Physicochemical and sensory evaluations of moisturising lip balm using natural pigment from Beta vulgaris

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Physicochemical and sensory evaluations of moisturising lip balm using natural pigment from Beta vulgaris

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Abstract: Nowadays, people are demanding on the naturally derived cosmetic products, including lip balms. However, there is a lack of studies for the physico-chemical properties of the formulated lip balms. Besides, there are little publications found on the use of beetroot as an active ingredient and colourant in lip balm formulation. Thus, this study aims to formulate lip balm using beetroot; and test the physicochemical properties of formulated lip balms to get three best formulations. The stability of these best lip balms was conducted for 4 weeks in room temperature and chiller conditions. Finally, a sensory evaluation was conducted to identify consumer acceptance towards the best lip balms. Lip balms placed in room temperature were all stable while few changes occurred for the lip balms placed in a chiller. All panelist preferred the same lip balm in the sensory test. The result of this study can be extended to assess the potential of beetroot in the formulation of other cosmetic products.

Subjects: Environment & Agriculture; Food Science & Technology; Physical Sciences; Engineering & Technology

Keywords: beetroot; lip balm; physicochemical properties; stability; sensory evaluation

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PUBLIC INTEREST STATEMENT

Lip balm becomes essentials for both women and men as it commonly applied to prevent either inflammation or swelling of lips. People nowadays prefer to use natural and home-made lip balm without notices any adverse effects on the lips if the required tested are not performed after the product formulation. Therefore, the purpose of this study is to find the best all-natural lip balm formulation in term of its appearance (using natural colourant from beetroot) and safety to the user. This naturally derived lip balm will leave your lips soft, moist, non-greasy and healthy-looking.
1. Introduction

Lips are a part of the human body that is very thin as compared to face skin (Kadu et al., 2015) even though it consists of three to four layers of the skin. Lips are susceptible where lips disorder such as inflammation and swelling of lips can quickly occur. Inflammation occurs when the corner of lips become painful, cracked and irritated (Kadu et al., 2015) while swelling happens when some people may have an allergic to certain foods such as seafood and causing the lips to become swelling. Besides, a hereditary condition also can lead to lip swelling (Chaudhari et al., 2018). Therefore, lips must be moisturised using any lip product such as lip balm when the lips disorder appears. The use of tested lip balm is essential to prevent any disease happens.

Currently, lip products made from enormous chemical ingredients have been grown in the market even though it will bring adverse side effects to the users (Kadu et al., 2015). People nowadays are demanding on the naturally derived ingredients as they believed that the natural ingredients could contribute to sound effects on their lips. However, most of the home-made lip balm products (purposely to sell the product) have not tested for their physicochemical properties to make sure the suitability of the product to human lips. The tested lip balm will help in preventing any adverse side effects that could bring any lips disorder to the users.

Formulation of natural lip balm consists of natural ingredients such as beeswax (acts as a base), oil (acts as a solvent) and also natural colouring agent. The natural colouring agent can be varied from any plant, including Beta vulgaris or commonly known as beetroot. Beetroot is a perishable vegetable which the minerals content such as iron and calcium may increase because of the reduction of water mass during storage before it is used (Dhawan et al., 2019). Beetroot contains purplish-red pigment, termed as betalain (Emerton, 2008), a water-soluble and nitrogen-containing dye. The compound comprises the red-violet betacyanin and the yellow betaxanthins (Mereddy et al., 2017). Apart from the colouring agent, there are several active ingredients in beetroot that are beneficial to the human body such as carotenoids (Singh et al., 2014) and anthocyanins (Gengatharan et al., 2015). Carotenoid in beetroot acts as anti-inflammatory, anti-microbial, anti-fungal and rich in nutrients (Kale et al., 2018) while anthocyanins functions as a potent antioxidant, anti-inflammatory and cardioprotective activity (Ahmed et al., 2013; Gerardi et al., 2018; Kong et al., 2003). Anthocyanins can also be a source of red pigment. However, betalains are more water-soluble than anthocyanins, and they have three times higher colouring strength than anthocyanins (Mereddy et al., 2017). Besides, betalains are more stable to pH and temperature than anthocyanins even though it has been widely used in industries. Beetroot is also a good source of dietary fibre that contains vitamins, minerals, and high nutritional because it contains high glucose content (Da Silva et al., 2019).

Therefore, the use of beetroot in the formulation of lip balm can be profitable to the lips. The most important reason is it will be used as a colouring agent due to non-toxic effects on the human and have a natural and safe alternative to be used as red dyes (Elbandy & Abdefadeel, 2008). In this study, all-natural ingredients were utilized in the lip balm formulation to find the best concentration of base, oil and beetroot. Virgin coconut oil was used as oil because it contains natural vitamin E that acts as an antioxidant and helps to increase the shelf life of oil-based products (Rizvi et al., 2014). Beeswax was used as a base for the lip balm formulation because it is more stable due to the higher melting point (Kadu et al., 2015) as compared to another base like cocoa butter. This study focuses on the determination of the best ratio for all ingredients to formulate lip balm from beetroot. It is an important step to balance the concentration of all ingredients used to develop a lip balm to make sure the stability of the product (Ribeiro Fernandes et al., 2013). Besides, this study emphasized the analysis of physicochemical properties of the formulated lip balms before these products are tested in sensory evaluation.
2. Materials and methods

2.1. Plant sample
Beetroot sample was purchased from a local market located at Kota Bharu, Kelantan Malaysia, in May 2019. Beetroot sample was washed under running tap water to remove all residues and impurities. The clean sample was dried using cloth paper for further use.

2.2. Beetroot powder
The spray dryer Model SD 06 supplied by Lab Quip (Cumbria, England) was used to get the dried powder of beetroot. 100 g of cleaned beetroot was cut into small pieces before it was mixed with 500 ml of distilled water and 30 g of Arabic gum that acts as a binder. The mixture was ground and filtered using a muslin cloth with 2 mm pore size. The filtrate was added with distilled water until the solution achieved 1000 ml before it was processed into dried powder using a spray dryer. In this process, the temperature applied was 92°C. The process took 5 hours to get small particle size dried powder of beetroot (<50 μm) and ready to be used in the lip balm formulation. From this drying process, it was found that the moisture content of beetroot sample used was 94.6%.

2.3. Formulation of lip balm
In this study, all-natural ingredients which are virgin coconut oil (VCO), beeswax and dried powder of beetroot were used in the lip balm formulation. The ratio used for beeswax to VCO is ranging from 1:2 to 1:10, and for each ratio, 0.5 g to 2.5 g of beetroot powder was added to the lip balm formulation as stated in Table 1. Twenty-five lip balms were formulated using these ratios and mass of beetroot powder (different red tone as shown in Figure 1). The formulation started by the mixing of VCO with beetroot powder into falcon tube and vortexed the mixture until the powder mixed well with the oil. Then, the solution was filtered using a muslin cloth to prevent any precipitation in the lip balm container. At the same time, the beeswax was heated with a hot plate using an indirect flaming (double boiling) method at 75-85°C with the continuous stirring. The filtered solution of VCO and beetroot powder was added to the hot and fully melted beeswax while continuously stirred the mixture until a proper dispersion achieved. The hot mix of beeswax, VCO and beetroot powder was poured into lip balm containers and placed in the refrigerator at 5°C for an hour and stored at room temperature to stabilize the composition of the lip balm.

2.4. Physicochemical analysis of formulated lip balm

2.4.1. Texture
The formulated lip balm sample was placed on the base of the AMETEK Brookfield CT-3 Texture Analyzer. Cylinder probe (TA39) was attached to the load cell since it is the most suitable probe for cosmetic products (Cosmetics industry applications | Food Technology Corporation, 2019). Then, the probe was lowered to a few millimetres above the sample to align the lip balm sample under the cylinder probe centrally. The hardness readings were recorded for all formulated lip balm samples.

2.4.2. Colour
The colour analysis of lip balms was evaluated using the Konica Minolta CR-400 chroma meter. This chroma meter has three indicators which contributing to lightness (L*), redness (a*) and yellowness (b*) of the tested sample. Based on these three indicators, all lip balms formulated from beetroot are more favour on redness which is a* value.

2.4.3. pH
In this study, the pH meter model HI-2211-01 was used to measure the pH value for all formulated lip balm. The pH meter was calibrated using a buffer solution before continuing the pH measurement of the lip balm. The pH value for the lip balm sample was measured and recorded.
2.4.4. Greasiness
Greasiness test was examined to identify the amount of oil in the formulated lip balm. In this study, 4 g of lip balm was placed on the filter paper, and the sample was left at room temperature for 24 hours. The diameter of the oily ring produced was measured.

2.5. Comparison of formulated and commercial lip balms
Texture, colour, pH and greasiness tests were measured for all formulated lip balm in this study. The commercial lip balm also was tested and measured for all the criteria to compare the value with the expressed lip balm values. The physicochemical properties of the commercial lip balm were used as a benchmark to find the best formulation of lip balm made up from beetroot.

2.6. Stability evaluation of formulated lip balm
Three best formulations that showed the nearest values of all physicochemical properties with the commercial lip balm were selected and scrutinised for their stability. The stability test for lip balms was conducted for 4 weeks to evaluate the sustainability of them when placed at two different temperatures (chiller, 4 ± 1°C and room temperature, 27 ± 1°C). The other physicochemical tests, including pH, colour and texture of the lip balm were conducted for every week of the stability test period. Besides, the spreadability test was also done by spreading the lip balm on the top of the glass slide, and any deformation or breakdown that happened was evaluated as tested by Kadu et

| Formulation | Ratio (Beeswax: VCO) | Beetroot powder(g) |
|-------------|----------------------|--------------------|
| 1           | 1:2                  | 0.5                |
| 2           |                      | 1.0                |
| 3           |                      | 1.5                |
| 4           |                      | 2.0                |
| 5           |                      | 2.5                |
| 6           | 1:4                  | 0.5                |
| 7           |                      | 1.0                |
| 8           |                      | 1.5                |
| 9           |                      | 2.0                |
| 10          |                      | 2.5                |
| 11          | 1:6                  | 0.5                |
| 12          |                      | 1.0                |
| 13          |                      | 1.5                |
| 14          |                      | 2.0                |
| 15          |                      | 2.5                |
| 16          | 1:8                  | 0.5                |
| 17          |                      | 1.0                |
| 18          |                      | 1.5                |
| 19          |                      | 2.0                |
| 20          |                      | 2.5                |
| 21          | 1:10                 | 0.5                |
| 22          |                      | 1.0                |
| 23          |                      | 1.5                |
| 24          |                      | 2.0                |
| 25          |                      | 2.5                |

Table 1. Formulations of lip balm

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Stability and physicochemical properties tests in this part were conducted to prove that the formulated lip balms can be long-lasting and stable for at least 1 month.

2.7. Sensory evaluation test
Sensory evaluation was done based on many purposes and useful for separate the visual olfactory-gustatory assessment (Marcazzan et al., 2017). In this study, the evaluation was conducted to determine the acceptance or likeness of the 100 untrained panels from Universiti Malaysia Kelantan Jeli Campus, Kelantan Malaysia towards the formulated lip balm product. Colour, odour, spreadability and overall acceptance of the lip balm were observed in this sensory evaluation test. Hedonic scale rating was used to collect the information from the panels since the assessment was subjective and based on people-oriented.

3. Results and discussion

3.1. Physicochemical analysis of the formulated lip balm

3.1.1. Hardness
All 25 formulations were analysed based on their texture in term of hardness. Based on Figure 2, the formulation that gives the highest hardness value (308 g) is a lip balm with a ratio of 1:2 (beeswax to VCO) and added with 0.5 g of beetroot powder. Meanwhile, the lip balm formulated using the ratio of 1:10 (beeswax to VCO) and 2.5 g of beetroot powder shows the lowest value of hardness which is 9.33 g. The vast difference between the hardness value for these two
formulations shows that the amount of beeswax and VCO affect the texture of the lip balm (Kasparaviciene et al., 2016).

Based on this figure, formulations with ratios of beeswax to VCO, 1:2, 1:4 have higher values of hardness as compared to the other ratios. The visual observation by naked eyes shows that the data for ratios of beeswax to VCO, 1:2, 1:4 seen to have very thick semi-solid texture. In contrast, the other ratios 1:6, 1:8, 1:10 have a cream-like touch due to the high amount of oil used and make the lip balm feel a bit heavy on the lips (Handmade Lip Balms Deciphered and a Tutorial, 2013). It can be deduced that the higher amount of beeswax in the formulation, the higher the hardness value is. However, some formulations show the fluctuating trends of hardness. These situations occur due to the addition of beetroot powder (solid form) to the formulation, make the formulation harder or softer and affect the value of lip balm hardness. As an example, the addition of 2.5 g of beeswax powder in 1:2 ratio of beeswax to VCO makes the lip balm becomes harder (hardness value is 81.33 g) as compared to the addition of 2.0 g of beetroot powder (hardness value is 65.67 g) with the same ratio of beeswax to VCO ratio. The same case occurs for the formulation with a ratio of beeswax to VCO, 1:6 where the hardness value is 34.67 g for the addition of 2 g of beetroot powder while the value is 19.33 g for the acquisition of 1.5 g of beetroot powder. The mentioned cases obtain because of the increasing amount of solid beetroot powder help in making the lip balm harder.

Conversely, some formulations become softer with the increasing amount of beetroot powder. It can be observed for the formulation with the ratio of beeswax to VCO 1:2 with 1 g of beetroot powder (hardness value is 65.33 g) as compared to the same ratio with 0.5 g of beetroot powder (hardness value is 308 g). The same conditions occur for the ratio of beeswax to VCO 1:4 (addition of 0.5 g and 1 g of beetroot powder), 1:6 (addition of 2 g and 2.5 g of beetroot powder), 1:8 (addition of 1.5 g and 2 g of beetroot powder) and 1:10 (addition of 2 g and 2.5 g of beetroot powder). These cases occur because of the addition of a tremendous amount of liquid (VCO) compared to the solid particles (beeswax and beetroot powder) in the formulations. The increased amount of liquid form (VCO) helps the lip balm become softer.

3.1.2. Colour
Red colour analysis for 25 lip balm formulated with beetroot is presented in Figure 3. In overall, as the amount of VCO increases, the a* value decreases. This phenomenon occurred because of the liquid VCO dissolving the beetroot powder make the solution become dilute and faded colour. In this test, the highest reading (23.94) for colour analysis comes from 1:2, the ratio of beeswax to VCO with the addition of 1 g of beetroot powder. It shows that 1 g of beetroot powder in the lip
balm formulation is the best amount, where the amount exceeding this value decreases $a^*$ readings. Colour analysis for all the lip balms in the ratio of 1:10 for beeswax to VCO shows the lowest among the other ratio where the values range from 14.19 to 18.29. This ratio contains a considerable amount of VCO compared to the beeswax. For this case, VCO dissolves the beetroot powdered and effect the colour when added to the lip balm formulation. A high amount of VCO added in lip balm formulation causes degradation towards the lip balm because betalain stability is influenced by light exposure. These results are supported by those related by Cai et al. (1998) indicating an inverse relationship between light exposure and betalain stability. As betalain is dissolved in a massive amount of VCO, the degradation quickly occurs because of this colourless VCO exposes this pigment to the light. These results are supported by those related by Azeredo et al. (2007) indicating that clear jars suffered higher degradation than that packed in dark jars. In this study, translucent jars can be considered as colourless VCO, while a small amount of VCO can be regarded as dark jars.

3.1.3. pH
Result for pH analysis for all 25 formulations is illustrated in Figure 4. The pigment in beetroot is stable in the pH range 3 to 7 and thus suitable for colouring from sour to neutral foods and cosmetic products (Esatbeyoglu et al., 2015). Although betalains exhibit broad pH stability ranging from pH 3 to 7 (Stintzing & Carle, 2004), pH conditions beyond this range readily induce betalain
degradation. As shown in this figure, the pH values for all formulations range from 4.36 to 5.3. The values are within the scope of stable betalains (pH 3 to 7). The formulation contains 2.5 g beetroot powder and 1:2 ratio of beeswax to VCO shows the highest pH value (pH value is 5.16) while the lowest (pH value is 4.36) is the lip balm contains 0.5 g of beetroot powder and 1:2 of beeswax to VCO.

The cosmetic product with a pH range between 4 and 6.5 is considered as usual for skin, including lips (Lambers et al., 2006). Therefore, the pH of the formulated lip balms in this study is safe and in an ideal range for average human’s skin.

3.1.4. Greasiness
Based on the 25 different formulations, the greasiness for each lip balm was analyzed after 24 hours left on the filter paper in room temperature. The diameter ring of oil was measured to obtain the greasiness of each lip balm. Figure 5 shows that formulations for 1:10 ratio of beeswax to VCO with the addition of 1.5 g, 2.0 g and 2.5 g beetroot powder, respectively, give the highest and the same greasiness value, which is 12.4 cm. This phenomenon occurs because all of the three formulations contain the highest amount of VCO as compared to the other formulations. The same ratio of beeswax to VCO 1:10 with different amount of beetroot powder, 0.5 g and 1.0 g show 10.7 cm and 11.7 cm diameter of oil ring, respectively. In this case, the VCO may be absorbed by beeswax and beetroot powder more than it was incorporated in the other three formulations. Meanwhile, 1:2 ratio of beeswax to VCO with the addition of 0.5 g of beetroot powder shows the lowest value of greasiness because it contained a vast amount of beeswax, which is 6.50 g.

In overall, the greasiness of lip balm depends on the amount of beeswax and VCO. A high amount of beeswax causing less greasy and resistant to oxidation, bacteria and moisture of the formulated lip balm (Kadu et al., 2015). Meanwhile, for the three formulations (1:10 ratio of beeswax to VCO with the addition of 1.5 g, 2.0 g and 2.5 g beetroot powder) that exhibit the highest value of greasiness consisting of a high amount of VCO and a small amount of beeswax. The VCO causes the lip balm to become very greasy and not comfortable for the lips. The greasiness value of commercial lip balm is on the average diameter of ring 7.7 cm. Lip balm that has a similar average reading with commercial is non-greasy and comfortable for lips.

3.2. Comparison of physicochemical analysis of formulated and commercial lip balm
The purpose of measuring all stated physicochemical properties of the formulated lip balms is to find the best lip balm to be tested for stability and sensory. Therefore, a commercial lip balm brand NIVEA with blackberry shine flavour was used to compare its physicochemical properties values with the formulated lip balm. This commercial lip balm was used because the colour of this lip balm is similar to the formulated beetroot lip balm.

Figure 5. Greasiness of lip balm with ratio beeswax to VCO.
Table 2. Best formulation based on commercial lip balm

| Physicochemical properties | Physicochemical properties of commercial lip balm | Best formulation |
|-----------------------------|-----------------------------------------------|-----------------|
| Hardness                    | 50–65 g                                       | • 1:2 (beeswax to VCO) and 1.0 g of beetroot powder  |
|                             |                                               | • 1:4 (beeswax to VCO) and 1.5 g of beetroot powder  |
|                             |                                               | • 1:2 (beeswax to VCO) and 2.0 g of beetroot powder  |
| Colour                      | a* 23.58                                      | • Can be any formulation as long as it gives a* value |
| pH                          | 4–6.5                                        | • All 25 formulations  |
| Greasiness                  | ≤ 7.7 cm                                      | • 1:2 (beeswax to VCO) and 0.5 g of beetroot powder  |
|                             |                                               | • 1:2 (beeswax to VCO) and 1.0 g of beetroot powder  |
|                             |                                               | • 1:2 (beeswax to VCO) and 1.5 g of beetroot powder  |
|                             |                                               | • 1:2 (beeswax to VCO) and 2.0 g of beetroot powder  |
|                             |                                               | • 1:2 (beeswax to VCO) and 2.5 g of beetroot powder  |

Table 2 shows the result for the best lip balm formulation as compared to the commercial lip balm. Three most selected lip balms that give the physicochemical properties in the range of features of commercial lip balm are 1:2 ratio of beeswax to VCO with the addition of 1 g, 2 g and 2.5 g of beetroot powder, respectively. These three formulations were utilized in the stability assessment and sensory evaluation.

3.3. Stability evaluation

Three lip balm formulations, namely, X (1:2 beeswax to VCO with the addition of 2.5 g beetroot powder), Y (1:2 beeswax to VCO with the addition of 2.0 g beetroot powder) and Z (1:2 beeswax to VCO with the addition of 2.5 g beetroot powder) were placed at two different conditions for the stability test. The stability test was conducted to predict the possible changes that may occur on the lip balms (Ribeiro Fernandes et al., 2013). Any potential changes happened in the product can be identified within four period weeks. In this stability test, three selected lip balms were placed at room temperature (27 ± 1°C) and chiller (4 ± 1°C). The attributes such as hardness, colour, pH and spreadability were analyzed, and the result is shown in Table 2.

The hardness analysis of the lip balm was analyzed to identify any changes that occur on the texture of lip balm. Based on Table 3, the hardness of the lip balms placed in room temperature has remained normal (N) for 4 weeks of evaluation. Meanwhile, the hardness of lip balm when placed in the chiller was intensely modified (IM) because the hardness reading increased rapidly from the initial week. There was also white cast identified on the surface of lip balm. It may due to the fat bloom that has to harden during the storage (Kasparaviciene et al., 2016).

The colour changes of the X, Y and Z formulations placed in room temperature were considered as normal (N) for the whole 4 weeks. The purple-red colour of the lip balm made from beetroot was stable in room temperature. Meanwhile, the initial colour of the lip balms in chiller changed during the second, third and fourth weeks. By contrast, the colour assessment was considered as modified when the purple-red colour turned into a lighter colour. According to Kavitkar et al. (2017), the colour of the beetroot will degrade when the temperature is not stable. Hence, the modification of the colour occurred due to the degradation of active compounds in the beetroot.

Besides that, the pH of lip balm formulations in all conditions is within the range that suitable for human’s skin. The optimum pH value for the skin is within 4–6.5 (Lambers et al., 2006). The results for pH in 4 weeks have quite fluctuated, but it is still safe to be used for the lips. When this pH is too
### Table 3. Stability evaluation for 4 weeks

| Attribute       | Storage condition | Time |
|-----------------|-------------------|------|
|                 | Room Temperature  | Chiller |
|                 | t₀   | Week 1 | Week 2 | Week 3 | Week 4 | t₀   | Week 1 | Week 2 | Week 3 | Week 4 |
| Hardness        | X    | N      | N      | N      | N      | N    | IM    | IM    | IM    | IM    |
|                 | Y    | N      | N      | N      | N      | N    | IM    | IM    | IM    | IM    |
|                 | Z    | N      | N      | N      | N      | N    | IM    | IM    | IM    | IM    |
| Colour          | X    | N      | N      | N      | N      | N    | N     | M     | M     | M     |
|                 | Y    | N      | N      | N      | N      | N    | N     | M     | M     | M     |
|                 | Z    | N      | N      | N      | N      | N    | N     | M     | M     | M     |
| pH              | X    | 4.17 ± 0.23 | 3.90 ± 0.61 | 3.84 ± 0.26 | 4.22 ± 0.07 | 4.11 ± 0.10 | 4.31 ± 0.05 | 4.07 ± 0.47 | 4.23 ± 0.21 | 4.43 ± 0.02 | 4.00 ± 0.36 |
|                 | Y    | 4.21 ± 0.20 | 4.62 ± 0.21 | 4.04 ± 0.44 | 3.44 ± 0.50 | 3.35 ± 0.30 | 4.28 ± 0.39 | 3.97 ± 0.36 | 3.96 ± 0.29 | 4.08 ± 0.27 | 4.3 ± 0.24 |
|                 | Z    | 4.25 ± 0.16 | 4.52 ± 0.17 | 3.82 ± 0.32 | 3.99 ± 0.54 | 3.80 ± 0.40 | 4.29 ± 0.31 | 4.29 ± 0.12 | 4.30 ± 0.08 | 4.22 ± 0.21 | 3.86 ± 0.34 |
| Spread ability  | X    | G      | G      | G      | G      | G    | I     | I     | I     | I     |
|                 | Y    | G      | G      | G      | G      | G    | I     | I     | I     | I     |
|                 | Z    | G      | G      | G      | G      | G    | I     | I     | I     | I     |

**All parameters were evaluated in triplicate; t₀: Preparation of formulation after 24 hours, RT: Room Temperature (27°C ± 1), C: Chiller (4°C ± 1).**

For hardness and colour; N- Normal, M- Modified, IM- Intensely Modified

For spread ability: G- Good, I- Intermediate, B- Bad.
acidic, it will cause inflammation on the lips (Kadu et al., 2015). Thus, this study proved that the formulated lip balms are safe to be applied to the lips.

The spreadability of the lip balm was considered as good (G) when there was no deformation occur, and no fragment leaves on the glass slides. The formulations that stored in room temperature were considered as good because it showed an excellent uniformity upon the application. The formulations were also not leaving any fragmentation and deformation throughout the stability tests. The functionality of the lip balm remains normal. Meanwhile, the implementation of the lip balm under chiller condition was not really smooth or wholly inappropriate (I) application since the texture is harder than lip balm placed in room temperature. The spreadability in chiller condition is still uniform, but there are still few fragments left of the glass slides.

3.4. Sensory evaluation
Table 4 shows the ANOVA results. The significant value for colour is $p = 0.658$ where there is no significant difference among the three (X, Y and Z) formulations. The odour and spreadability significance values were $p = 0.546$ and $p = 0.613$, respectively. The spreadability analysis conducted also shows that there is no significant difference between the formulations. It can be concluded that there is no significant difference in all attributes of these three lip balm formulations. When the p-value is above than 0.05, the null hypothesis is accepted.

4. Conclusion
Twenty-five formulations of lip balm made from different ratios of beetroot were successfully formulated. The physicochemical properties of each lip balm were tested; hence, the best formulations were identified. The relationship between the factors (ingredients) and responses (physicochemical properties) had been studied. It showed that each of the ingredients affects the physicochemical properties of the lip balm. Besides, the stability assessment for the best formulations was conducted for 4 weeks to identify any changes occur towards the lip balm. Three best formulations gave almost similar physicochemical properties values (hardness, colour, pH and greasiness) as compared with the commercial

| Table 4. ANOVA for sensory evaluation analysis | Sum of Squares | df | Mean Square | F | Significant value, p |
|---|---|---|---|---|---|
| Colour | Between Groups | 0.827 | 2 | 0.413 | 0.418 | 0.658 |
| | Within Groups | 293.370 | 297 | 0.988 | 0.546 |
| | Total | 294.197 | 299 | | |
| Odour | Between Groups | 1.127 | 2 | 0.563 | 0.607 | 0.546 |
| | Within Groups | 275.540 | 297 | 0.928 | | |
| | Total | 276.667 | 299 | | |
| Spreadability | Between Groups | 0.887 | 2 | 0.443 | 0.490 | 0.613 |
| | Within Groups | 268.500 | 297 | 0.904 | | |
| | Total | 269.387 | 299 | | |
| Overall acceptability | Between Groups | 0.447 | 2 | 0.223 | 0.288 | 0.750 |
| | Within Groups | 230.700 | 297 | 0.777 | | |
| | Total | 231.147 | 299 | | |
lip balm. The colour, pH and greasiness of the best lip balms were within the range of the retail lip balm values. In the stability assessment for 4 weeks, all lip balms placed in room temperature were standard in hardness, pH, colour and had good spreadability. In chiller, these lip balms were intensely modified on the texture while colour and spreadability have slightly changed. Sensory evaluation was conducted, and the results showed that there is no significant difference among the three lip balms. Panelists chose all the lip balm in the same score, and there was an only slightly different. Therefore, these three lip balms (X, Y and Z formulations) can be utilized to be commercialized. From this research, beetroot can be applied as a natural colourant in the lip balm formulation. An attractive purplish-red colour is very suitable to be involved in another cosmetic product such as nailpolish.

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References
Ahmed, J. K., Salih, H. A. M., & Hadi, G. (2013). Anthocyanins in red beet juice act as scavengers for heavy metals ions such as lead and cadmium. Journal of Science and International Technology, 2(3), 269–274. http://www.josic.info/ContentPaper/2013/4-13.pdf
Azredo, H. M. C., Santos, A. N., Souza, A. C. R., Mendes, K. C. B., & Andrade, M. I. R. (2007). Betacyanin stability during processing and storage of a microencapsulated red beetroot extract. American Journal of Food Technology, 2(4), 307–312. https://doi.org/10.3923/ajft.2007.307.312
Cai, Y., Sun, M., & Corke, H. (1998). Colorant properties and stability of Amaranthus betacyanin pigments. Journal of Agricultural and Food Chemistry, 46(11), 4491–4495. https://doi.org/10.1021/jf980407n
Chaudhari, N. P., Chaudhari, N. U., Chaudhari, H., Premchandani, L. A., Dhankani, A. R., & Powar, S. P. (2019). A Review on Herbal Lipstick from Different Natural Colouring Pigment. Indian Journal of Drugs, 6(3), 174–179. http://drugresearch.in/pdf/Oct2018/IJOD-15-nileshwariChaudhari.pdf
Cosmetics industries applications | Food Technology Corporation. (2013). CT3 Texture Analyzer.
Do Silva, D. V. T., Dos Santos Baldo, D., de Oliveira Silva, F., Alves, G., Perrone, D., Del Aguila, E. M., & Flosi Paschoalini, V. M. (2019). Betanin, a natural food additive: Stability, bioavailability, antioxidant and preservative ability assessments. Molecules, 24(3), 458. https://doi.org/10.3390/molecules24203058
Dhawan, D., Sharma, S., Ph., I., & Scholar, D. (2019). Exploration of the nourishing, antioxidant and product development potential of beetroot (Beta vulgaris) flour. International Journal of Health Sciences & Research (www.ijshr.org), 9(6), 280. https://www.ijshr.org/IJHSR_Vol9_Issue6_June2019/IJHSR_Abtract.039.html
Elbandy, M. A., & Abdelfadel, M. G. (2008). Stability of betalain pigments from red beetroot (Beta vulgaris). International Journal of Food Science, 36, 49–60. https://doi.org/10.1016/j.foodchem.2008.12.015
Nurshafieera Idayu Mat Jaine, Azmin, Siti Nuurul Huda Mohammad, & Mohd Shukri Mat Nor, 2016. Studies on the effect of freezing on the stability of beetroot betalin pigments. Cogent Engineering, 3(1), 1678629.

Gengatharan, K., Alwan, S. U., & Elbandy, O. (2019). Betacyanins from Beta vulgaris: Stability and storage conditions. Current Research in Food Science, 2(3), 129–139. https://doi.org/10.1002/mnfr.201900689
Gerardi, C., Albanò, C., Calabriso, N., Carluccio, M. A., Durante, M., Mitò, G., Renna, M., Serio, F., & Blando, F. (2018). Techno-functional properties of tomato puree fortified with anthocyanin pigments. Food Chemistry, 240, 1184–1192. https://doi.org/10.1016/j.foodchem.2017.08.057
Jacobs, J., & Ford, T. (2013). Handmade lip balms deciphered and a tutorial.
Kadu, M., Vishwasrao, S., & Singh, S. (2015). Review on natural lip balm. International Journal of Research in Cosmetic Science, 5(1), 1–7. (2014 August).
Kale, R., Sawate, A., Kshirsagar, R., Patil, B., & Mane, R. (2010). Studies on evaluation of physical and chemical composition of beetroot (Beta vulgaris L.). International Journal of Chemical Studies, 6(2), 2977–2979. https://www.researchgate.net/profile/Rajan_Kale/publication/325057965_Studies_on_evaluation_of_physical_and_chemical_composition_of_beetroot_Beta_vulgaris_L_-_Kale_RG_Sawate_AR_Kshirsagar_RB_Patil_BM_and_Mane_RP/links/5af3edbaaca2720af9c47631/Studies-on
evaluation of physical and chemical composition of beetroot—Beta vulgaris L—Kale—RG—Sawate—AR—Chisrosgar—RP—Patil—BM—and—Mane—RP.pdf
Kasaraviciene, G., Savickas, A., Kalveniene, Z., Velziene, S., Kubiliene, L., & Bernatoniene, J. (2016). Evaluation of beeswax influence on physical properties of lipstick using instrumental and sensory methods. Evidence-Based Complementary and Alternative Medicine, 2016, 1–8. https://doi.org/10.1155/2016/3816460
Kavilkar, R. S., Rao, K. J., Mishra, D., Deshmukh, G. P., Prajapati, R., Jadhao, S. Y., & Scholar, P. D. (2017). Effect of beetroot extract on colour and sensory quality of flavoured milk. International Journal of Pure & Applied Bioscience, 5(5), 1177–1182. https://doi.org/10.18782/2320-7051.2879
Kong, J.-M., Chia, L.-S., Goh, N.-K., Chia, T.-F., & Brouillard, R. (2002). Analysis and biological activities of anthocyanins. Phytochemistry, 64(5), 923–933. https://doi.org/10.1016/S0031-9422(02)00438-2
Lambers, H., Pliessens, S., Bloem, A., Pronk, H., & Finkel, P. (2006). Natural skin surface pH is on average below 5, which is beneficial for its resident flora. International Journal of Cosmetic Science, 28(5), 359–370. https://doi.org/10.1111/j.1467-2949.2006.00344.x
Marczczan, G. L., Mucignot-Caretta, C., Marina Marchese, C., & Piano, M. L. (2017). A review of methods for honey sensory analysis. Journal of Apicultural Research, 57(1), 75–87. https://doi.org/10.1080/00218839.2017.1357940
Mereddy, R., Chan, A., Fanning, K., Nirmal, N., & Sultanbawa, Y. (2017). Betolain rich functional extract with reduced salts and nitrate content from red beetroot (Beta vulgaris L) using membrane separation technology. Food Chemistry, 215, 311–317. https://doi.org/10.1016/j.foodchem.2016.07.132
Ribeiro Fernandes, A., Ferrera Dario, M., Aparecida Sales de Oliveira Pinto, C., Mary Kaneko, T., Rolim Baby, A., & Valéria Robles Velasco, M. (2013). Stability evaluation of organic lip balm. Brazilian Journal of Pharmaceutical Sciences, 49(2), 293–299. https://www.scielo.br/pdf/bjps/v49n2/11.pdf
Rizvi, S., Raza, S. T., Ahmed, F., Ahmad, A., Abbas, S., & Mahdi, F. (2014). The role of vitamin E in human health and some diseases. Sultan Qaboos University Medical Journal, 14(2), 157–165. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3997530/pdf/squmj1402-e157-165.pdf
Singh, B., Hathan, B. S., Devi, C., Memorial, L., & Longowal, S. (2014). Chemical composition, functional properties and processing of beetroot — A review. International Journal of Scientific & Engineering Research, 5(1), 19–38. https://www.scielo.br/pdf/bjps/v49n2/11.pdf
Stintzing, F. C., & Corle, R. (2004). Functional properties of anthocyanins and betalains in plants, food, and in human nutrition. Trends in Food Science & Technology, 15(1), 19–38. https://doi.org/10.1016/j.tifs.2003.07.004