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EFFECTIVENESS OF ANTI-LICE SHAMPOO FROM LEMONGRASS STALK (*Cymbopogon citratus*)

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Abstract: Head lice (*Pediculus humanus capitis*) are ectoparasites that attack the human head, causing excessive itching. To overcome this problem, shampoo with active substances from natural sources can be used, namely, kitchen lemongrass (*Cymbopogon citratus*). This study was designed to determine the effects of various concentrations of lemongrass stalk extract and the optimal concentration of the anti-head lice shampoo on head louse mortality. The results of the phytochemical screening revealed that the lemongrass stalk extract contains flavonoids, tannins, terpenoids, saponins and alkaloids. Lemongrass stalk extract can be used as a shampoo formulation with several concentrations ranging from 3%, 5%, 7% and 9%. The characteristic results revealed a thick liquid, homogeneous, brown to blackish brown color, a characteristic smell of the lemongrass stalk, a foam height range of approximately 6–5 cm, a pH range of approximately 5–6, and a viscosity range of approximately 100–5700 cP. The optimum concentrations that can kill head lice effectively and meet the SNI requirements for an adequate and safe shampoo to use are shampoo at concentrations of 5% and 7%.

Keywords: Shampoo, Lemongrass, Antihead lice

Abstrak: Kutu rambut (*Pediculus humanus* capitis) merupakan ektoparasit yang menyerang kepala manusia sehingga menimbulkan rasa gatal yang berlebihan. Untuk mengatasi hal tersebut dapat digunakan sampo dengan bahan aktif yang berasal dari bahan alam yaitu serai dapur (*Cymbopogon citratus*). Penelitian ini dirancang untuk mengetahui karakteristik sampo anti kutu rambut dari variasi konsentrasi ekstrak batang serai dapur dan konsentrasi optimum pada sampo anti kutu rambut terhadap mortalitas kutu rambut. Berdasarkan hasil skrining fitokimia terbukti bahwa ekstrak batang serai dapur mengandung senyawa flavonoid, tanin, terpenoid, saponin dan alkaloid. Ekstrak batang serai dapur dapat digunakan sebagai formulasi sampo dengan beberapa variasi konsentrasi berkisar 3%, 5%, 7% dan 9%. Hasil karakteristik yang diperoleh yaitu berupa cairan kental, homogen, berwarna coklat hingga coklat kehitaman, memiliki bau khas batang serai dapur, kisaran tinggi busa sekitar 6-5 cm, kisaran pH sekitar 5-6, kisaran viskositas sekitar 100-5700 cP. Konsentrasi optimum yang mampu membunuh kutu rambut secara efektif dan memenuhi persyaratan SNI-06-2692-1992 untuk sampo yang memadai dan aman digunakan adalah sampo dengan konsentrasi 5% dan 7%.

Kata kunci: Sampo, Serai dapur, Anti kutu rambut

INTRODUCTION

Pediculosis is an ectoparasite caused by blood-sucking lice that attacks human skin. Among the three species of lice that attack humans, namely, body lice (*Ediculus humanus humanus*), head lice (*Pediculus humanus capitis*) and pubic lice (*Thirus pubis*) (Light *et al.,* 2008), head lice are the most important because they are considered very common (Heukelbach and Feldmeier, 2004). The global prevalence of head lice among school students in low- and middle-income countries is estimated to be 19.96% (Delie *et al.*, 2024). The most dominant factor in the incidence of pediculosis capitis is personal hygiene. Research has shown that children with poor personal hygiene are 8.713 times more likely to develop pediculosis capitis than are children with good personal hygiene (Sitorus *et al.,* 2020).

Head lice, which can suck blood, can cause anemia and itching. Itching can make it difficult for children to sleep and disrupt their concentration on learning, which can result in decreased performance at school. Head lice infestations cause high levels of anxiety among parents and children and can lead to negative social and psychological impacts such as stress and anxiety. Recent research findings support the cognitive–behavioral determinants of HLI transmission and prevention. Perceived collective family efficacy, perceived barriers, perceived self-efficacy, and individual response efficacy are important in predicting head lice prevention behavior. In addition, chronic lice lesions caused by lice bites can lead to pathogenic bacterial infections that can worsen a subject's condition (Babazadeh *et al.,* 2023). Therefore, solutions are needed to overcome the problems caused by small parasites found in the hair and scalp.

The number of human head lice can be controlled mechanically by keeping the scalp clean using antilice shampoo. Shampoos are generally made from chemicals, causing problems for the health of the scalp and hair (George and Potlapati, 2021). Pediculosis treatment generally uses synthetic chemical products such as lindane, pyrethrin, permethrin, and malathion (Iwata and Shimada, 2013). However, these substances are very persistent and toxic (Ahmad *et al.*, 2024) and cause the development and spread of resistant head lice populations (Soonwera *et al.,* 2018).

To overcome this, herbal shampoo is needed as an alternative to reduce the problems caused by shampoo-containing chemicals. Many medicinal plants have been reported to have beneficial effects on hair and are commonly used in shampoo

formulations. Topical ivermectin demonstrated high effectiveness in killing head lice, with a success rate of 86.2% after one application and 100% after the second application (Karthikeyan *et al.,* 2022). Natural pediculosides can be obtained from various kinds of plants, including medicinal plants containing active substances such as secondary metabolites believed to be able to treat various diseases (Chaachouay and Zidane, 2024). Research has shown that pure herbal shampoo has excellent performance, equivalent to that of commercially available shampoo (SK *et al.*, 2020). Another study revealed that a combination of several herbal plant extracts is promising as an ovicidal agent for controlling head lice (Soonwera, *et al.*, 2018). One of the herbal plants that can be used as an alternative for making shampoo is the kitchen lemongrass stalk.

Kitchen lemongrass (*Cymbopogon citratus*) is a natural insecticide widely used as a spice. Lemongrass plants in the health sector can be used as laxatives for urine, phlegm, sweat and body warmers to increase appetite and as a postpartum treatment to reduce fever and relieve convulsions. Several studies have been conducted on citral content, namely, geranial and neral (Vazquez-Briones, *et al.*, 2015), in lemongrass, which can be used as insecticides. The geranial content in the stalk was greater than that in the leaves. Lemongrass stalks contain 91.67% citral compounds, consisting of 55.172% geranial and 36.501% neral (Hartatie *et al.*, 2020). Research has shown that the essential oil aromatherapy of lemongrass candle is effective as an anti-mosquito agent against *Aedes aegypti,* with the active substance citronellal as a mosquito repellent (Dewi and Lusiyana, 2020). Lemongrass plant extract is effective in killing *Aedes* sp. larvae (Giroth *et al*., 2021) and effective in dealing with rice weevil pests (Rohma *et al*., 2021). Lemongrass extract has been used to reduce the mortality of termites (*Macrotermes gilvus* Hagen) via several application techniques (Yazid, 2024).

On the other hand, lemongrass extract is less convenient to use directly on hair because it can make hair sticky and tangled, so it is needed in an easier and more practical form, such as shampoo. Shampoo is a cosmetic used to clean hair and scalp to be clean and healthy. Commercial shampoos have several forms, such as liquids, gels or emulsions containing active substances, thickeners, moisturizers, preservatives and surfactants, which can produce foam (Pravitasari *et al.,* 2021). Things that must be considered when making shampoos include appropriate comparisons, preparation evaluations, and shampoo preparation

effectiveness. Therefore, shampoo can be used safely and healthily (Surani and Aliza, 2017).

Based on the background, this research was conducted to make shampoo from kitchen lemongrass stalk extract (*Cymbopogon citratus)* and to determine the effectiveness of mortality on head lice (*Pediculus humanus capitis*).

METHOD

Tools and materials

The tools used in this research include Fourier transform infrared (FTIR) (Thermo Fisher Scientific IS 50), a rotary evaporator (RE-100 Pro D-LAB), an analytical balance, a viscometer (VM-BF-RV-01) (Brookfield type RVT), a pH meter, a hotplate, a blender, and glassware.

The materials used in this research were distilled water (H_2O), iron (III) chloride ($FeCl_3$) 5% and 1%, lemongrass stalk (*Cymbopogon citratus*), ethanol (C_2H_5OH) 96%, hydroxyprophyl methylcellulose ($CH_3CH(OH)CH_2$), filter paper, methyl paraben ($C_8H_8O_3$), sodium lauryl sulfate, Dragendroff's reagent, Mayer's reagent, Liebermann-Burchard's reagent, Wagner's reagent, and head lice (*Pediculus humanus capitis*).

Work procedures

Extraction of lemongrass (Cymbopogon citratus) stalks

Samples of the lemongrass stalks were washed and dried at room temperature. The stalk was cut into small pieces and mashed with a blender. Ethanol (96% solvent) was added to a 1 kg sample until it was submerged, after which the maceration process was carried out in a closed container for 24 hours. The filtrate was filtered with gauze, and the residue was soaked again with the same solvent. This process was carried out 3 times. All filtrates were collected and evaporated on a rotary evaporator at a temperature of 59 °C until a thick extract was obtained (Tee and Badia, 2019) and (Soraya, *et al*., 2016)

Phytochemical screening

Phytochemical screening was designed to determine the secondary metabolite contents in the sample. Phytochemical screening includes flavonoid tests, saponin tests, tannin tests, alkaloid tests and terpenoid tests. Flavonoid testing was carried out by adding 5% $FeCl_3$ to the sample extract. The saponin test was carried out by adding 10 mL of water to the sample extract and shaking vigorously. The tannin test was carried out by adding 1% iron(III) chloride ($FeCl_3$) to the sample extract. The alkaline test was carried out by

using Mayer's reagent, Wagner's reagent and Dragendroff's reagent. The terpenoid test was carried out by using Liebermann–Burchard reagent. All of the changes were observed.

Fourier Transform Infrared (FTIR) Test

FTIR analysis was carried out to determine the functional groups associated with the presence of citronellal compounds in the lemongrass stalk extract. The sample was crushed until smooth and then put into a mold to form pellets, which were analyzed at a wavelength of 400 – 4000 cm^{-1}.

Shampoo Formulation

Shampoo was made with the formulations presented in Table 1 (Eryaputri *et al.*, 2023 and Tee and Badia, 2019) below:

Table 1. Anti-lice shampoo formula

Material	Concentration (%)				
	F0	**F1**	**F2**	**F3**	**F4**
Lemongrass stalk extract	0	3	5	7	9
Sodium lauryl sulfate	4	4	4	4	4
HPMC	1	1	1	1	1
Methyl paraben	0.5	0.5	0.5	0.5	0.5
Aquades	50 mL	50 mL	50 mL	50 mL	50 mL

(Source: (Eryaputri *et al.*, 2023 and Tee and Badia, 2019)

Characteristics of the shampoo preparation

The characteristics of the shampoo preparations included organoleptic test results, homogeneity, pH stability, foam height and viscosity. An organoleptic test consists of identifying shampoo preparations by observing their form, color and aroma. The homogeneity test was carried out by shaking the shampoo mixture in triplicate. The pH stability test was carried out by weighing 1.0 g of the sample extract, dissolving it in 10 mL of distilled water and measuring it with a digital pH meter, which was carried out 3 times. The viscosity test was carried out using a Brookfield viscometer at 50 rpm in 100 mL. The foam height test was carried out by placing 0.1 g of shampoo into a test tube. Then, 10 mL of distilled water was added, and the test tube was shaken for 20 seconds.

Testing the Effectiveness of Anti-Lice

Testing the effectiveness of the antilice effect of the shampoo was carried out by preparing filter paper in a Petri dish and then dripping it evenly with 0.5 mL of shampoo extract. After that, 5 head lice were placed in a Petri dish, covered and observed every 5 minutes for 2 hours 3 times.

RESULTS AND DISCUSSION

The production of antihead louse shampoo from lemongrass stalk extract consists of several stages, namely, sample extraction, phytochemical screening, FTIR analysis, shampoo characterization and lice effectiveness testing.

Results of secondary metabolite content identification

Table 2. Phytochemical screening test results

Active compound	Reagent	Observation	Information
Flavonoids	$FeCl_3$ 5%	Blackish yellow	Positive
Saponins	Aquades	Foam	Positive
Terpenoids	Liebermann-Burchard	Brownish ring	Positive
Tannins	$FeCl_3$ 1%	Blue	Positive
Alkaloids	Wagner	White precipitate	Positive
	Mayer	Brown precipitate	Positive
	Dragendorff	Red precipitate	Positive

Phytochemical screening was carried out to determine the contents of secondary metabolite compounds contained in the shampoo extract. The test results revealed that it contained flavonoids, saponins, terpenoids, tannins and alkaloids. This finding is consistent with several studies revealing that lemongrass contains tannins, flavonoids and steroids (Hamad *et al.,* 2019) and (Alfiyanti *et al.,* 2023).

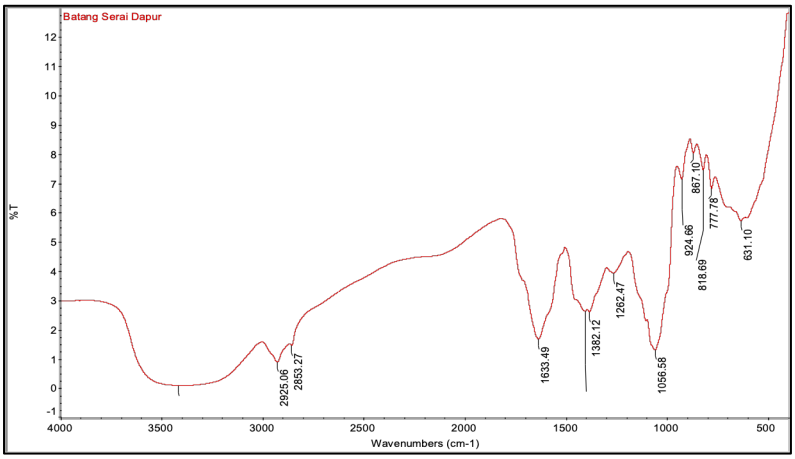

Figure 1 FTIR spectrum of the lemongrass stalk extract

The FTIR test was designed to determine the functional groups contained in the sample. The FTIR spectrum (Figure 1) revealed the secondary metabolite content contained in the lemongrass stalk extract. The peak at 2925.96 cm^{-1} was

attributed to -CH_3 stretching as a saturated aliphatic alkyl group. At 2853.27 cm^{-1}, the asymmetric stretching of -CH_2 was observed. Stretching of the C=O aldehyde group was observed at 1633.49 cm^{-1}. The peak at 1382.12 cm^{-1} was attributed to methyl C-H symmetric bending. At 818.69 $cm^{-1,}$ a 1,3 distribution and a 1,4 distribution were observed. These results are similar to those of other studies (Vazquez-Briones, *et al.*, 2015 and Wany *et al.*, 2014) that indicated that citral (geranial and neral) was obtained.

Table 3. Characteristics of the antiharm lice shampoo

Characteristics	Shampoo Concentration (%w/v)				
	0	3	5	7	9
Organoleptic:					
Color	Chocolate	Dark chocolate	Dark chocolate	Dark chocolate	Dark chocolate
Being	Viscous liquid	Viscous liquid	Viscous liquid	Viscous liquid	Viscous liquid
Smell	Odorless	Lemongrass stalk odor	Lemongrass stalk odor	Lemongrass stalk odor	Lemongrass stalk odor
Homogeneity	Homogeneous	Homogeneous	Homogeneous	Homogeneous	Homogeneous
Foam height	5.27 ±0.36	5.68 ± 0.24	5.83 ± 0.22	6.16 ± 0.22	6.33 ± 0.22
pH	6.98 ± 0.026	5.64 ± 0.01	5.47 ± 0.01	5.38 ± 0.006	5.37 ± 0.066
Viscosity	100 cP	200 cP	2700 cP	3700 cP	5700 cP

Characteristics of shampoo preparations

An organoleptic test is a physical parameter used to determine the stability of a shampoo. Organoleptic analysis was carried out by observing changes in the shape, odor and color of the shampoo. The results of the organoleptic observations in Table 3 revealed that the greater the concentration of shampoo, the stronger the color and smell of the lemongrass stalk, and the greater the amount of foam in the shampoo preparation. The shampoo preparation produced a thick liquid, and no sediment formed.

Figure 2. (a) Lemongrass extract, (b) Lemongrass shampoo

The homogeneity test results revealed that the shampoo preparation at each concentration was homogeneous, which was characterized by being evenly mixed and having no small particles. The results of the foam height test revealed that

the greater the concentration of shampoo was, the greater the amount of foam produced. This was influenced by the surfactant and saponin contents of the lemongrass stalks. The results of the foam height test at all concentrations met the foam height requirements, namely, 1.3 – 22.0 cm.

The pH test revealed that the higher the concentration was, the lower the acidity of the shampoo preparation. Other studies (Rahayu and Mentari, 2024) revealed that increasing the concentration of shampoo extract led to a pH closer to the ideal range. Even though the pH of this lemongrass shampoo has decreased, it still fulfills the acidity requirements set by the Indonesian National Standard (SNI) 06-2692-1992 (Badan Standarisasi Nasional, 1992), namely, 5.0 – 9.0.

A viscosity test was carried out to observe and measure the thickness of the shampoo preparation. The results of the viscosity test shown in Table 4 revealed that the concentration of shampoo extract can affect the viscosity of shampoo. The analysis results revealed that shampoo with adequate viscosity SNI required concentrations of 5% and 7%, namely, in the range of 400 – 4000 centipoise (cP) (Tee and Badia, 2019).

Table 4. Test of the effectiveness of the antilice shampoo

Formulation	Time (min)						Number of dead lice
	5	**10**	**15**	**20**	**25**	**30**	
Shampoo	-	-	-	-	1	-	1
0%	-	-	-	-	-	-	-
	-	-	-	-	-	1	1
Shampoo	1	1	-	1	1	1	5
3%	1	1	1	1	1	-	5
	-	1	1	1	1	1	5
Shampoo	2	1	-	1	1	-	5
5%	3	1	1	-	-	-	5
	2	3	-	-	-	-	5
Shampoo	5	-	-	-	-	-	5
7%	5	-	-	-	-	-	5
	4	1	-	-	-	-	5
Shampoo	5	-	-	-	-	-	5
9%	5	-	-	-	-	-	5
	5	-	-	-	-	-	5
Aquadest	-	-	-	-	-	-	5
	-	-	-	-	-	-	5
	-	-	-	-	-	-	5
Peditox	1	1	-	1	1	1	5
	2	3	-	-	-	-	5
	1	1	1	2	-	-	5

The effectiveness of the lemongrass shampoo test can be seen in Table 4. The use of distilled water as a negative control resulted in no dead head lice, and the use of

peditox as a positive control with a 1% permethrin content killed 5 lice. Shampoo at a 0% concentration killed only 1 head louse. Compared with the negative control, shampoo at concentrations of 3%, 5%, 7% and 9% killed 5 head lice faster. This finding revealed that the greater the concentration of shampoo was, the less time was taken to kill the lice.The active substances that can kill head lice are flavonoids, terpenoids, saponins, tannins and alkaloids. Flavonoids are antioxidant compounds that kill head lice because they can act as stomach poisons by disrupting digestive organs, resulting in biotransformation during the metabolism of head lice (Sulistiyana *et al*., 2022). Tannins also enter an insect's body as a poison in the digestive system by binding proteins to produce complex proteins, causing head lice to lose their appetite so that their growth rate and survival ability decrease.

Saponins can enter the body of lice in two ways, namely, through respiratory system and physical contact. Saponins inhibit enzymes in the respiratory process, causing damage to the cell membrane and the death of lice. Alkaloids can inhibit acetylcholinesterase enzymes in the nervous respiratory system of lice (Sulistiyana *et al*., 2022).

One of the active components in kitchen lemongrass stalks is citral (Hartatie *et al.*, 2020), an essential oil terpenoid that can act as a repellent for head lice and insects (Plata-Rueda *et al.*, 2020 and Li *et al.*, 2020). The essential oil, which is a desiccant poison, can kill lice via direct contact. The poison enters the lice's body area through the exoskelet (hard framework on the outside of the organism) by the tarsus (toes) and causes the lice's body to continuously lose fluids, causing unresolved evaporation of water in the body. In the end, the lice die because of dryness (Meri *et al.,* 2017).

CONCLUSION

The characteristics of the antihead lice shampoo from the lemongrass stalk extract and the lemongrass (*Cymbopogon citratus)* stalk extract are brown to blackish in color, have a characteristic baying smell of the lemongrass, form a thick, homogeneous liquid, have a foam height of 5 – 6, a stable degree of acidity (pH) of 6 – 5 and a viscosity of 100 – 5700 cP. The optimum concentration of antiharm lice shampoo from lemongrass (*Cymbopogon citratus)* stalk extract for reducing head louse mortality and meeting the requirements of SNI-06-2692-1992 is 5% and 7% shampoo.

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