



## Activity and safety test combination of garlic extract (*Alium sativum*l) and candlenut extract (*Aleurites molucanna*) as fertilizer rabbit fur

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### ABSTRACT

Efficacy of candlenut oil and garlic oil with three concentrations of 1000 ppm, 500 ppm and 100 ppm tested its activity as a fertilizer of rabbit fur by applying hazelnut oil and garlic oil each of approximately 1 ml at spot prepared twice daily for 28 days. The result of the third activity test of this concentration tested the data with statistics and got the most optimum result as a grower of rabbit fur is candlenuts oil with the concentration of 1000 ppm with the average growth of length for 28 days is 0,6250 cm and garlic oil with concentration 500 ppm with the average length of growth for 28 days is 0.8417 cm, while for the most optimum rabbit fur weights are with garlic concentration of 1000 ppm with an average weight of 28 days is 0.3627 g, while the growth of rabbit fur weight most optimum for 28 days is candlenut 1000 ppm with an average weight for 28 days = 0.3518 g result of significant analysis at ( $p \leq 0,05$ ).

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### INTRODUCTION

Garlic is a perennial herbal plant that forms a tuber. This plant grows in clumps and stands upright to a height of 30-75 cm. The stem that appears above the ground is a pseudo-stem consisting of leaf midribs. While the real stem is in the ground. From the base of the stem grows many small, fibrous roots less than 10 cm in length. The roots that grow on the main stem are rudimentary, functioning as a means of sucking food (Carruthers et al., 2016).

Garlic is the name of the plant from the genus *Allium*,

as well as the name of the tuber produced. Garlic is capable of producing essential oils, one of which can be obtained from the refining process. In the wider community, garlic essential oil is known to contain several components that can be used for the health world, especially in the cosmetic field (Censi et al., 2018).

Like most other plants, garlic contains more than 100 secondary metabolites, which are very useful biologically. Among the several bioactive components found in garlic, sulfide compounds are the ones that are abundant. These compounds include diallyl sulfide or in an oxidized form called Allysin. Just like other phenolic compounds, Allysin is thought to have a very broad physiological function, including antioxidants, anticancer, anti thrombotic, anti-inflammatory, lowering blood pressure, and can lower blood cholesterol (Fuji and Root, 2016).

Epidemiological data also shows that there is a correlation between garlic consumption and a decrease in cardiovascular diseases, such as atherosclerosis (fat accumulation), coronary heart disease, and hypertension. These compounds contain mostly sul-

fur which is responsible for the taste, aroma, and pharmacological properties of garlic. The two most important organosulfur compounds in garlic tubers, namely the non-volatile amino acid  $\alpha$ -glutamyl-S-alk (en) il-L-cysteine (1) and the essential oil S-alk (en) ilsisteinsulfoxide or alliin (2). The two compounds above are the precursors of most of the other organosulfur compounds (Muhammad and Abu, 2014).

Levels can reach 82% of all organosulfur compounds in tubers which can function as anti-implantation. The compound glutamyl-S-alk (en) il-L-cysteine (1) is an intermediate compound for the formation of other organosulfur compounds, including alliin (2). This compound is formed from the acid biosynthetic pathway. The reaction process of  $\alpha$ -glutamyl-S-alk breakdown (en) il-L-cysteine (1) takes place with the help of the enzymes  $\alpha$ -glutamyl - transpeptidase and  $\alpha$ -glutamyl-peptidaseoxidase, and will produce alliin (Ridland and Halloran, 1981).

In Indonesia, hazelnut, which is extracted into hazelnut oil, has long been trusted by most of the Indonesian population as a fertilizer and reduces hair loss. The core of hazelnut seeds contains 60-66% of hazelnut fatty acids. The oil content in hazelnut seeds is high, namely 55 - 66% of the weight of the seeds. The main component of hazelnut oil is unsaturated fatty acids, but it also contains a relatively small percentage of saturated fatty acids. The hazelnut oil contained in the seeds also has many benefits, including ingredients for cosmetics, medicines, varnishes, paints, and fuels (Director General of Drug and Food Control, 1989) (Table 2).

Hazelnut oil functions as hair fertilizer; besides that, it can also be used as a hair growth stimulant or as an additive in hair care. Currently, high-quality hazelnut oil has become the main commercial product and is widely sold in the cosmetics industry. Candlenut functions to treat hair loss by strengthening the hair root mechanism, which is usually produced and packaged in hair oil preparations. Besides that, pecan also contains vitamin B of 0.06 mg in 100 g of pecan, which plays a role in strengthening the roots. HairBecause the structure of the eyebrows is the same as the structure of the hair, and it should be assumed that hazelnut oil is able to function as a growth or even as a fertilizer for eyebrows (Director-General of Drug and Food Control, 2016) (Figure 2).

Candlenut oil is a type of oil that is easy to obtain and has often been used empirically in everyday life for various properties. The relationship as hair fertilizer from these plants has been shown by various studies. Based on the research in cosmeti-

cologi, the combination of hazelnut essential oil and mangkokan leaf extract has better hair growth activity than single-dosage extracts. The difference with this research is that pecan essential oil is combined with garlic essential oil, which functions as an eyebrow grower and maintains the condition of human eyebrows aged over 20 years to 50 years (Figure 3).

## METHOD

### Tools and Materials

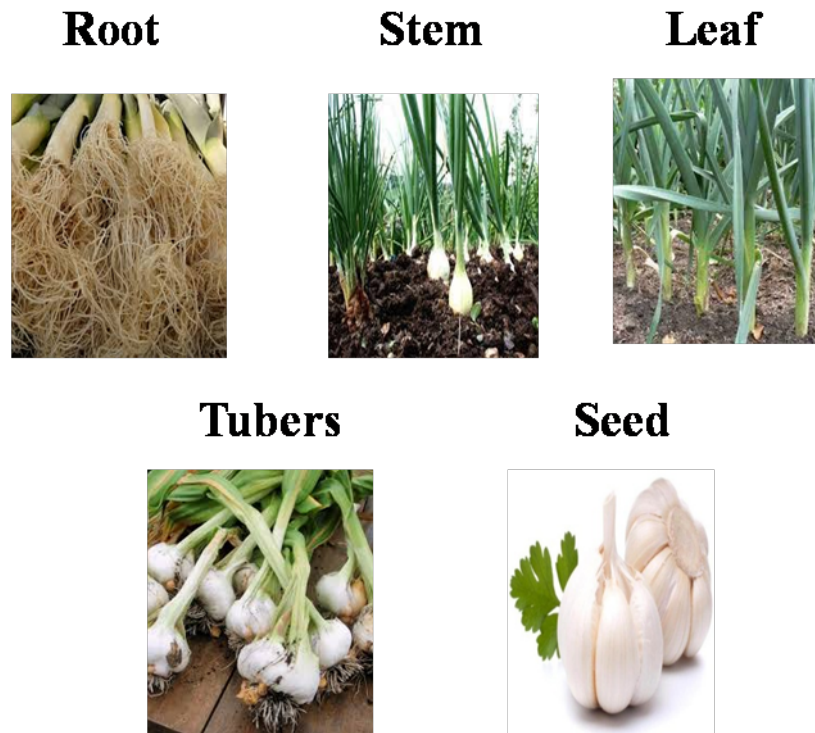
The ingredients used in this study were: Garlic Oil which has been tested for its simplicia quality and oil quality at (Anafarma Phytochemical Laboratory), Candlenut Essential Oil which has been tested for simplicia quality and oil quality at (Anafarma Phytochemical Laboratory), 70% Ethanol (Merck), Aquadest (AnafarmaLaboratorium), Eyebrow Pencil brand X with Batch number \_ Exp., DMSO as a solvent (Roy et al., 2008).

The research used instruments in the form of glassware with the Pyrex brand, steam distillation by Germany, Heating coat with the Bardead Electrothermal brand, Sartorius branded Analytical Scales, Pyrex Picnometer, Oswald Viscosimeter with Scopus brand, Oven with Memmerth brand, pH meter with Methorm brand, Centrifuge with Methorm brand, Evaporator Rotary with Eyela Brand, Agilent Gas Chromatography, Scanning Electron Microscope with Brand, Architectural Slide with Mutoh Brand.1.1 Work Procedures

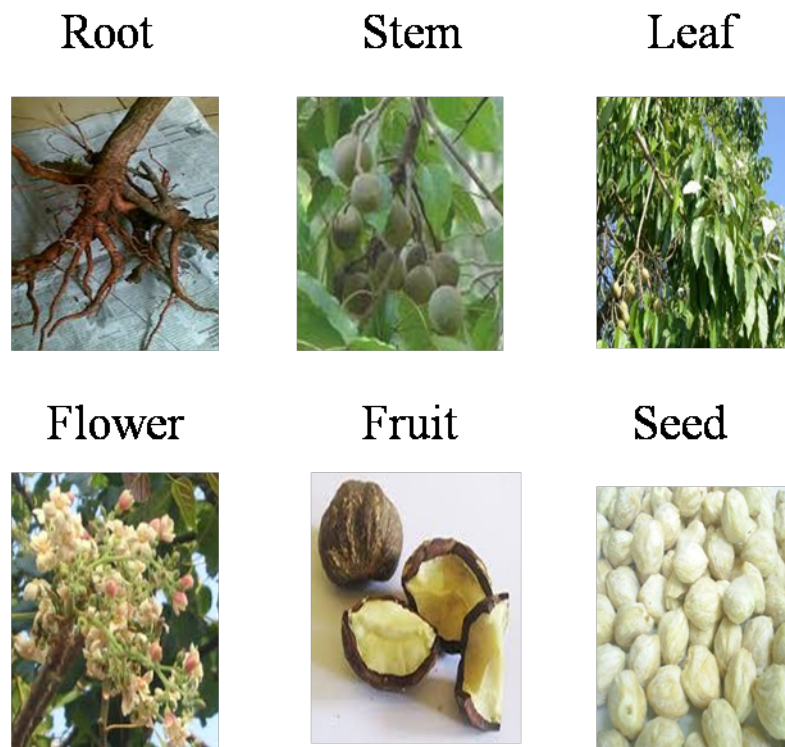
### Activity Test on Animals

The test animals used were male white rabbits aged 3-6 months with a body weight of 2-3 kg with; the number of rabbits used was determined using Federer's empirical formula:  $(n-1)(t-1) \geq 15$ , t indicates the number of treatments and n indicates the number of animals per treatment based on Federer's empirical formula, the number of animals used is 4 male white rabbits. On the back of each rabbit, 9 basting areas were made, namely combination 1, combination 2, combination 3, normal control, negative control and positive control. The combination that gives the most optimum results is measured in grams and cm in weight and length (Director-General of Drug and Food Control, 2016).

In this study, the eyebrow fertilizer activity test was carried out in a combination of hazelnut oil and garlic oil in three (3) combinations, namely combination 1 (200 ppm hazelnut oil and 200 ppm garlic oil), 2 combinations (100 ppm hazelnut oil and 200 ppm garlic oil). Ppm) and a combination of 3 (50 ppm hazelnut oil and 200 ppm garlic oil). Previously conducted activity tests of single extract of 1000 ppm,



**Figure 1: Garlic Plant**



**Figure 2: Candlenuts Plant**

### Candlenuts Seed ( 10x)

### Garlic Seed ( 10x)

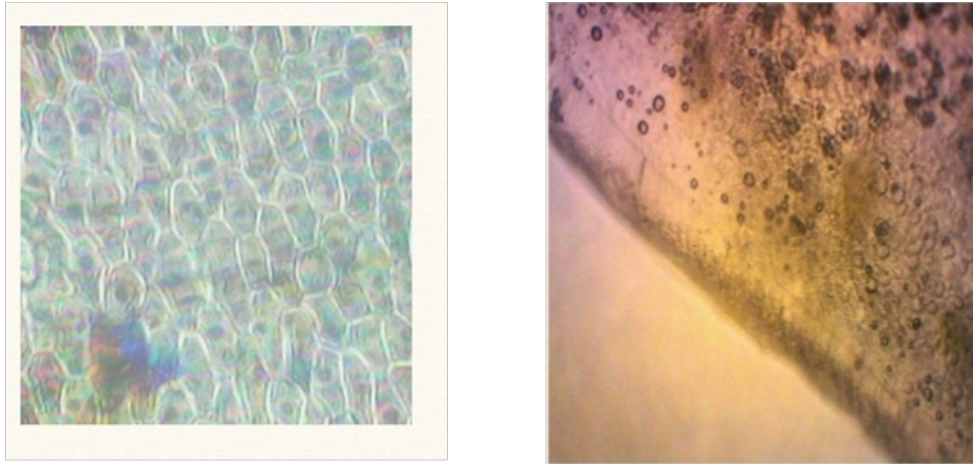


Figure 3: Cross Section of Candlenut and Garlic

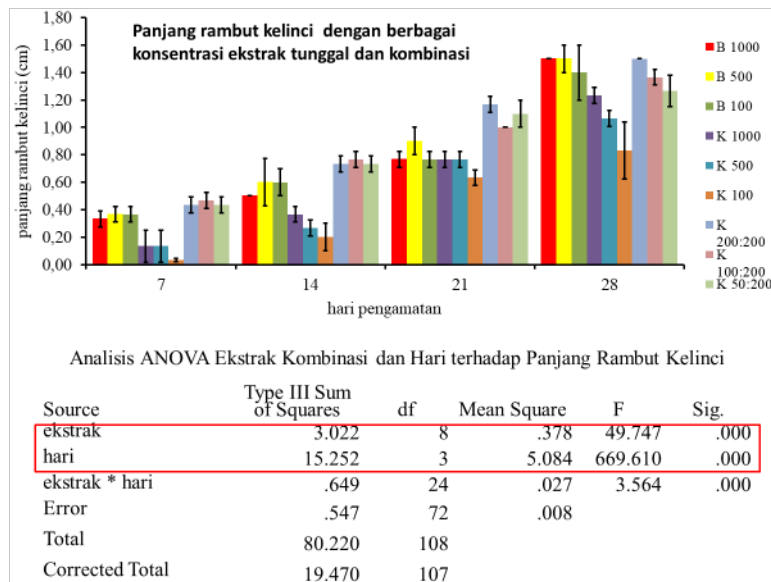
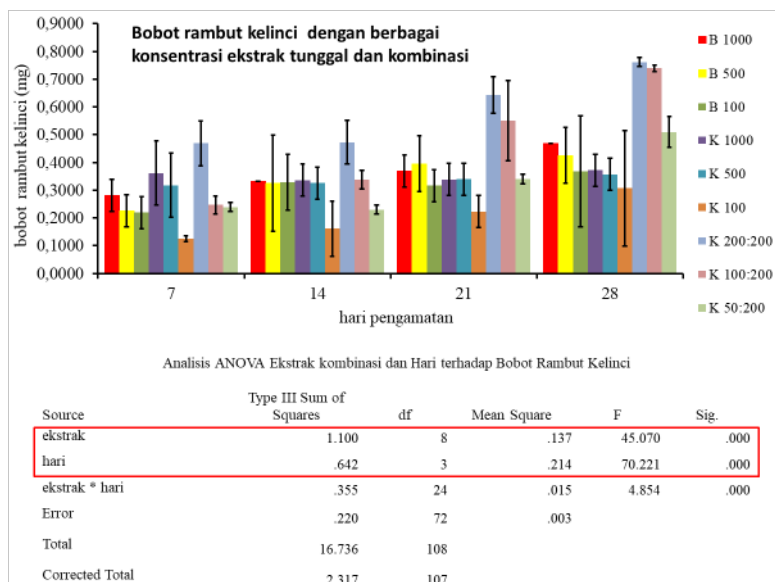


Figure 4: ANOVA analysis of rabbit hair length from various concentrations of single and combination extracts

Table 1: Garlic Plant Morphology

No.	Specification	Information
1.	Colour	Green Leaves and White Petiole Stolons
2.	Surface	Average
3.	Average Size	2 sd 6 cm
4.	Leaf Tip	Pointed
5.	Leaf Base	Thick White
6.	Leaf Bone Arrangement	Single
7.	Number of Leaves	One Tuber Six to Seven Strands
8.	Shape	Whistled to Form a Round





**Figure 5: ANOVA analysis of rabbit hair weights from various concentrations of single and combination extracts**

**Table 2: Candlenuts Morphology**

No.	Specification	Informations
1.	Colour	The Seeds are Yellowish, Wrapped in a Hard Shell that is Dark Brown.
2.	Space	Jagged
3.	Size	Seeds Vary, with a Shell Thickness of 0.5-1 mm, a Seed Diameter of 2-3 cm
4.	Form	Round

**Table 3: Identity Candlenuts end Garlic Oils**

No.	Description	Candlenuts Oil	Garlic Oil
1.	Form	Thick	Thick
2.	Colour	Yellow	Yellow Light Brown
3.	Smell	Specific	Specific
4.	Taste	Bargain	Speisy

**Table 4: Total Ash Content**

No.	Treatment	Total Ash Content Candlenuts Oil	Total Ash Content Garlic Oil (%)
1.	Simplisia	5,79	11,80
2.	Ekstract	2,26	0,74

**Table 5: Water Content Candlenuts Oil end Garlic Oil**

No.	Name of Sample	Content water I (%)	Content water II (%)	Content water III (%)	Average (%)
1.	Candlenuts Oil	4,77	4,58	4,80	4,72
2.	Garlic Oil	1,47	1,42	1,44	1,44

**Table 6: Residual Solvent of candlenuts end garlic Oils**

No.	Sample	Residual Solvent (%)	Information
1.	Candlenuts oil	0,000005	
2.	Garlic oil	0,000005	

**Table 7: pH Candlenuts and Garlic Oils**

No.	Sample	pH	Information
1.	Candlenuts Oil	7	Lampiran
2.	Garlic Oil	6,45	

**Table 8: The viscosity of Candlenut Oil and Garlic Oil**

No.	Sample	Viscosity	Information
1.	Candlenut Oil	7677 cP	Achives
2.	Garlic Oil	7567 cP	

**Table 9: Phytochemical Screening of Garlic Oil**

Phytochemical Screening	Test	The Result	Information
Alkaloid	Meyer Dragendorf Bouchardat	Positive	A turbid solution is formed Formed in orange color A Chocolate Sediment Forms
Saponin	base	Positive	Forms a foam that does not disappear
Tanin	Dischange with FeCl <sub>3</sub>	Negative	No blackish-blue or blackish green color is formed
Flavonoid	Reagen alkali Dischange Mg	Negative	No intense yellow color is formed No color on the amyl alcohol coating
Terpenoid	Salkawski	Positive	Reddish-brown in interphase coating
Steroid	Salkawski	Positive	Red color on the bottom layer

**Table 10: Phytochemical Screening of Candlenuts Oil**

Phytochemical Screening	Test	The Result	Informations
Alkaloid	Meyer Dragendorf Bouchardat	Positive	A turbid solution is formed Formed in orange color A Chocolate Sediment Forms
Saponin	base	Positive	Forms a foam that does not disappear
Tanin	Dischange FeCl <sub>3</sub>	Negative	No blackish-blue or blackish green color is formed
Flavonoid	alkali Dischange Mg	Negative	No intense yellow color is formed No color on the amyl alcohol coating
Terpenoid	Salkawski	Positive	Reddish-brown in interphase coating
Steroid	Salkawski	Positive	Red color on the bottom layer

500 ppm and 100 ppm of hazelnut oil, 1000 ppm of garlic oil, 500 ppm and 100 ppm.

Every single extract of pecan, garlic oil and a combination of hazelnut oil and garlic oil were tested for their activity on rabbits in-vitro. The in-vitro test was performed by shaving the back hair of the rabbit with nine spots. The first spot is smeared with 1000 ppm of hazelnut oil, the second spot is smeared with 500 ppm of hazelnut oil, the third spot is smeared with 100 ppm of hazelnut oil, the fourth spot is smeared with 1000 ppm of garlic oil, the 5th spot is smeared with 500 ppm of garlic oil, the 6th spot is smeared with oil 100 ppm of garlic, 7th spot is applied with control + (using gastrol oil), 7th spot is applied with an eyebrow pencil, 8th spot is applied with control - (using distilled water) and 9th spot is not smeared with any solution as a normal condition) More rubbing less 1 ml of the solution twice a day for 28 days (Edisi III, 2009).

The activity of the combination of hazelnut oil and garlic oil was tested after knowing the most optimum activity of the single extract. The combination of hazelnut oil with garlic is made in 3 combinations. Namely, combination A is a combination of 200 ppm hazelnut oil with 200 ppm garlic oil, combination B is a combination of 100 ppm hazelnut oil with 200 ppm garlic oil, combination C is a combination of 50 ppm hazelnut oil with 200 ppm of garlic oil which gives the most optimum results which will be continued to the brow growth formulation stage (brow growth.).

This study did not conduct an irritation test because, based on the "Journal of Botanical of Herbal Cosmetics in the Asia Pacific", it was explained that for cosmetics derived from herbs, there is no need for an irritation test because the oil comes from plants that are consumed by humans.

This test has received Ethical Clearance from the Health Research Ethics Commission of Jakarta Health Polytechnic II.

## RESULT AND DISCUSSION

### Plant Determination

Plant determination at LIPI Cibinong, Biology Laboratory, Jalan Raya Bogor KM.46 Bogor, West Java. Plant determination is carried out to find out the correct identity of the plants used in the research as intended. From the results of the determination that has been done, the results indicate that the plant is a plant of Garlic (*Allium sativum* L) familia. And the pecan plant (*Aleurites moluccana*) (Figure 3).

Morphological data on garlic tubers showed that the

tubers were round and tubers like tubers. The size varies yellowish white.15 (Table 1, Figure 1)

The resulting candlenuts oil and garlic have the following descriptions.

### Ash content

$$\% \text{ Ash Content} = \frac{(\text{krus} + \text{ash}) - (\text{krus blank}) \times 100\%}{\text{Ekstrakt}}$$

The principle of determining the ash content is that a number of materials are heated to a temperature where the organic compounds and their derivatives are digested and evaporated, leaving only mineral and inorganic elements left (Table 3).

The results of the determination of the total ash content provide an overview of the internal and external mineral content of both organic and inorganic compounds from the initial process to the formation of simplicia and extracts either naturally derived from plants or contaminants during the process (Table 4) (Ridland and Halloran, 1981).

### Water Content

The water content required in the Herbal Pharmacopeia (2008) for hazelnut oil is not more than 10%, and garlic is not more than 12% (Table 5).

### Residual Solvent

The remaining solvent level needs to be determined because it will affect the oil's activity. The remaining solvent for hazelnut oil was 0.000005 and the remaining solvent for garlic oil was 0.000005, which was determined by Gas Chromatography (Table 6) (Ridland and Halloran, 1981).

### pH of Candlenut Oil and Garlic Oil

From the test results, the pH of the hazelnut oil was 7 (Neutral), while the pH of the garlic extract was 6.45 (slightly acidic). The pH of the oil needs to be determined because the pH of the preparation must match the pH of the skin, which is neutral because if the pH is acidic, it will irritate the skin, and if the pH is alkaline, it will cause the skin to become dry (Table 7) (Takeo, 2016).

### The viscosity of Candlenut Oil and Garlic Oil

Viscosity needs to be determined so that the resulting preparation can stick evenly to the Table 8.

### Phytochemical Screening

The phytochemical testing of garlic extract and hazelnut extract was carried out by six methods, as shown in Tables 9 and 10 below.

### Activity Test on Animals

Eye brow growing activity is an activity that occurs on a rabbit's back hair, which is applied a single extract of 1 ml of each spot which is applied twice a

day for 28 days. The results are measured by weighing the weight and measuring the length (Edisi III, 2009).

The rabbit's back was made a spot measuring 0.5 cm x 1 cm by shaving. Nine spots are made. 1st spot smeared with 1000 ppm hazelnut oil, 2nd spot with 500 ppm hazelnut oil, 3rd spot with 100 ppm hazelnut oil, 4th spot with 1000 ppm garlic oil, 5th spot with onion oil putih 500 ppm, 6th spot with garlic oil with 100 ppm, 7th spot with the positive control, 8th spot with positive control and 9th spot with normal. Replication rabbits, there are 3 tails. The results of statistical data processing can be explained in Figures 4 and 5 above (Roy et al., 2008). There was no significant difference in the length of rabbit fur at different concentrations of single garlic oil, while the hazelnut oil gave a significant difference to the length of rabbit fur with different concentrations. The combination concentration of garlic oil with hazelnut oil gave significant rabbit hair length at a concentration of 200: 200 ( $p \leq 0.05$ ) (Figure 4) (Edisi III, 2009).

There was a significant difference between the weight of rabbit hair and the ratio of various concentrations of the combination of garlic oil and hazelnut oil at a significant level ( $p \leq 0.05$ ). (Figure 5) (Nguyen, 2014).

## CONCLUSION

Since the combination of garlic oil with hazelnut oil gives significant results as fertilizer for rabbit fur, it is reasonable to suspect that the combination of hazelnut oil with garlic oil can also have an effect as a fertilizer for human eyebrows.

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The authors declare that they have no funding support for this study.

## Conflict of Interest

The authors declares that there is no conflict of interest for this study.

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