrheal activity of hexane extract of Citrus limon peel in an experimental animal model. *Journal of Integrative Medicine*, 15, 158–164.

Alam MM, Emon NU, Alam S, Rudra S, Akhter N, Mamun MMR, Ganguly A. 2021. Assessment of pharmacological activities of Lygodium microphyllum Cav. leaves in the management of pain, inflammation, pyrexia, diarrhea, and helminths: In vivo, in vitro and in silico approaches. *Biomedicine &*

Pharmacotherapy, 139, 111644.

Amiliah, Nurhamidah, Handayani D. 2021. Aktivitas antibakteri kulit buah jeruk kalamansi (Citrofortunella microcarpa) terhadap bakteri Staphylococcus aureus dan Escherichia coli. *Jurnal Pendidikan dan Ilmu Kimia*, 5, 92–105.

Anggraeni R, Alfiar I, Suhendi H. 2020. Uji aktivitas daun mareme (Glochidion borneense (Müll Arg) Boerl) sebagai antidiare pada mencit jantan galur swiss webster. *Jurnal Farmasi Muhammadiyah Kuningan*, 5, 59–69.

Ayalew M, Bekele A, Mengistie MG, Atnafie SA. 2022. Evaluation of the antidiarrheal activity of 80% methanol extract and solvent fractions of the leaf of Bersama abyssinica Fresen (Melianthaceae) in mice. *BMC Complementary Medicine and Therapy,* 22, 8.

Barluado MJG, Lagang MJM, Gordonas IFM, Bosas CDB. 2016. Antiangiogenic and antioxidant properties of calamansi Citrus microcarpa peel ethanolic extract. *UIC Research Journal*, 9, 123–134.

Debora N, Prabowo WC, Ibrahim A, Rijai L. 2016. Uji efek antidiare kombinasi ekstrak biji pepaya (Carica papaya L.) dengan daun kesumba keling (Bixa orellana L.) pada mencit (Mus musculus), in: *Proceeding of the 4th Mulawarman Pharmaceuticals Conferences*. Fakultas Farmasi, Universitas Mulawarman, Samarinda.

Ezeja IM, Ezeigbo II, Madubuike KG, Udeh NE, Ukweni IA, Akomas SC, Ifenkwe DC. 2012. Antidiarrheal activity of Pterocarpus erinaceus methanol leaf extract in experimentally-induced diarrhea *Asian Pacific Journal of Tropical Medicine*, 5, 147–150.

El Fihry N, El Mabrouk K, Eeckhout M, Schols HA, Filali-Zegzouti Y, Hajjaj H. 2022. Physicochemical and functional characterization of pectin extracted from Moroccan citrus peels. *LWT*, 162, 113508.

Goeswin, A., 2008. *Pengembangan Sediaan Farmasi*. ITB Press, Bandung.

Hidayati N, Styawan A, Khotimah K. 2020. Formulasi dan uji sifat fisis sirup ekstrak etanol daun sukun (Artocarpus altilis) (Parkinson ex F.A.Zorn) Fosberg, in: *The 12th University Research Colloqium 2020*. Stikes Muhammadiyah Klaten, Klaten.

Husni E, Ismed F, Afriyandi D. 2020. Standardization study of simplicia and extract of calamondin (Citrus microcarpa Bunge) peel, quantification of hesperidin and antibacterial assay. *Pharmacognosy Journal*, 12, 777–783.

Juliana RA. 2017. Uji efek antidiare ekstrak kulit jeruk manis (Citrus sinensis L.) pada tikus putih jantan (Rattus novergicus) yang diinduksi magnesium sulfat. *Skripsi*. Politeknik Kesehatan Palembang, Palembang.

Khuluq H, Marlina E. 2021. Evaluation of antidiarrheal effect of combination of salam leaves (Syzygium polyanthum) and jackfruit leaves (Artocarpus heterophyllus Lam.) infusum in rats induced by castor oil. *Pharmacy Education*, 148–151.

Kola-Mustapha AT, Ghazali YO, Ayotunde HT, Atunwa SA, Usman SO. 2019. Evaluation of the antidiarrheal activity of the leaf extract of Parquetina nigrescens and formulation into oral suspensions. *Journal of Experimental Pharmacology*, 11, 65–72.

Lee GJ, Lee SY, Kang NG, Jin MH. 2022. A multi-faceted comparison of phytochemicals in seven citrus peels and improvement of chemical composition and antioxidant activity by steaming. *LWT*, 160, 113297.

Lee SW, Najiah M, Wendy W, Nadirah M. 2009. Antibiogram and heavy metal resistance of pathogenic bacteria isolated from moribund cage cultured silver catfish (Pangasius sutchi) and red hybrid tilapia (Tilapia sp.). *Frontiers of Agriculture in China,* 8, 880–886.

Liu N, Li X, Zhao P, Zhang X, Qiao O, Huang L, Guo L, Gao W. 2021. A review of chemical constituents and health-promoting effects of citrus peels. *Food Chemistry*, 365, 130585.

Lota ML, de Rocca Serra D, lix Tomi F, Casanova J. 2001. Chemical variability of peel and leaf essential oils of 15 species of mandarins. *Biochemical Systematics and Ecology*, 29(1), 77-104.

Maesaroh I, Marini. 2021. Uji aktivitas anti diare sirup liofilisat ekstrak buah harendong (Melastoma affine D. Don) terhadap mencit (Mus musculus L.). *Jurnal Ilmiah Manuntung*, 7, 12–20.

Pongoh GS, Hariyadi, Maarisit W, Tapehe Y. 2020. Uji efektivitas ekstrak kulit buah jeruk bali Citrus maxima sebagai antidiare pada tikus putih jantan Rattus norvegitus. *Jurnal Biofarmasetikal Tropis*, 3, 39–45.

Puspitarini PMA, Pratama IS, Suryadi BF. 2020. Anti-diarrheal activity of aqueous extract of nagasari flowers (Mesua ferrea L.) in BALB/c mice induced by Escherichia coli. *Journal of Pharmaceutical Sciences and Community*, 16, 50–55.

Sanches VL, Cunha TA, Viganó J, de Souza Mesquita LM, Faccioli LH, Breitkreitz MC, Rostagno MA. 2022. Comprehensive analysis of phenolics compounds in citrus fruits peels by UPLC-PDA and UPLC-Q/TOF MS using a fused-core column. *Food Chemistry X*, 14, 100262.

Sari D, Ariansyah S, Shinta S, Beniardi W. 2021. Face tonic formulation from ethanol extract of Maranta arundinacea L. With variety of cosolvent and surfactant: Propylene glycol and polysorbate 80, in: Mursidi, A. (Ed.), *The Economic Optimism 2021 Perspectives: Education, Business, Health, Engineering and Technologi, Productive Enterprises and Law*. STKIP Singkawang, Singkawang. 34–39.

Sari DY, Widyasari R, Puspita W. 2021. Formulasi hair tonic dari ekstrak etanol, fraksi etanol, dan fraksi kloroform-metanol kayu secang (Caesalpinia sappan L.). *Jurnal Farmasi Indonesia*, 18, 109–120.

Sharma P, Vidyasagar G, Bhandari A, Singh S, Bhadoriya U, Ghule S, Dubey N. 2012. Pharmacological evaluation of the antidiarrhoeal activity of leaves extract of Murraya koenigii in experimentally induced diarrhoea in rats. *Asian Pacific Journal of Tropical Disease*, 2, 230–233.

Song Y, Luo Y, Yu B, He J, Zheng P, Mao X, Huang Z, Luo J, Luo Y, Yan H, Wang Q, Wang H, Chen D, Yu J. 2021. Tannic acid extracted from gallnut prevents post-weaning diarrhea and improves intestinal health of weaned piglets. *Animal Nutrition*, 7, 1078–1086.

Suri S, Singh A, Nema PK. 2022. Current applications of citrus fruit processing waste: A scientific outlook. *Applied Food Research*, 2, 100050.

Susanti M, Wijayanti R, Dwi A, Resty D, Nurferawati D. Aeni S. 2017. Aktivitas antibakteri in vitro dan efektivitas antidiare in vivo ekstrak biji carica (Carica pubescens) pada mencit jantan (Swiss Webster) yang diinduksi minyak jarak. *Jurnal Farmasi Sains dan Praktis*, 3, 29–38.

Toemon A, Bertilova A, Widiarti A, Mutiasari D. 2019. Pengaruh pemberian ekstrak etanol buah masisin (Rhodomyrtus tomentosa Wight) terhadap frekuensi, konsistensi, dan durasi diare pada hewan coba mencit putih jantan (Mus musculus) yang diinduksi oleum ricini. *Jurnal Surya Medika*, 5, 22–40.

Wang MM, Wang F, Li G, Tang MT, Wang C, Zhou QQ, Zhou T, Gu Q, 2022. Antioxidant and hypolipidemic activities of pectin isolated from citrus canning processing water. *LWT*, 159, 113203.

Alam U, Fawwad A, Shaheen F, Tahir B, Basit A, Malik RA, 2017. Improvement in neuropathy-specific quality of life in patients with diabetes after vitamin D supplementation. *Journal of Diabetes Research*, 2017, 7928083.