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Formulation and evaluation of lip balm preparations of ethanolic extract from beetroot (Beta vulgaris L.)

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Abstract

Beetroot (Beta vulgaris L.) is a source of betacyanin that could be used as a natural dye and is known as an antioxidant that can protect the skin from exposure to free radicals. Lip balm, one of the popular cosmetics, often uses natural ingredients as active compounds to enhance colour and promote healthy skin. This study aims to formulate beetroot ethanolic extract into a topical lip balm preparation. The beetroot was extracted using ethanol solvent. The phytochemical screening was carried out to identify phytochemicals of flavonoids, saponins, tannins, and alkaloids within the ethanolic extract. The extract was formulated into a lip balm preparation with extract concentrations of 0% (F1), 5% (F2), 10% (F3), and 15% (F4). After formulation, the lip balm was physically examined for organoleptic, homogeneity, acidity, spreadability, and adhesivity of the product. The phytochemical screening exhibited positive results in all phytochemical groups tested in the beetroot ethanolic extract. Meanwhile, physical evaluation of lip balm demonstrates that all formulations had a semi-solid texture, cacao scent, and red color that increased in intensity proportionate with the extracts used (F2, F3, F4). Other parameters indicate a homogeneous product with suitable acidity (pH valued at 9) and adhesivity (>4 seconds). However, the spreadability test displayed low dispersion capabilities of the lip balm. Even though the spreadability values increased with higher beetroot extract, this value remained lower than the standard minimum dispersion of lip balm, thus indicating poor spreadability of the formula. The hedonic evaluation indicates that respondents tended to like the third formulation (F3). Based on this, the best and most preferred formula is at a concentration of 10% active substance (F3).

Keywords: Lip Balm; Formulation; Beetroot; Betacyanin

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INTRODUCTION

Beetroot (Beta vulgaris L.) is included in the type of tubers widely cultivated in Indonesia. The utilization and processing of beetroot in Indonesia, however, are still rare and poorly known by most people. Poor taste, a strong earthy smell when consuming, and too striking a color can be reasons causing the lack of liking of beetroot.[1] Beetroot is, in fact, a plant

useful as a natural dye and a good source of minerals and vitamins, including B vitamins, calcium, phosphorus, and iron. Furthermore, beetroot contains red pigments, namely betacyanin.[1,2] Betacyanin is also widely used because of its usefulness in addition to being a natural dye as well as an antioxidant. This pigment produces a purplish red color that can be used as a natural dye.[3–5] The properties of betacyanin made this pigment suitable for color ingredients in the formulation of cosmetic products like lip balm.

Lip balm is a popular cosmetic mainly used to prevent dryness of the lips by increasing moisture and enhancing the natural color of the lips. Lip balm uses bases like oil, wax, or fat mixed with active ingredients, pigments or coloring agents, and preservatives.[6] The lipophilic bases principally cover the skin layer of the lips, trapping moisture in the skin and guarding against lip damage.[7,8] The coloring agent of lip balm commonly uses pigments and other natural coloring compounds. One of the plants that has the potential to be developed as a lip balm is the beetroot tuber plant. In addition, betacyanin has two advantages: as a pigment constituent that boosts red-based color and a protective antioxidant against free radicals.[2,9,10] Therefore, this study aims to develop a lip balm that uses beetroot as a source of red color. The formulation of lip balm from natural ingredients was done by balancing the concentration of the main ingredient with other additives. Physical evaluation and hedonistic of lip balm used in this study were to assess both the customer's favorability and the product's physical properties.

METHODS

Tools and materials

The tools utilized included Beaker Glass (Pyrex), Erlenmeyer Glass (Pyrex), Measuring Glass (Pyrex), Micropipette (Ohauss), Digital analytic (Ohauss), Powder machine, Buchner funnel, Stirrer magnetic (Ohauss), Water bath (WT-6H), Ph Meter(Hanna), Oven (Memmert), mortar, stamper, and others common tools to mix ingredients.

The ingredients used in this study were beetroot (Beta Vulgaris L.), Ethanol p.a (Merck), HCL (Merck), $FeCl_3$ (Merck), Wagner reagent (Merck), cera flava, lanolin, glycerin (Bratachem), nipagin (Bratachem), liquid paraffin (Bratachem), and oleum cacao (Bratachem).

Procedures

The Maceration of Beetroot

The beetroots were cleaned under running water. After that, they were thinly chopped, dried in the sunlight with a covered black cloth, and then powdered using a powder machine. The process of making extracts was carried out by maceration, by simply soaking 300 grams of beetroot powder with 1500 ml of ethanol (1:4), with the addition of 1% ascorbic acid (pH solution becomes 4.5), added to prevent degradation of betacyanin. The maceration was conducted for 3 x 24 hours. The macerate was filtered and then evaporated using a rotary evaporator at a temperature of 40°C until a thick extract was obtained.[3,11]

Phytochemical Screening

The phytochemical screening consisted of qualitative tests of flavonoids, saponins, tannins, and alkaloids.

Flavonoids

The liquid ethanolic extract of beetroot was heated for approximately 2 minutes, and then 4-5 drops of concentrated 2N HCl were added. The yellow to dark red mixture indicated positive results within 3 minutes.[12]

Saponins

The ethanolic beetroot extract was added with warm distilled water into a test tube shaken vertically for 10 seconds. The results were positive for forming stable foam for 10 minutes minimum 1 cm.[13]

Tannins

The ethanolic beetroot extract was reconstituted, and 3 drops of 1% $FeCl_3$ solution were added. The blackish green color of the mixture indicated the presence of tannin.[13]

Alkaloids

Ethanolic extract of beetroot, which had been reconstituted, was put in a test tube, added 1mL HCl 2N, heated on a water bath for 2 minutes, desired, did filtering, and then added 2-5 drops of Wagner reagent. The positive results were obtained from the formation of brown precipitates [12]

Lip Balm Formulation

The lip balm preparation material used oleum cacao, cera flava, and lanolin as a lipophilic base. These three compounds were heated until they became liquid and mixed until homogeneous. After that, nipagin and glycerin were added to the mixture and stirred until evenly mixed. The beetroot extract was added constantly until a semi-solid lip balm mixture was formed. The formulation of lip balm can be seen in Table 1.

Table 1. Beetroot Extract Lip Balm Dosage Formulation

Material	Formulation (% b/b)				Material usability
	I	II	III	IV	
Beetroot extract	0	10	15	20	Active substances
Cera flava	5	5	5	5	Base
Lanolin	50	50	50	50	Base
Glycerin	2.5	2.5	2.5	2.5	Humectants
Nipagin	0.18	0.18	0.18	0.18	Preservatives
Liquid Paraffin	15	15	15	15	Emollient
Oleum cacao	Add 100	Add 100	Add 100	Add 100	Base

Physical Evaluation and Hedonic Test of Lip Balm

Physical evaluation of lip balm includes homogeneity, acidity, spreadability, and adhesivity tests.

Organoleptic test:

Organoleptic tests determined the lip balm's color, texture, and scent.

Homogeneity test:

A small amount of lip balm was sampled on a suitable or transparent glass object. After that, the observation was conducted using a different background of black and white. The homogeneity of the sample indicated by the absence of coarse-grained or clumped ingredients.[15]

Acidity test:

A small amount of lip balm taken from the upper, middle and bottom layers of lip balm was measured using pH indicators.[15]

Spreadability test:

A 0.5-gram sample was placed on object glass and given pressure ranging from 50, 100, and 150 grams. A dispersion value of the sample was measured on the diameter of the sample after 1 minute.[15]

Adhesivity test:

The adhesion test was carried out by weighing 0.5 grams of the sample on the glass object and attaching it to the glass, giving a load of 500 grams for 5 minutes. Measurement of adhesion properties of the sample used the time of glass release.[15,16]

Hedonistic test:

The hedonic test was performed on a group of twenty panellists (panellists had already agreed on informed consent) who were chosen based on random gender and different skin colours. After that, the most preferred formula based on texture, colour, and scent was chosen.[17]

RESULT AND DISCUSSION

Maceration of Beetroot

A fresh beetroot was ground into powder, dried and then macerated using ethanol solvent. A total of 7 kg of beetroot was collected from the traditional market in Cirebon. After drying and powder processing, this amount was reduced to only 5% of the initial weight in powdered dry beetroot. Some literature describes beetroot as having more than 87% water content.[1,18,19] The sorting and peeling process of beetroot also contributes to further reducing the dry powder of beetroot.

The maceration of beetroot was modified from the common maceration process, adding an acidic condition to prevent degradation of betacyanin. The uses of ascorbic acid in a previous study showed stabilisation of betacyanin and acted as an antioxidant to further prevent betacyanin oxidation.[20] The maceration process yielded 59 grams of ethanolic extract (16,8 g from the dry powder of beetroot). The step of fresh beetroot processed to maceration of beetroot can be observed in Figure 1.

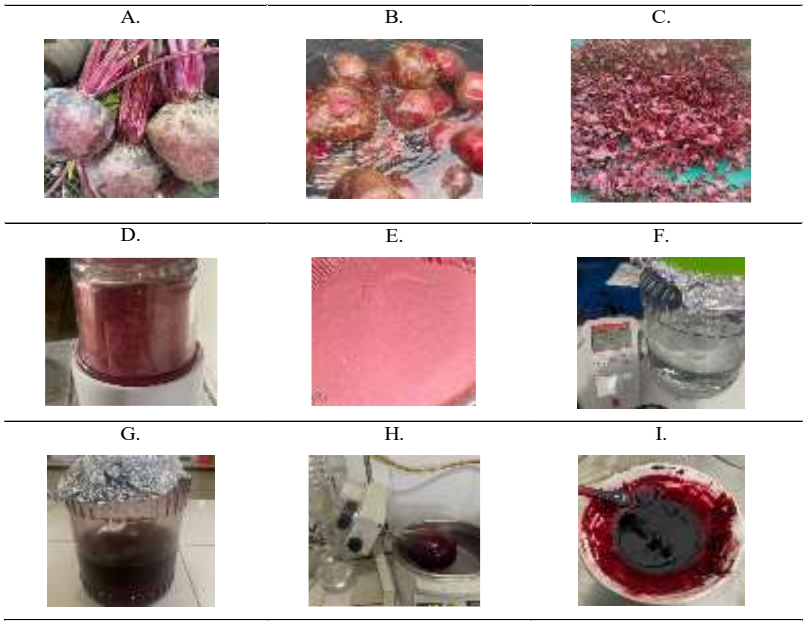

Figure 1. Step-by-step for beetroot extraction. A). Fresh beetroot was collected from a traditional market; B). Washing and cleaning process; C). Drying process; D) Powdered process of beetroot; E). Dry powder of beetroot; F). Adjustment of acidity using ascorbic acid; G). Maceration process; H). Evaporation of macerate; I) Extract of beetroot

Phytochemical screening of beetroot

The phytochemical screening was conducted to identify some bioactive compounds in the extract. The phytochemical groups tested include saponin, flavonoid, alkaloid, and tannin. The processing of beetroots with each step and condition assumed could reduce the bioactive content of beetroots. Based on these assumptions, the screening proved that this process could retain bioactive compounds. The results exhibited positive values for all tested groups of phytochemicals. This indicated the maceration process did not degrade the bioactive content of beetroot. The phytochemical screening can be seen in Figure 2.

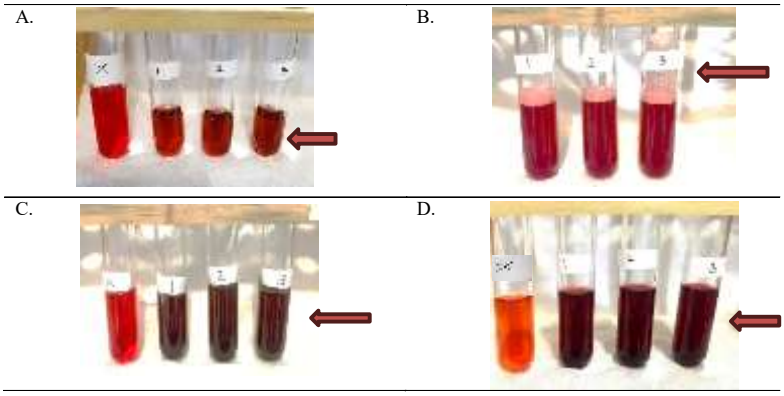

Figure 2. Bioactive compounds were identified in triplication (The number indicated the sample used). A). Flavonoids test; (B). Saponins test; (C) Tannins test; (D) Alkaloids test.

Formulation of lip balm and evaluation of the product

The formulation was designed with 4 formulas (F1, F2, F3, and F4) with different concentrations of beetroot extract. The F1 was used as a base formula without extract, and others were given beetroot extract, respectively, with concentrations of 5% (F2), 10% (F3) and 15% (F4). The final product of each formula can be seen in Figure 3.

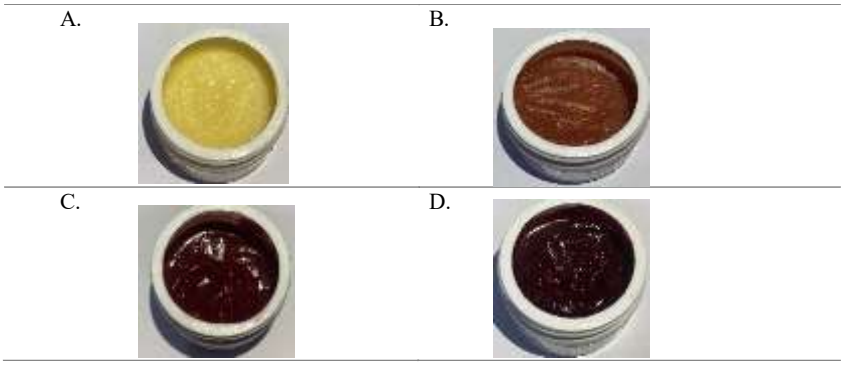

Figure 3. The lip balm was produced from formulas of F1 (A); F2 (B); F3(C); F4 (D)

Each formula has distinctive features based on texture, color and scent. The organoleptic test of each formula indicated a semi-solid texture and a strong scent of cacao due to no essence added in the formula. The beetroot contains sesquiterpene geosmin that gives an earthly beet-like scent, which in this formula is covered by the strong smell of cacao (21). The formula's color ranged from pale red to dark red, proportionate to the increase of beetroot extract. The organoleptic test and other physical evaluations can be seen in Table 2, while the hedonic evaluation is illustrated in Figure 4.

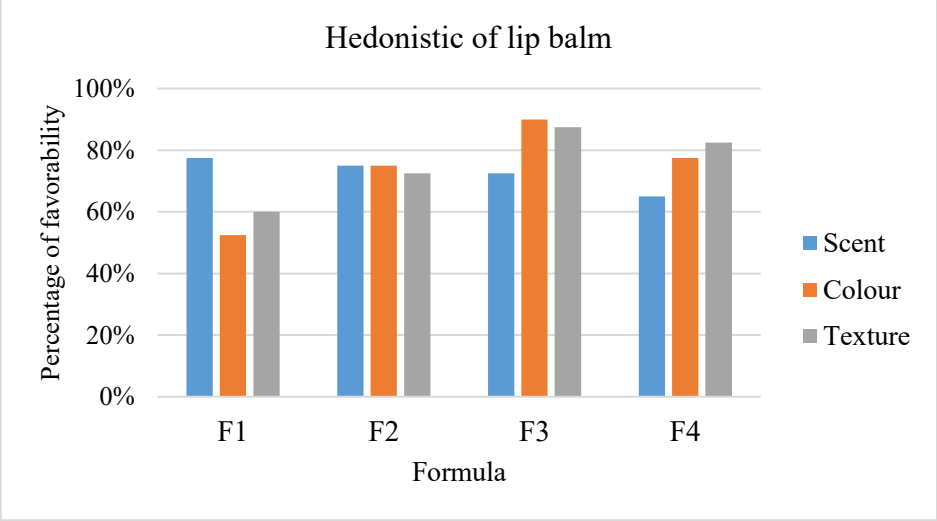

Figure 4. Hedonistic test to determine the favorability of the respondent based on scent, color, and texture

Table 2. Physical evaluation of formulas (F1, F2, F3, and F4)

Physical evaluation	Formula			
	F1(0%)	F2(5%)	F3(10%)	F4 (15%)
Organoleptic : - Visual/color - Scent - Texture *	Yellow Cacao scent Semi solid	Red Cacao scent Semi solid	Dark Red Cacao scent Semi solid	Dark Red Cacao scent Semi solid
Homogeneity	Homogeneous	Homogeneous	Homogeneous	Homogeneous
Acidity (pH)	5	4.8	4.7	4.5
Adhesivity (second)	23.36	20.10	09.44	04.58
Spreadability (Cm)*	1.6 – 2.8 Cm	1.6-2.8 Cm	2.5-3.5 Cm	3.0-4.7 Cm

Note: *) spreadibility range weight from lowest (0 grams) to highest (150 grams)

Physical evaluation could be used to determine the stability and properties of the product. In addition, it also provides safety and comfort from using the product. The homogeneity test shows homogeneity of all formulas. These results ensure an even mixture of ingredients. Besides, the homogeneity of the product is associated with good manufacturing practice.[22] The acidity of all products remains in a tolerable range for lip usage (pH 4-6). The higher pH value could affect the stability of betacyanin, while a lower pH value results in incompatibility and irritability on the skin.[20] The adhesivity and spreadability properties describe the product's capability to stick and spread on skin. Both always hold the opposite value, with a lower adhesivity impact on higher spreadability.

In this study, the adhesivity test exhibited good adhesivity (>1 second). Despite that, the spreadability properties of each formula were lower than the standard value (5-7 cm), indicating poor product spreadability.[23] Interestingly, the higher beetroot extract used made the product lower in adhesiveness while also increasing spreadability. This phenomenon was caused by decreased intermolecular forces (Van der Waals interaction, hydrogen bond, etc.) at the skin interface due to the addition of polarity properties of beetroot extract.[23] The formula's evaluation is also used on the respondents, who give favorability based on texture, scent, and color. The hedonistic test further shows the respondent most favorable in F1 for scent; F3 for color and texture. The comfort of cosmetics is influenced by aesthetic value, taste, and smell. However, considering the few respondents, the results could differ because of different aesthetics, sense, or smell values.

CONCLUSION

This research concludes that the best lip balm preparation formula with ethanolic beetroot extract is F3 (10% extract). This

conclusion is based on physical and hedonic evaluation of lip balm.

CONFLICT OF INTEREST

The authors and co-authors state that this research has no conflict of interest.

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