

Formulation of An Anti-lice Shampoo Soursop Leaves Extract (*Annona muricata* L)

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Abstract. Soursop (*Annona muricata* L.) leaves contain acetogenin which is effective for killing head lice (*Pediculus humanus* Linnaeus). Head lice can be treated using an anti-lice shampoo. To get a shampoo with good viscosity and pH, variations in the concentration of carbopol and citric acid are used. The purpose of this study was to determine variations of carbopol and citric acid which have the best physical properties and are the most effective in dealing with head lice. Soursop leaves extract is made in 3 formulas shampoo with variations of carbopol: citric acid. Formula I 2%: 0.1%, Formula II 3%: 0.2%, and Formula III 4%: 0.3%. Then evaluated by organoleptic, pH, viscosity and foam stability. Anti-lice effectiveness testing was carried out by looking at the number of head lice deaths for 5 minutes for 3 formulas, distilled water as negative control, and Peditox[®] as positive control. The results showed that shampoo with 2% carbopol: 0.1% citric acid produced the best physical properties with a pH value was 6 and viscosity value was 2.7dPas and was able to kill head lice in the first minute. Least Significance Different test results showed that no difference compared to Peditox[®] ($p > 0.05$).

1. Introduction

Head lice (*Pediculus humanus* Linnaeus) or head lice infection is a problem that often occurs in children aged 3-11 years in the world. The prevalence of head lice (*Pediculus humanus* Linnaeus) in several countries in the world is still quite high. In Jordan 26.6% of elementary school children suffer from head lice [1]. The prevalence of head lice in the Indian tribe is 28.3% and Malays 18.9% higher when compared to the Chinese tribe which is 4.6%. Lice infestation in East Bangkok averaged 23.32% more in girls 47.12% than boys 0% [2]. Whereas in Indonesia, there has not yet been a general incidence of head lice and in school-age children [3].

Hair lice (*Pediculus humanus* Linnaeus) is an ectoparasite that lives on the human scalp. Head lice (*Pediculus humanus* Linnaeus) that occur can be symptomatic or asymptomatic. Symptomatic state, itching will be found in a high percentage of variables in patients [4].

Head lice also tend to be resistant to conventional hair treatments. The tendency of resistance and toxic hazards makes the importance of safe but effective alternative treatments in eradicating head lice [3]. Some families in Indonesia treat head lice with insect poisons or anti-lice medicines without regard to safety principles.

Indonesia is a country rich in plant diversity which has potential as a medicinal plant, including soursop leaves (*Annona muricata* L.). The content of acetogenin compounds (asimicin, integer, and squamosin) in soursop leaves as a parasitic biopesticide that can kill head lice [5].



Shampoo is the most suitable and easy to use anti-lice preparation. An anti-lice shampoo requires a special formulation [6]. In the shampoo formulation needed appropriate additional ingredients. The most influential ingredient is carbopol because it can affect the physical properties of the resulting gel. According to carbopol can increase the viscosity of a shampoo preparation because carbopol with a small concentration can produce a shampoo with a high viscosity [7].

This study aims to determine the formula of soursop leaves extract which is good based on variations in the concentration of carbopol and citric acid and its effectiveness in killing head lice.

2. Methods

One thousand gram of soursop leaves were obtained from Basin, Kebonarum, Klaten, Central Java, Indonesia. Soursop leaves were blended and macerated with 96 % of ethanol for 5 days. The solution then filtered with flannel, to obtain a thin filtrate. The aqueous filtrate is then evaporated on the water bath until all the ethanol has evaporated and a thick filtrate is produced. The resulting viscous extract was then made into 3 variations of the shampoo formula with each composition as shown in Table 1:

Table 1 Formulation of an anti-lice shampoo soursop leaves extract

Material	Account (g)		
	FormulaI	FormulaII	FormulaIII
Extract	25	25	25
Carbopol	2	3	4
Citric Acid	0.1	0.2	0.3
Metil Paraben	0.4	0.4	0.4
<i>Sodium lauryl sulphate</i> (SLS)	10	10	10
<i>Cocamidopropyl betaine</i>	10	10	10
Parfume	q.s	q.s	q.s
Distilled water	100 ml	100 ml	100ml

Physical property tests carried out on each shampoo formula include:

Organoleptic Test. Shampoo preparations that have been made are observed in color, odor, and shape. The experiment was repeated 3 times.

Homogeneity Test. The ready-made shampoo is poured into a glass beaker. Observed in a bright place whether there are particles that clot or not. The experiment was repeated 3 times.

pH test. Takes 0.5 grams of shampoo preparation dissolved in 5 ml aquadestilata. Dip the pH stick in the shampoo preparation. Seen changes in color on the pH stick. Adjust the color with a predetermined pH indicator paper. The experiment was repeated 3 times [8].

Viscosity test. The RION VT-04E viscosity rotor is placed in the center of the basket containing the shampoo. Turned on so that the rotor can spin. The viscosity value is seen on the scale found on the viskometer after the needle is fixed on a certain scale. The experiment was repeated 3 times [7].

Foam stability test. 0.5 g of soursop leaves extract shampoo is mixed with 50 ml of water. Put it in a measuring cup and close it. The mixture is shaken for 20 minutes by turning the measuring cup regularly. Observe the height of the foam formed and 5 minutes later it is observed again. Calculate the height difference of foam [7].

Anti-lice effectiveness test. The shampoo formula which has the best physical properties is then tested to determine its effectiveness as an anti-lice shampoo, using 3 petri dishes. The first petri dishes was given 5 lice and 0.5 ml of soursop leaves extract shampoo; the second cup was given 5 lice and 0.5 ml of Peditox® as a positive control; the third cup was given 5 ticks and 0.5 ml distilled water as a negative control. Anti-lice effectiveness testing was carried out by looking at the number of head lice deaths for 5 minutes for 3 formulas [9].

3. Results

3.1 Soursop leaves extract (*Annonamuricata* L.) Extract

From 1 kg of soursop leaves macerated with 96% ethanol solvent produced 76.8 grams of thick extract, extract yield of 7.68% w/w. In the form of thick green-black thick extract with a distinctive odor of soursop leaves.

3.2 Physical Properties of 3 Shampoo Formula

Organoleptic test, organoleptic testing includes testing of color, odor and dosage form. Table 2 show the organoleptic test results. Homogeneity test, table 3 show that formulas I and III are homogeneous, while formula II is not. The homogeneity of the dosage indicated by the mixing of the ingredients used in the shampoo formula, both active ingredients and additives that are evenly distributed. pH test, table 4 show the result of pH test, where all 3 formula chemically acid ($\text{pH} < 7$). ANOVA test result test show that there are difference of pH among the formulas significantly, $p = 0.000$). Viscosity test, table 5 shows the result of viscosity test all of 3 formulas. The value of viscosity for shampoo preparations ranged 2dPas - 4dPas because with these thickening shampoos can spread well and convenient to use [10]. Formulas I, II, and III, have an average viscosity of 2.74 dPas, 2.5 dPas, and 4.7 dPas, respectively. ANOVA and LSD test shows that there are significance difference of viscosity among the 3 formula, $p = 0.000$. Foam stability test, table 6 shows the result of foam stability test all of the 3 formula. The Kruskal Wallis test results showed a significant difference in foam stability ($p = 0.029$), as well as the LSD test results.

Table 2 The result of organoleptic test

Formula	Colour	Odors	Consistency
I	Dark Green	Thypical of soursop	Viscous
II	Dark Green	Thypical of sourop	Viscous
III	Dark Green	Thypical of soursop	Viscous

Table 3 The result of homogeneity test

Formula	Charecteristic	Homogeneity
I	No coarse grain	Homogen
II	There ara coarse grain	Not Homogen
III	No coarse grain	Homogen

Table 4 The result of pH test

Formula	pH			X \pm SD
	Replication			
	1	2	3	
I	6	6	6	6 \pm 0
II	6	6	6	6 \pm 0
III	5	5	5	5 \pm 0

Table 5 The result of viscosity test

Formula	Viscosity (dPas)			X ± SD
	Replikasi			
	1	2	3	
I	2,7	2,7	2,7	2,7 ± 0,0
II	2,5	2,5	2,5	2,5± 0,0
III	4,7	4,7	4,7	4,7± 0,0

Table 6 The result of foam stability test

Formula	Foam height (%)
I	66,7
II	62,5
III	42,11

From the result of physical properties test, we conclude that formula I (i.e. carbopol 2 %, and citric acid 0.1 %) is the best formula, and to be continued to anti lice activity.

3.3 Anti-lice activity test

Table 7 shows that the number of lice that died in the first minute after being a soursop leave extract shampoo was 5, equal to peditox[®]. Meanwhile, in the aquadestilata group there were no lice deaths until the 5th minute.

Table 7 Anti-lice activity test

Group	Hait-lice (tail)	Minute into					
		0	1	2	3	4	5
Extract soursop shampoo	5	5	5	-	-	-	-
Peditox [®]	5	5	5	-	-	-	-
Aquadestilata	5	5	0	0	0	0	0

4. Discussion

The extraction process selected maceration because without heating so that the compound acitogenin be taken not damaged or evaporate due to take place in cold conditions acetogenin properties are compounds that are not heat resistant and at temperatures >60°C may undergo changes in structure [11].

Solvents used in the process of maceration is 96% ethanol as the ethanol itself is a solvent that is easily accessible, versatile, soluble in water and organic solvents and has a polarity that high can extract more material [12] used 96% as it can attract component which is polar and non-polar that the compounds contained in the sample can be extracted more. Leaves of the soursop (*Annona muricata* L.) contain active compounds acetogenin among others: acimisin, bulatacin, squamosin and the compounds are non-polar solvents that are used are [13].

Having obtained liquid extraction leaves of the soursop (*Annona muricata* L.) is separated by solvent by filtration using a flannel. Steam the liquid extract over the water bath to get the thick liquid extracted. The goal is that the solvent evaporation of active substances evaporate and the only remaining pure extract of the leaves of the soursop (*Annona muricata* L.). Extracts obtained viscous greenish-black with a distinctive odor extracts. Obtained extract as much as 76.8 grams and 7.68% for the results rendamen standard soursop leaves extract (*Annona muricata* L.) 11.53% [14].

From the results of physical properties tests that have been carried out the best formula is formula I. Because the physical properties test for formula I can meet the requirements of organoleptic dark green color, liquid form and the typical bubblegum odor of the extract. For homogeneity of the formula I homogeneous preparations no coarse grains, for the pH of the shampoo preparation meets the requirements of pH 6 and for viscosity of 2.7 dPas to meet the requirements of 2-4 dPas [10] and for the high foam the stability of 66.7% does not meet 40-50% but for the shampoo preparation there are no minimum and maximum requirements for foam stability [15]. Whereas in the formula II in the physical test the homogeneity of the soursop leaves extract shampoo is not homogeneous, there are coarse grains that can irritate the scalp because when developing carbopol the water is lacking. As for formula III, it does not meet the physical viscosity test requirements because the concentration of carbopol is too large so that the soursop leaves extract (*Annona muricata* L.) shampoo is too thick and difficult to pour. From formulas I, II and III with variations of carbopol and citric acid can only affect viscosity and pH. So, the effectiveness of head lice (*Pediculus humanus* Linnaeus) was tested with a comparison of negative controls using distilled water and positive controls using peditox[®].

The results of the effectiveness of the experiment using 0.5 ml of the dosage was put into a petri dishes by infestation of 5 lice in each preparation within 0 to 5 minutes of the death of the lice, wherein the formula I shampoo with 1 to 5 minutes of observation was deadly 5 head lice or 100%, treatment with negative control or distilled water with observations of 1 to 5 minutes there are also no signs of lice death then 0 lice or 0%, treat with positive control or use peditox[®] with a duration of observation of 1 to 5 minutes can be deadly 5 ticks or 100%. According to [16] in the research of Srikaya leaves extract (*Annona squamosa* L.) in the concentration of 25% extract with 10-tailed lice syrup in 5 minutes can kill 8 lice or 80% so if compared with soursop leaves extract does not differ much to kill head lice (*Pediculus humanus* Linnaeus).

Previous research by Sulis [13] showed that pure soursop leaves extract with a concentration of 25% can kill 5 lice in 1-5 minutes. This means that shampoo-making ingredients do not affect the death of head lice.

Anti-lice shampoo from soursop leaves extract can kill head lice because soursop leaves contains acetogenin, active compounds including: acimisin, bulacin, squamosin are non-polar so that the appropriate solvent is used i.e ethanol 96% [12]. The mechanism of action of acetogenin is to destroy the cell wall and precipitation of precipitation of cell proteins or microorganisms so that coagulation occurs and malfunction of these microorganisms [17].

There are some weaknes in this study, evaporation process which involves high temperatures and a long time, possible there are solvents that are still left behind or do not evaporate properly. From the pH shampoo test, a pH stick tool is used, preferably a pH meter that is used so that the results obtained are more accurate. Besides the viscosity test used a manual viskotester should use a digital viskotester so that it can distinguish and more accurate results.

5. Conclusions

Based on the explanations above, we can conclude that soursop leaves extract shampoo with 2% carbopol: 0.1% citric acid produced the best physical and was able to kill head lice in the first minute. But, there is no difference compared to Peditox® ($p > 0.05$).

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