The Potential of Turmeric and Tamarind Leaves Extract (Curcuma Domestica Val - Tamarindus Indica L) as Anti-collagenase Cream

Sri Mulyani¹*, Bambang Admadi Harsodjuwono¹ and Anak Agung Gde Putra Wiraguna²

¹Departement of Agroindustrial Technology, Udayana University, Indonesia
²Department of Health Sciences and Dermatology, Udayana University, Indonesia

ABSTRACT

The aim of this study was to observe the activity of anti-collagenase from turmeric and tamarind leaves extract, the preferable concentration of the extract in the cream, and ability of the cream as anti-collagenase. This research followed some stages as follows. Ability of the extract, as anti-collagenase with concentration (0, 20, 40 and 60) µg/L and the concentration of the extract in cream (0, 50, 100 and 150) mg GAE/100 g cream. Cream from the extracted plants was stored for six weeks and observed for its physical characteristics, organoleptic, and anti-collagenase activity. The extracts of turmeric and tamarind leaves show an anti-collagenase activity with IC₅₀ of 4.403 mg/mL. The cream from the extracted plants with total phenolic of 50 mg GAE/100g indicated a stable condition until the sixth week. The homogeneous cream showed a high viscosity; 10,114 cp and spreading power: 5.6 cm stuck for 30.1 seconds. The separation ratio and the pH of the cream were 1 and 7.43 respectively. The observation results suggested common acceptable organoleptic color, scent, and spreading power with low anti-collagenase activity, IC₅₀: 1.792.75µg/mL.

Keywords: Turmeric; Tamarind-leaf extract; Cream; Anti-collagenase

INTRODUCTION

Food, medicine, and cosmetic industries in Indonesia have been attempting to develop local products based on local wisdom. Turmeric and tamarind are plants widely cultivated in Indonesia and are frequently used in the cosmetic production. The phenolic compounds contained in turmeric and tamarind leaves have a potential as an anti-ageing remedy since both plants provide a skin protection and an anti-wrinkle effect.

Skin ages naturally due to the decreased collagen synthesis and the increased matriks metaloproteinase/MMP expression [1]. Ultraviolet (UV) radiation plays a major role in surging the MMP-1 expression [2]. MMP-1 is a collagenase enzyme [3]. The MMP-1 activity and the number of dermis collagen can be inhibited by natural antioxidants contained in green tea [4] which belong to the phenolic group. Phenolic compounds are effective to impede a collagenase activity [5]. Curcumin is a phenolic antioxidant compound found in turmeric. Clinically, it is known as a good free radical scavenger [6]. Meanwhile, the antioxidants bio-activity found in tamarind leaves are exhibited by vitamin C and a phenolic compound [7].

The combination of two antioxidants and the synergy of both can improve their effectiveness [8]. Turmeric and tamarind leaves extract drink (sinom) has been proven effective in inhibiting the activity of the α-glucoside at 10 ppm as the lowest concentration and 20 ppm as the most optimal concentration [9]. The extract drink has also been proven able to maintain the activity of the SOD enzyme until 56, 26% on a diabetic rat compared to other diabetic rat which was not given the extract drink (14, 16%) [9]. The synergy of antioxidants found in turmeric and tamarind leaves occurred at a ratio of 8:2, with total phenolic of 11.48 ± 0.174 g GAE/100 g [10].

Skin ageing is marked by wrinkles which result from a collagenase activity [3]. The activity of this enzyme can be inhibited by an antioxidant which contains active phenolic compounds (0.05- 0.26 mg GAE/mL) [5]. The total
The phenolic content of turmeric and tamarind leaves extract is 0.12 mg GAE/mL [10]. Therefore, it can be categorized as a collagenase inhibitor. In vivo cream extract of Veronica officinalis (IC50: 105.93 µg/ml) which was smeared on skin for 56 days was able to reduce wrinkles by 66%. [11]. The novelty of the research were found extracts of turmeric and tamarind leaves as anti-collagenase.

MATERIALS AND METHODS

Materials

The Turina-1 turmeric variety plants from BPPT Bogor-Indonesia which were cultivated in Antap village, Tabanan Bali were harvested at the age of nine months. The tamarind leaves used for this research were taken from the buds of local plants in Jimbaran, Badung, Bali, Virgin Coconut Oil/VCO (Sudamala Bali). Other additional materials were Tricine, Sodium chloride, Sodium hydroxide solution, Calcium chloride, dihydrate Hydrochloric acid solution, FALGPA, mineral oil, DPPH, gallic acid (Sigma), Folin ciocalteu phenol, Tiobarbituric acid and sodium carbonate (Merck), stearic acid, triethanolamine, cetyl alcohol, propylene glycol, glycerin and sorbitol (SAP Chemical), TBHQ, buffer phosphate, ethanol (Brathaco Chemical), and cold ultrapure water.

Methods

Stage 1: The ability of turmeric and tamarind leaves extract as anti-collagenase:

The experiment conducted using turmeric and tamarind leaves extract (8:2) was repeated three times with different concentration (0, 20, 40 and 60 µg/L). Data obtained from the trials was analyzed descriptively to determine the IC50 value and the ability of the extract to inhibit collagenase enzymes.

Formulation of turmeric and tamarind leaves extract: Turmeric plants which were harvested at the age of nine months were washed and drained overnight. Each plant was sliced into disks approximately 0.1 cm thick, dried in the oven at 55°C until water content reached 10%. Tamarind leaves were harvested late in the afternoon, washed, and drained overnight. The leaves were put into an oven until water content reached 10%. The turmeric plants and tamarind leaves were mashed into powder with a size of 80 mesh. The turmeric powder was weighed (500 g) and extracted with ethanol 96% at ratio of material and solvent 1:6. The materials were macerated for 24 hours, stirred two times. Filtrate was separated and the pulp was re-macerated. Both filtrates were mixed and steamed using an evaporator at 100 rpm, 40°C, 50 mBar. The same extraction process was repeated on the tamarind leaves until the two extract was obtained.

Research procedures: Samples of turmeric and tamarind leaves extract (8:2) were weighed and diluted based on the three treatment groups 0, 20, 40 and 60, µg/L. The samples were mixed with a solution containing the collagenase substrates and enzymes from Clostridium histolyticum (CHC-EC.3.4.23.3) until the total volume reached 3 mL. The observation of the inhibitory activity performed by the extract was examined using the collagenase test [12]. Data was analyzed descriptively to get a percentage of 50% collagenase inhibition (IC50).

Stage 2: The preferable concentration extract in cream:

Research design: The complete randomized design was applied in three treatment groups of turmeric and tamarind leaves extract concentration (0, 50, 100 and 150 mg GAE/50 g of cream). The experiment was repeated four times until there were 16 units of treatments performed. The variance of the total phenolic content was analyzed and the BNT test was conducted. Organoleptic variables examined were color, aroma, spreading power, and the acceptance of the cream. The data was analyzed using the Friedman test.

Research procedures: The formulated cream materials are presented in Table 1. The turmeric and tamarind leaves extract was mixed and diluted in VCO. The solution was then added with stearic acid, propyl paraben, mineral oil, and setyl alcohol and heated at 65°C. Water-soluble materials were put together to generate moisturizer conditioner which contained the mixture of propylene glycol, glycerin, and sorbitol (2: 1: 1). It was then added with TEA, stearic acid, and water, and heated at 65°C. The water phase was gradually turned into oil phase until it thickened and reached the temperature of 40°C, with constant stirring.
Table 1: Formula of turmeric and tamarind leaves extract cream modified from Bakkara [13]

<table>
<thead>
<tr>
<th>No</th>
<th>Ingredient</th>
<th>Total phenolic concentration mg GAE/g</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0</td>
</tr>
<tr>
<td>1</td>
<td>Turmeric and tamarind leaves extract (g)</td>
<td>0.00</td>
</tr>
<tr>
<td>2</td>
<td>Stearic acid (g)</td>
<td>5.00</td>
</tr>
<tr>
<td>3</td>
<td>Triethanolamine (g)</td>
<td>0.67</td>
</tr>
<tr>
<td>4</td>
<td>Virgin coconut oil (g)</td>
<td>1.67</td>
</tr>
<tr>
<td>5</td>
<td>Mineral oil (g)</td>
<td>1.04</td>
</tr>
<tr>
<td>6</td>
<td>Moisturizer conditioner</td>
<td></td>
</tr>
<tr>
<td>6.1</td>
<td>Propilen glicol (g)</td>
<td>2.50</td>
</tr>
<tr>
<td>6.2</td>
<td>Glicerin (g)</td>
<td>1.25</td>
</tr>
<tr>
<td>6.3</td>
<td>Sorbitol (g)</td>
<td>1.25</td>
</tr>
<tr>
<td></td>
<td>Total ingredients</td>
<td>13.38</td>
</tr>
<tr>
<td></td>
<td>Aquades qs , (g)</td>
<td>50.00</td>
</tr>
</tbody>
</table>

Stage 3: Determining anti-collagenase of the cream:

Research design: The best results of the turmeric and tamarind leaves extract indicated by research stage 2 were observed in terms of their organoleptic and physical characteristics as well as their ability as anti-collagenase enzymes (IC$_{50}$). The analysis was performed for six weeks: the observation was conducted every two weeks. The variance of the homogeneity, viscosity, spreading power, sticking time, pH, and separation ratio of the cream was examined and further analyzed using the BNT test. The organoleptic variables tested included the color, aroma, spreading power, and acceptance of the cream. The data was analyzed using the Friedman test. Meanwhile, data on the anti-collagenase of the cream was analyzed descriptively to determine the IC$_{50}$ value.

The observation procedures:

Measuring the total phenolic [14]: The sample (± 0.1 g) was diluted in methanol until 5 mL in an erlenmeyer flask. It was filtered, and 10 µl was taken out and added with 490 of methanol. The result was taken with a pipette (200 µl) and added with 200 µl of methanol and 400 µl reagen folin-Ciocelteu, and vortexed until it became homogeneous. It was left out for 6 minutes and added with 4.2 mL of 5% sodium carbonate solution. The sample was put in a room temperature for 30 minutes and read at λ=760 nm.

Determining the anti-collagenase activity [15]: The collagenase inhibitor activity was tested out using a method suggested by Moore and Stein, [15] and adjusted with the sigma quality control test procedure enzymatic assay of collagenase using FALGPA. The enzyme solution which contained 2 unit/mL of collagenase in cold ultrapure water (2–8°C) was prepared. In every 3 mL of solution mix, the final concentration measured was 48.3 mM tricine, 9.67 mM Calcium Chloride, 387 mM Sodium Chloride, 0.967 mM FALGPA, and 0.20 unit of collagenase. All was put into the reagen reaction tube using a pipette: (a) test: FALGPA 2.9 mL and (b) blank: FALGPA 2.9 mL + 0.1 mL ultrapure cold water. Mixed with an inversion and equilibrate at 25 ºC, the solution was monitored at (λ) = 345 (A$_{345}$) until it reached a constant condition, using a spectrophotometer and thermostat. The solution was then put into the test tube (a): 0.1 mL enzyme solution. It was mixed with an inversion and observed at (λ) = 345 for 5 minutes. The maximum speed of both reactions was determined, (a) test and (b) blank based on at least one minute interval and four points of data. The inhibitory activity was calculated using the following formula:

\[
\text{Inhibition (\%)} = \left( \frac{A_{\text{Kontrol}} - A_{\text{Sample}}}{A_{\text{Kontrol}}} \right) \times 100\%
\]

Test on the physical characteristics of the cream:

a. **Homogeneity test:** The homogeneity test was performed visually by applying the cream on a petri dish thoroughly. It was observed every two weeks for over a six week period of time [16].

b. **Viscosity test:** The viscosity test was conducted using a viscometer (Brookfield engineering Laboratories, Inc at 2, 4, 10, 20 rpm, spindel no 7. It was also observed every two weeks for over a six week period of time.

c. **Spreading power test:** 0.5 gram cream was put in the middle of a petri dish, covered with another petri dish loaded with 150 gram cream and left out for one minute. The spreading power test was measured afterwards [17].
d. **Sticking time test**: An object glass was marked 4 x 2.5 cm and 0.25 gram cream was put in the middle of the area and covered with another object glass which loaded 1 kg thing for about 5 minutes. A test tool loaded 80 gram was installed on both object glasses which had been attached to each other. Starting from the condition when both glasses were attached to each other until they were separated was determined as the sticking time [16].

e. **Test on cream separation ratio**: This test was performed using the centrifuged fastened separation phase method. The cream was weighed (2 g) and put into a tube. It was centrifuged at 3750 rpm for five hours at a one-hour interval. The separation ratio of oil phase and water phase was measured [18]. The separation volume was calculated using the following formula:

\[
F = \frac{Hu (Separated)}{Ho (Initial)}
\]

f. **pH test**: pH was measured using a digital pH meter.

**Test on the organoleptic characteristics of the cream:**

Organoleptic Characteristics Test: The organoleptic test was conducted using a hedonic test. There were 25 females aged 17-30 years participating in this test. The test was administered in at around 10-12 in the afternoon.

**RESULTS AND DISCUSSION**

**Activity of the Turmeric and Tamarind Leaves Extract as Anti-collagenase**

The average collagenase unit used in this research was 3.78 unit/mg enzyme. The total phenolic content of the turmeric and tamarind leaves extract was 11.48g GAE/100g [10]. The results of the test suggest that the extract could inhibit 50% of the collagenase enzyme activity IC₅₀: 4.403μg/mL. The IC₅₀ is better compared to the IC₅₀ value obtained from the linseed butanol (78.80 μg/ml) [19], and the Pomace grapes mixture (20.3 μg/ml) [20]. The turmeric and tamarind leaves extract was proven stronger in inhibiting radical due to its higher phenolic content value. The phenolic compound found in the extract acted as the complex or settling agent in the collagenase inhibition process [21].

The turmeric and tamarind leaves extract at ratio 8:2 was a much stronger inhibitor because of the synergy effects [10]. Some research have reported that the extract of butanol and *Ginkgo biloba* seeds, pomegranates (*P. granatum*), and mulberry are able to inhibit 50.8%, 4.4%, and 19.5% of collagenase activity respectively (MMP-9) [22]. The mixture of the pomegranates and mulberry plants extract show higher collagenase inhibitory activity. At a concentration of 5 mg/mL, the extract of the fruit mixture could inhibit the enzyme by 67.45% [23]. The high inhibitory activity was performed due to the synergy effect. The collagenase confirmation was changed by the polyphenols compounds [24]. Collagenase is believed to cut off the collagen amino acid bound and break collagen and elastin into pieces. This process sustains the damage on skin which in the end results in wrinkles [25].

**Preferable Concentration of the Turmeric and Tamarind Leaves Extract in Cream**

**Effects of the concentration on cream organoleptic:**

The turmeric and tamarind leaves extract can be categorized into components of biologically active which can be also used for cosmeceuticals [26]. The results of the organoleptic test on the turmeric and tamarind leaves extract are presented in Table 2. The table shows that the cream phenolic content had a significant effect on the organoleptic color (*p*<0.05). The panelists were asked to evaluate the color of the cream and provide responses from “do not really like” to “like” (4.68-5.36). The higher phenolic content makes the cream even yellower. Turmeric contains curcumin phenolic which adds the yellow color to the cream. This color was not liked by the panelists. However, phenolic content had no effects on cream aroma or scent (*p*>0.05) (Table 2). The panelists evaluated the turmeric and virgin coconut oil (VCO) scent as “do not really like” (4.52-4.68). Higher phenolic content results in stronger turmeric scent left on the product. This is caused by the turmeric essential oil content (5%) which consists of turmerone, borneol, sineol, phellandrene, curcumin, and zingeron [27]. The dominant VCO aroma made the panelists unable to distinguish the extract concentration of the cream.
The addition of the extract did not have any effects on the spreading power of the cream \( (p>0.05) \) (Table 2). The panelists provided “do not really like” response \( (4.72-5.16) \). The biggest component contained in the turmeric extract is phenolic compounds which consist of curcumin \((15.95\%),\) bidesmetoksi curcumin \((15.95\%),\) demetoksi curcumin \((5.95\%),\) meanwhile the tamarind leaves extract contains 1-methoxy-4-[2-(4-phenylphenyl)ethenyl] benzene \((8.68%)\) [28]. These compounds are oil-soluble so that they can be thoroughly dispersed in cream emulsion. Therefore, they do not affect the spreading power of the cream. The spreading power of the cream needs to be measured in order that the cream can be applied well, and washed easily with water.

The turmeric and tamarind leaves extract did not have any effects on the general acceptance of the cream \( (p>0.05) \) (Table 2). The panelists evaluated it as \( (4.83-5.08) \) which belongs to “do not really like” category. The acceptance value of the cream might be mostly affected by the aroma and the spreading power of the cream, so that the extract concentration had no effect on the acceptance value by the panelists. This research finding suggests that the panelist may prefer a scent addition to the cream to disguise the VCO and turmeric smell.

**Effects of the concentration on the cream total phenolic content:**
The concentration of the turmeric and tamarind leaves extract had a significant effect on the total phenolic content of the cream \( (p<0.05) \) (Table 3). The addition of the extract to the cream \((50-150 \text{ mg GAE/g})\) did not have any effect on the total phenolic content of the cream. It indicates that the cream from the turmeric and tamarind leaves extract belong to the cosmeceuticals category which contains active phenolic compounds [26].

The phenolic compounds contained in the cream has an important role since they function to catch free radicals and inhibit the collagenase enzyme activity. The natural collagenase inhibitor produced from turmeric and tamarind leaves extract is much safer as an anti-ageing. The increase extract concentration \((50-150 \text{ mg GAE/g})\) had no significant effect on the phenolic compounds content. It occurred because the extract concentration added was small [5].

The results of the organoleptic test shows that the extract concentration \((150 \text{ mg GAE/g})\) did not have any real effect on the cream organoleptic variables, except on color. The panelists gave the highest score to color produced by the extract addition at \(50 \text{ mg GAE/g}\). These results also suggest that the addition of \(50 \text{ mg GAE/g}\) of the extract to the cream apparently had no effect on the total phenolic content of the cream. Therefore, \(50 \text{ mg GAE/g}\) is considered as the best concentration to add to the cream.

**Table 3: The average total phenolic of cream turmeric and tamarind leaves extract**

<table>
<thead>
<tr>
<th>Concentration phenolic of turmeric and tamarind leaves extract (mg GAE / 100 g cream)</th>
<th>Average phenolics (mg GAE / 100 g cream)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>(1765 \pm 0.699^*)</td>
</tr>
<tr>
<td>50</td>
<td>(3449 \pm 0.294)</td>
</tr>
<tr>
<td>100</td>
<td>(4602 \pm 1056)</td>
</tr>
<tr>
<td>150</td>
<td>(4671 \pm 1278)</td>
</tr>
</tbody>
</table>

\(^*\)Average grade testing ± standard deviation; \(n=4\)

**The Characteristics and Ability of the Cream as Anti-collagenase**

**Physical characteristics of the cream:**

**Homogeneity:** The cream had remained homogeneous for six weeks. The homogeneity of the cream is required to show the effectiveness of the active substances contained in it. Homogeneous cream will affect the effectiveness of the therapy if it is applied thoroughly on the skin with the same concentration. Cream can be determined stable if during the storage process, it is not separated and the viscosity value of the cream remains the same for five weeks [29].

**Cream viscosity:** Up to week-6, the viscosity of the cream was more likely to decrease (Table 4). The decline was caused by the decreased stability of the emulsion from time to time. It was marked by the increased globule size at the internal phase and the decreased globule speed so that the liquid flow became slower. The ups and downs in the viscosity value during storage indicate that the cream is unstable. Bushe [30] states that the viscosity value of the


c...
cream that is sold in the market must be at least 30,000 cp, 5 rpm, 25°C. Research findings suggest that the viscosity of the cream at week 6 was 10,113.0 cp which means that the cream has complied with the terms and conditions.

The power spreading of the cream had been stable for six weeks (Table 4). The cream stability indicates that the emulsion of the cream is constant so that no globule separation is reported. The turmeric and tamarind leaves extract has met the specification standard (SNI) of spreading power which is around 5-7 cm [31]. High power spreading value indicates wide skin surface that can be applied with the cream. Spreading power of more than 7 cm results in the decreased effectiveness of the cream because the concentration of the active substances get lower.

The results of the sticking time observation for six weeks show a decrease (Table 4). The decline was caused by the changing viscosity value of the cream although it still complied with the standard at the sixth week. Sticking time of the cream is related to the duration of the cream contact with the skin and users friendliness. Excellent cream will guarantee that the cream can have an effective contact with the skin, or in other words, not too sticky. Sticking time also ensures the effectiveness of the active substances of the cream. The longer the cream is put on the skin, the more effective the active substances will be.

During the storage process, the pH of the cream tended to increase (Table 4). The purpose of the pH test was to examine the safety of the cream; whether it does not leave skin irritation. The changing pH of the samples indicates the unstable emulsion which release triethanolamine as the emulsifier. Triethanolamine is base released if the emulsion is damaged and can therefore increase pH. The cream pH during the storage process can exceed the limit of the skin physiology pH (4-7). If it gets lower than the skin physiology pH, skin irritation may occur. However, higher pH can also result in skin irritation and dry skin [32].

The effect of storage on cream aroma or scent has not been reported (F=1). This finding suggest that the cream was stable after being centrifuged at 3,750 rpm for 5 h. It was reported that [18] a centrifugation at 3,750 rpm for 5 h is equal to a year gravitation effect. Therefore, it can be concluded that cream from the turmeric and tamarind leaves can be stored for a year at the room temperature without having to experience the separation phase.

**Organoleptic characteristics of the cream:**

**Color and spreading power:** The results of the test on the organoleptic characteristics of the cream suggest that six-week storage can adversely affect the color (p<0.05) and the spreading power (p<0.05) of the cream (Table 5). The results of the observation show that the cream became much darker after six weeks. This color changing has resulted from the phenolic compounds oxidation which makes the cream much darker eventually [33].

Cream storage also influences the spreading power of the cream (Table 5). The spreading power is related to the distribution of the cream when it is applied. The panelists gave the highest score to the cream that was stored the longest. It means that the panelists preferred cream that can be easily spread on skin. An increase in the spreading power may also cause by the decreased viscosity of the cream (Table 4).

Research findings suggest that storage had no effect on the cream aroma or scent (p>0.05) (Table 5). The panelists evaluated the aroma as 4.17-4.67 which means that they did not really like it. Such responses resulted from the fact that the cream contains strong turmeric and VCO smell which makes the panelists unable to distinguish the storage effect. Therefore, it is recommended to add more fragrance to the cream so that people are going to like it.

![Table 4: Viscosity, scattering, stickiness and pH cream of turmeric and tamarind leaves extract](image)

<table>
<thead>
<tr>
<th>Time observation (week)</th>
<th>Viscosity at 5 rpm</th>
<th>Scattering (cm)</th>
<th>Stickiness (second)</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>6,293.5</td>
<td>5.81</td>
<td>111.33</td>
<td>7.36</td>
</tr>
<tr>
<td>2</td>
<td>6,774.0</td>
<td>5.83</td>
<td>78.00</td>
<td>7.44</td>
</tr>
<tr>
<td>4</td>
<td>9,440.3</td>
<td>5.89</td>
<td>50.17</td>
<td>7.40</td>
</tr>
<tr>
<td>6</td>
<td>10,114.0</td>
<td>5.57</td>
<td>30.10</td>
<td>7.40</td>
</tr>
</tbody>
</table>

The panelists was not able to distinguish the storage effect because the cream contained strong turmeric and VCO smell which makes the panelists unable to distinguish the storage effect. Therefore, it is recommended to add more fragrance to the cream so that people are going to like it.

![Table 5: The organoleptic test of color, spreading power, aroma and overall acceptance of turmeric and tamarind leaves extract cream](image)

<table>
<thead>
<tr>
<th>Observation (week)</th>
<th>Color</th>
<th>Spreading power</th>
<th>Aroma</th>
<th>Overall acceptance</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>5.33 ± 0.43</td>
<td>5.17 ± 0.37*</td>
<td>4.67 ± 1.17</td>
<td>5.17 ± 1.494*</td>
</tr>
<tr>
<td>2</td>
<td>4.67 ± 0.39</td>
<td>5.33 ± 0.51</td>
<td>4.54 ± 0.88</td>
<td>5.04 ± 0.955</td>
</tr>
<tr>
<td>4</td>
<td>4.92 ± 0.32</td>
<td>6.04 ± 0.69</td>
<td>4.58 ± 1.02</td>
<td>5.04 ± 0.550</td>
</tr>
<tr>
<td>6</td>
<td>4.13 ± 0.42</td>
<td>5.42 ± 0.49</td>
<td>4.17 ± 1.20</td>
<td>4.75 ± 0.897</td>
</tr>
<tr>
<td>P/ Significance</td>
<td>0.014</td>
<td>0.021</td>
<td>0.695</td>
<td>0.204</td>
</tr>
</tbody>
</table>

*Average grade testing ± standard deviation; n = 24

Table 5 presents the six-week storage did not have any effect on the general acceptance of the cream (p>0.05). The general acceptance of the cream ranged from 4.83 to 5.08 which means that the panelists did not really like it. The
panelists did not provide different responses towards the cream until week 6. It indicates that the six-week storage did not adversely affect the panelists’ responses and they can still use it after all.

Collagenase Inhibitory Test of Cream Extract Turmeric and Tamarind Leaves
Collagenase activity inhibition test showed that the IC$_{50}$ cream increased during storage for six weeks. The values of IC$_{50}$ cream from weeks 0, 2, 4 and 6 were 1.545.0, 1.743.99, 1.768.14 and 1.792.75 µg/mL, respectively, during storage of cream decreased collagenase inhibition activity. The cream experienced a decline in its ability to inhibit collagenase until 16.0%. This finding is contradictory with the results of the research conducted by KT Farrel [27] which shows a stable IC$_{50}$ value for six weeks. Storing the extract for six weeks in three different temperature levels (25 ± 2)°C; (0 ± 2)°C and (-10 ± 2)°C did not have any effect on the difference in the IC$_{50}$ value. Plants extract which is able to inhibit the activity of the collagenase enzyme has total phenolic content of (0.05-0.26) mg GAE/mL [5]. The turmeric and tamarind leaves extract with total phenolic content of 0.115 mg GAE/mL is therefore able to serve as the collagenase inhibitor. Cream preferred by the panelists contains low phenolic (0.04 mgGAE/mL) so that the inhibitory power is also low. The collagenase inhibitor is important as an anti-ageing which results from the reduced collagen found in dermis due to the collagenase activity. It was reported that wrinkles appear as the number of collagen fiber of skin is decreasing [34].

CONCLUSION
As a conclusion of this paper is: turmeric and tamarind leaves extract at ratio 8:2 contains high total phenolic 11.48 g GAE/100 g. with anti-collagenase, IC$_{50}$: 4.403 µg/mL. Cream produced from the preferred concentration of turmeric and tamarind leaves extract contains 50 mg GAE/100 g. The cream was considered stable during storage. The physical characteristics of the cream are homogeneous and viscosity of 10.114 cp, spreading power of 5.6 cm, sticking time of 30.1 s, and separation ratio: 1, and pH 7.43. The panelists liked the color and the aroma, but not too much. The spreading power and the general acceptance of the cream belong to the “do not really like” category. The anti-collagenase of the cream IC$_{50}$: 1.792.75µg/mL, potential is still considered low.

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