

# ประสิทธิผลและความปลอดภัยของแชมพูสารสกัดใบยอผสมข่า เปรียบเทียบกับโลชั่นเบนซิลเบนโซเอต ในการกำจัดเหา

## Efficacy and Safety of Indian Mulberry Leaf Mixed with Galanga Extract Shampoo versus Benzyl Benzoate Lotion for the Treatment of Head Lice Infestation

นิพนธ์ต้นฉบับ

Original Article

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### บทคัดย่อ

วัตถุประสงค์: เพื่อศึกษาประสิทธิผลและความปลอดภัยของแชมพูสารสกัดใบยอและข่าเปรียบเทียบกับโลชั่นเบนซิลเบนโซเอตในการกำจัดเหา วิธีการศึกษา: ตัวอย่างเป็นเด็กนักเรียนหญิงชั้นประถมศึกษา โรงเรียนบ้านหนองมะค่า อ.โคกเจริญ จ.ลพบุรี ที่เป็นเหงาจำนวน 36 คน แบ่งเป็นกลุ่มทดลองที่ใช้แชมพูสารสกัดใบยอและข่า 18 คน และกลุ่มควบคุมที่ใช้โลชั่นเบนซิลเบนโซเอต 18 คนนาน 28 วัน ใช้แบบประเมินการตรวจนับเหา แบบประเมินความปลอดภัยในการใช้แชมพูสารสกัดใบยอและข่าและโลชั่นเบนซิลเบนโซเอต และแบบประเมินความพึงพอใจเพื่อเก็บข้อมูล สถิติที่ใช้ในการวิเคราะห์ข้อมูล ได้แก่ ค่าความถี่ ค่าเฉลี่ย ส่วนเบี่ยงเบนมาตรฐาน ทดสอบความแตกต่างระหว่างกลุ่มด้วยไควสแควร์ และที่เทสต์ ผลการศึกษา: แชมพูสารสกัดใบยอและข่ามีประสิทธิภาพในการกำจัดเหาดีกว่าและมีความปลอดภัยมากกว่าโลชั่นเบนซิลเบนโซเอตอย่างมีนัยสำคัญทางสถิติ ( $P$ -value < 0.05) ความพึงพอใจต่อการใช้แชมพูสารสกัดใบยอและข่าสูงกว่าโลชั่นเบนซิลเบนโซเอตอย่างมีนัยสำคัญ ( $P$ -value < 0.05) สรุป: แชมพูสารสกัดใบยอและข่ามีประสิทธิภาพและความปลอดภัยที่ดีกว่าในการกำจัดเหาเมื่อเปรียบเทียบกับโลชั่นเบนซิลเบนโซเอต

คำสำคัญ: เหา, แชมพู, ใบยอ, ข่า, เบนซิลเบนโซเอต

#### Editorial note

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### Abstract

**Objective:** To determine efficacy and safety of Indian mulberry leaf mixed with galanga extract shampoo versus benzyl benzoate lotion for the treatment of head lice infestation. **Method:** Thirty-six female students studying at primary school at Ban Nong Makha School, Khok Chareem district, Lopburi province were recruited and randomized into the experimental (test shampoo) and control (benzyl benzoate lotion) groups of equal size. Treatment lasted 28 days. Data were collected using the lice counting evaluation form, safety assessment form, and satisfaction assessment form. Statistical data analysis included frequency with percentages and mean with standard deviation. Comparisons between groups were done using chi-square test and independent t-test as appropriate. **Results:** Test shampoo was more efficacious and safer than benzyl benzoate lotion ( $P$ -value < 0.05) in treating head lice. Satisfaction toward the test shampoo was superior to that of benzyl benzoate lotion ( $P$ -value < 0.05). **Conclusion:** The Indian mulberry leaf mixed with galanga extract shampoo has better efficacy and safety in treating head lice compared with benzyl benzoate lotion.

**Keywords:** head lice, shampoo, Indian mulberry leaf, galanga, benzyl benzoate

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## Introduction

Pediculosis is a worldwide public health problem. Head lice infestation is an ongoing and recurring problem that leads to serious health problems if left untreated.<sup>1</sup> It is most common in children, especially in school-age children.<sup>2</sup> The highest prevalence of head lice infestation occurs in the age range of 5 - 11 years.<sup>3</sup> Typically, head lice spreads through head to head contact with an infected person and other routes are sharing of hairbrushes, hats, mats, towels, bed linen, clothes, combs, pillows, or other personal items.<sup>4</sup> Schools are at high risk for infestation of head lice. Additionally, the prevalence of head lice was 50% in females and just 3% in males.<sup>5</sup>

Head lice infestations can cause physical symptoms and lead to psychological stress. For physical symptoms, the main

symptom is itching, caused by an allergic reaction to louse saliva, which begins at about 4 to 6 weeks after the first infestation.<sup>2</sup> Other clinical manifestations of head lice include inflammation of the scalp and neck, local rash, wound, and scars as well as a chronic heavy infestation that may lead to anemia.<sup>4,6</sup> Furthermore, head lice carry certain diseases such as epidemic typhus, louse-borne typhus, louse borne relapsing fever, and louse trench fever, which can cause death without proper and timely treatment.<sup>7</sup>

Moreover, head lice can lead to other health problems. Children with head lice often have itching at night, which makes them unable to sleep. It can result in a loss of personality, an inability to concentrate on studying as well as

activities. Additionally, head lice infestations can also induce distress, social stigma, and absence from school.<sup>4</sup> Getting rid of head lice will help prevent and reduce the problem of students' personality, health, and academic performance.

Head lice treatment can be done in several ways. In Thailand, the gold standard is benzyl benzoate lotion, permethrin, and Ivermectin. All three drugs are on the A-List in the National List of Essential Medicine.<sup>5,8</sup> Other treatments are also used such as using a nit comb and cutting or shaving their hair.<sup>9</sup> The above drugs have resistance issues in many countries.<sup>10</sup> It also causes detrimental effects on children's health. Generally, 25% of benzyl benzoate concentration requires hair fermentation for 24 hours to kill head lice. Side effects are irritation and burning of the scalp and it also has a pungent smell that leads to dizziness.<sup>11</sup>

Nowadays, many medicinal plants are used in the treatment of head lice. The medicinal plants are safe, easy to find locally, and as effective in getting rid of lice as chemicals or drugs. They are available in herbal juices, extracts, and products such as shampoos, creams, gels, and lotions.<sup>12-14</sup> Medicinal plants in the Zingiberaceae or Ginger family such as turmeric, ginger, galanga, and fingerroot were used in the treatment of head lice. A study has been conducted where those plants were extracted with 95% ethyl alcohol and used to test the killing efficacy of adult head lice by the contact method. The result showed that 30% galanga extract has the best effect on killing lice.<sup>15</sup> *Morinda Citrifolia* L. or Indian mulberry, a plant in the Rubiaceae family, is one of the traditional herbs that have been used in the treatment of head lice. Squeezed juice and shampoo have been used in head lice treatment.<sup>16,17</sup>

In Khok Charoen district, Lopburi province, it was found to have an outbreak among school children with head lice infestations especially female students. From the 246 female students in this area, 186 of them had head lice infestations (75.61%).<sup>18</sup> Researchers realized the importance of the development of herbs that are very effective in terms of uses as a herbal product. Therefore, we are interested in developing herbal shampoo from extracts of Indian mulberry leaves and galanga. These plants are common in every region of the country. They are easily available and inexpensive. Thus, this study aimed to evaluate the efficacy and safety of Indian mulberry leaf mixed with galanga extract shampoo versus benzyl benzoate lotion (gold standard) for the treatment of head lice infestation in female students. The

study also aimed to compare the female students' satisfaction toward head lice infestation between the treatment of Indian mulberry leaf mixed with galanga extract shampoo and benzyl benzoate lotion. If the developed shampoo is an effective product to get rid of head lice, it could be another option to solve the problem of head lice in school children in Thailand using local medicinal plants.

## Methods

In this randomized controlled trial, study population was 113 female students of Ban Nong Makha primary school, Khok Charoen district, Lopburi province. Study sample was 36 female students who were infested with head lice recruited based on their willingness and their parent's permission. To be eligible, students had to have live head lice (adults or nymphs), not just eggs, on the hair or scalp, which was determined by visual inspection and dry-combing of hair with a head louse comb. Exclusion criteria were students affected by scalp disorders, wounds, history of irritation or sensitivity to pediculicides or hair care products from an herb, treatment with a pediculicide within 4 weeks before the study or during the study.

The eligible 36 participants were randomized into the experimental and control group of equal size. Each participant group was matched paired by the amount of head lice and then were randomly selected through lottery method. The participant was individually given a code (A or B) that was written on slips of paper to define group interventions. The experimental group used Indian mulberry leaf mixed with galanga extract shampoo and the control group used benzyl benzoate lotion for head lice treatment.

### Research interventions

Twenty-five percent concentration of benzyl benzoate lotion (Inpac Pharma<sup>®</sup>) was chosen as the chemical comparator for this study as it is the gold standard of head lice treatment in Thailand. Benzyl benzoate lotion was applied once a week, as per the manufacturers' instructions, i.e., on days 1, 8, 15, and 22. Thirty milliliters of lotion was left on the scalp covered with a protective cap for 24 hours. Thereafter, the lotion was rinsed off with a plenty of water and the hair was washed with ordinary shampoo. After the hair was dried, lice and nits were carefully combed out with head louse combs.

The Indian mulberry leaf and the galanga rhizome were washed with water and chopped into small pieces. Then the plants were macerated with 95% ethanol based on the methods from the work of Assavasirijinda (2010) with a ratio of plant sample to solvent of 1:10 for 7 days at room temperature.<sup>19</sup> Extracts were filtered and the solvent was evaporated by a rotary evaporator and dried on a boiling water bath until a constant weight was obtained. The shampoo was developed with the Indian mulberry leaf mixed with galanga extracts following the guidelines from Kaolak (2016).<sup>20</sup> The shampoo included 25% Indian mulberry leaf extract, 25% galanga extract, 95% ethyl alcohol, texapon N8000, aminon, lanolin, sodium salt, perfume, and sterile water. Indian mulberry leaf mixed with galanga extract shampoo was applied every three days<sup>21</sup>, i.e., on days 1, 4, 7, 10, 13, 16, 19, 22, 25 and 28. Thirty milliliters of shampoo was left on the scalp for 5 mins on wet hair and rinsed off with water. After the hair was dried, lice and nits were carefully combed out with head louse combs.

#### Outcome assessments

The hair was divided into 10 parts, and each part was combed 3 times with a head louse comb to keep the efficacy/bias as low as possible. Obtained head lice were placed onto white paper and examined for viability. The viability of the lice was primarily determined by their mobility. The nits were also observed. The efficacy of the treatment was judged by the disappearance of head lice and nits at days 0, 5, 10, 15, 20, 25 and 28. The cure rate was defined as the percentage of female students cured after application of the treatment. Determination of the cure rate was performed by using a head louse comb. The lice were counted and the infestation was scored as 0-none, 1-(light, or < 10 lice), 2-(moderate, or 10 – 20 lice), and 3-(severe, or > 20 lice). A patient was cured if the score was 0 (no live lice) or failed if the score was 1 or higher (one or more live lice).<sup>1</sup>

The safety assessment included the presence of dryness, redness, irritation, and flakiness. The response range from 0-absent, 1- mild, 2-moderate, and 3-severe.<sup>1</sup> These clinical signs were assessed at days 0, 5, 10, 15, 20, 25 and 28 after application of both treatments.

At the end of the study, satisfaction was evaluated by using a three-item questionnaire examining (1) the quality of the product, (2) the feature of the product, and (3) the packaging of the product of either Indian mulberry leaf mixed

with galanga extract shampoo or benzyl benzoate lotion for head lice treatment. This study used a Likert-type rating scale ranging from 1-strongly dislike, 2-dislike, 3-undecided, 4-like and 5-strongly like. Overall average score of the 3 items was used to categorize levels of satisfaction as strongly dislike, dislike, undecided, like and strongly like for 1.00 – 1.80, 1.81 – 2.60, 2.61 – 3.40, 3.41 – 4.20, and 4.21 – 5.00 points, respectively.

#### Ethical consideration

The study protocol was approved by the Human Research Ethics Committee of the King Narai Hospital, Lopburi province on September 11, 2020; with ethics number KNH 34/2563. An informed consent letter was prepared for the participants. Objectives and process of the study were explained to one of the parents or caregivers (or responsible relative). Voluntary nature of the study was informed to the students and their parents or caregiver. Participants were free to withdraw from the trial at any time. Study process was started after the informed consent form was signed.

#### Statistical data analysis

Descriptive statistics including frequency with percentage and mean with standard deviation (S.D.) were used to describe demographic characteristics and study results. Differences between the two groups were tested by Chi-square for categorical variables and independent t-test for continuous variables. Statistical significance was set at a type I error of 5% and a 95% confidence interval. All statistical analyses were performed using SPSS software version 16.0.

## Results

At day 0, most participants had severe head lice infestation in both groups (61.11% for both groups), followed by moderate level (38.89% for both groups) (Table 1). No participants were at the light and none level in both groups. At the end of the study (i.e., day 28), most participants in the experimental group were at the none (or cured) and light level of head lice infestation (83.33% and 16.67%, respectively). On the other hand, only 77.77% of participants in the control group were at the none level (or cured), followed by 16.67% at the moderate level and 5.56% at the light level. Such difference was statistically significant (*P*-value 0.038) (Table 1).

**Table 1** Number of head lice before and after treatment (N = 36).

Day of detection	Head lice level	N (%) by group			
		Experimental group (n = 18)		Control group (n = 18)	
Day 0	None	0	0.00	0	0.00
	Light	0	0.00	0	0.00
	Moderate	7	38.89	7	38.89
	Severe	11	61.11	11	61.11
Day 5	None	0	0.00	0	0.00
	Light	5	27.78	2	11.11
	Moderate	9	50.00	12	66.67
	Severe	4	22.22	4	22.22
Day 10	None	0	0.00	1	5.56
	Light	6	33.33	6	33.33
	Moderate	11	61.11	9	50.00
	Severe	1	5.56	2	11.11
Day 15	None	0	0.00	2	11.11
	Light	9	50.00	10	55.56
	Moderate	9	50.00	6	33.33
	Severe	0	0.00	0	0.00
Day 20	None	0	0.00	2	11.11
	Light	14	77.78	10	55.56
	Moderate	4	22.22	6	33.33
	Severe	0	0.00	0	0.00
Day 25	None	3	16.67	8	44.44
	Light	11	61.11	4	22.22
	Moderate	4	22.22	5	27.78
	Severe	0	0.00	1	5.56
Day 28*	None	15	83.33	14	77.77
	Light	3	16.67	1	5.56
	Moderate	0	0.00	3	16.67
	Severe	0	0.00	0	0.00

\* Comparisons of number of participants with different head lice level between the two groups at Day 28,  $\chi^2 = 6.514$ ,  $P$ -value = 0.038.

In terms of safety, 72.22% in the control group experienced mild dryness while only 5.56% in the experimental group did so (Table 2). For redness, 11.11% and 5.56% of participants in the control and experimental groups experienced mild redness, respectively. For irritation, 50.00% and 11.11% of participants in the control and experimental groups experienced mild irritation, respectively. In addition, 22.22% in the control group had moderate irritation and none in the experimental group did so. When any signs of adverse events were considered, 16.67% of participants experienced mild adverse events while 77.78% of those in the control group did so. More importantly, 22.22% in the control group experienced moderate events while none in the experimental group did so. Such difference was statistically significant ( $P$ -value < 0.001) (Table 2).

Participants in the experimental group showed satisfaction toward the Indian mulberry leaf mixed with galanga extract shampoo at "like" level or higher for each of the three aspects and overall satisfaction (Table 3). On the other hand, participants in the control group showed their satisfaction

toward the benzyl benzoate lotion at the "undecided" level for each of the three aspects and overall satisfaction. For overall satisfaction, the overall score of 4.20 points in the experimental group was significantly higher than 2.78 points in the control group ( $P$ -value = 0.002) (Table 3).

**Table 2** Safety outcomes of the participants (N = 36).

Clinical signs	Severity level	N (%) by group			
		Experimental group (n = 18)		Control group (n = 18)	
Dryness	Absent	17	94.44	5	27.78
	Mild	1	5.56	13	72.22
	Moderate	0	0.00	0	0.00
	severe	0	0.00	0	0.00
Redness	Absent	17	94.44	16	88.89
	Mild	1	5.56	2	11.11
	Moderate	0	0.00	0	0.00
	severe	0	0.00	0	0.00
Irritation	Absent	16	88.89	5	27.78
	Mild	2	11.11	9	50.00
	Moderate	0	0.00	4	22.22
	severe	0	0.00	0	0.00
Flakiness	Absent	18	100.00	18	100.00
	Mild	0	0.00	0	0.00
	Moderate	0	0.00	0	0.00
	severe	0	0.00	0	0.00
Any sign*	Absent	15	83.33	0	0.00
	Mild	3	16.67	14	77.78
	Moderate	0	0.00	4	22.22
	severe	0	0.00	0	0.00

\* Comparisons of number of participants with different level of any signs of adverse events between the two groups,  $\chi^2 = 12.600$ ,  $P$ -value < 0.001.

**Table 3** Mean scores and level of satisfaction toward the products of the two groups (N = 36).

Aspects of satisfaction	Mean, SD, and level of satisfaction by group					
	Experiment group (n=18)			Control group (n=18)		
Feature of product	4.11	1.040	Like	2.94	1.280	Undecided
Quality of product	4.59	0.567	Strongly like	2.61	1.583	Undecided
Packaging of product	3.89	0.816	Like	2.80	1.155	Undecided
<b>Overall*</b>	<b>4.20</b>	<b>0.876</b>	<b>Like</b>	<b>2.78</b>	<b>1.350</b>	<b>Undecided</b>

\* Comparisons of mean scores of overall satisfaction between the two groups,  $t$ -test = -3.61,  $P$ -value = 0.002.

## Discussions and Conclusion

In this randomized controlled trial, 83.33% female students in Ban Nong Makha School, Khok Charoen district, Lopburi province were recovered from head lice infestation using the Indian mulberry leaf mixed with galanga extract shampoo. As an standard treatment, benzyl benzoate lotion offered a 77.78% cure rate among those in the control group. This difference was statistically significant ( $P$ -value 0.038).

Galanga, a plant in Zingiberaceae, contains 1,8-cineol,  $\alpha$ -fenchyl acetate,  $\beta$ -farnesene,  $\beta$ -bisabolene,  $\alpha$ -bergamotene,

$\beta$ -pinene, and 1'-acetoxy chavicol acetate for insecticidal activity.<sup>23</sup> Moreover, galanga extracted with 95% ethyl alcohol at a concentration of 30% was the most effective in eliminating adult head lice when compared to other plants in the same family with 100% death after 8 hours of exposure.<sup>16</sup> For Indian mulberry, it has been reported that the active substance in leaves has activity against helminths and insects.<sup>24</sup> There was a study on the effectiveness of Indian mulberry leaf shampoo on head lice in grade 2 to grade 5 students at Matchipoom School, Trang province.<sup>18</sup> The research found that Indian mulberry leaf shampoo was effective at the removal of head lice and eggs at a significance level of 0.05. The female students who used the shampoo two times per week for a period of three weeks showed a 32.3% decrease in head lice infestation.<sup>18</sup>

This research found the side effects of the Indian mulberry leaf mixed with galanga extract shampoo were less than the use of benzyl benzoate lotion, which is a chemical drug. Among participants using benzyl benzoate lotion, 100% of them had clinical symptoms ranging from mild to moderate allergic reactions. After using the test shampoo, only 16.67% of participants clinical signs in light allergic reactions. The benzyl benzoate lotion has to be left on the scalp for up to 24 hours to be effective in causing lice death.<sup>16</sup> In addition, many side effects and pungent smell that lead to dizziness have occurred.<sup>12</sup> Children could not keep the benzyl benzoate lotion in their hair for the duration of the drug's requirement thus resulting in failed treatment.

New products should be evaluated for safety and effectiveness.<sup>6</sup> The results of this study indicated that the Indian mulberry leaf mixed with galanga extracts shampoo was effective and safe. This shampoo could be an alternative to treat head lice. Additionally, the satisfaction of female students is a reflection of shampoo quality. It stated that the female students were satisfied with the quality of Indian mulberry leaf mixed with galanga extract shampoo for head lice treatment. However, the feature and packaging of the product affected the female student's satisfaction. Therefore, more pleasant feature and packaging are crucial for customer satisfaction.

This study had certain limitations. This study used a small sample size and tested the product with very local target users. Power of analysis could be limited and generalization should be caution. Future research with a larger and more diverse group of participants is obligatory. The results of this

study are used as the basis for the development of herbal products for use in daily life. The shampoo may be an alternative for head lice treatment, especially in remote areas. Moreover, such product could improve the value of the medicinal plants readily available in the country.

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