The development of Moringa leaves effervescent granules with effervescent agent of citric acid and sodium bicarbonate

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Submitted: 10-06-2021 Reviewed: 10-07-2021 Accepted: 26-07-2021

ABSTRACT

Moringa leaves are part of Moringa Oleifera, which have a high antioxidant content, therefore, it is beneficial for overcoming free radicals in the body. Furthermore, the leaves contents have the potential to be developed into a functional drink in the form of effervescent granules. The important aspects influencing the physical characteristics, stability, and acceptability of granule is the type and ratio of the effervescent agent. In this study, citric acid and sodium bicarbonate were used as effervescent agents because they both provide acceptable texture and mouthfeel. This study aimed to develop an effervescent granule formula of Moringa leaves and to optimize the molar ratio of citric acid and sodium bicarbonate as the effervescent agent. Three molar ratios of citric acid and sodium bicarbonate were optimized, namely 1:3.11 (Formulation 1), 1:3 (Formulation 2), and 1:2.93 (Formulation 3). These formulations were developed using the wet granulation method. The results showed effervescent granules of the three formulations exhibited good flowability and uniform particle size distribution. Furthermore, formulation 3 showed better granule flow characteristics than the others. The results after reconstitution showed the granules were quickly dispersed within 207-234 secs, the pH of the preparation ranged between 5.74-5.92 (neutral pH), the viscosity was between 326-333 cps, and exhibited dilatant flow characteristics. Also, the organoleptic and sensory evaluation results showed formulation 3 was the most acceptable in terms of color, taste, aroma, and texture. Based on these findings, it was concluded that formulation 3 which used (citric acid ratio, sodium bicarbonate 1:2.93) was the optimal formulation.

Keywords: Moringa leaves, effervescent, granules, citric acid, sodium bicarbonate

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Journal homepage: http://journal.uad.ac.id/index.php/PHARMACIANA
INTRODUCTION

Moringa oleifera is a potential source of antioxidants which is due to the presence of phenolics phytochemical compounds such as quercetin and kaempferol (Arabshahi-D et al., 2007). Besides, other compounds which act as natural antioxidants are vitamin A, C, and E. The natural antioxidant content in dried weight basis of Moringa leaves includes 74-210 μmol/g for phenolics, 70-100 μmol/g for vitamin C, 1.1-2.8 μmol/g for carotene, and 0.7 1.1 μmol/g for tocopherols (vitamin E) (Yang et al., 2006). The antioxidants in the leaves are higher than other vegetables and fruits such as strawberries, carrots, and soybeans. Therefore, Moringa leaves are a source of natural antioxidants which are useful for counteracting free radicals and inhibiting cellular oxidation (Gopalakrishnan et al., 2016).

Moringa leaves with high antioxidants can be further processed to maintain the component of functional compounds. Previous studies showed drying and freezing is the selected method to allow for long-term storage while maintaining the contents (Nambiar and Parnami, 2008). Drying is a simple and cost-effective method for processing the leaves, hence it can be stored for a longer time. The results showed that drying the leaves at a temperature of 50-60°C maintained the nutrients content and other phytochemical compounds, except for vitamin C (Olabode et al., 2015). Also, increasing the functionality of the leaves as raw material and fortification in food, beverage, and pharmaceutical preparations was carried out by processing into powder. The results showed the addition of Moringa leaves powder in food and beverage products increased the nutritional and antioxidant content (Sengev et al., 2013).

One of the suitable preparation forms to be developed with the main content of Moringa leaves powder is effervescent granules. This is a functional drink in the form of granules designed to be dissolved or dispersed in water before consumption (Aslani and Jahangiri, 2013). The advantages of the granules include, ease of swallowing, rapid onset of action, do not irritate the gastrointestinal tract, have an acceptable taste, higher stability than liquid preparations, and is easy to handle after packaging (Aslani and Jahangiri, 2013). Furthermore, the granules produce carbon dioxide gas (CO2) due to the reaction between an acid source (citric acid, malic acid, tartaric acid, and other suitable acid sources) and bicarbonate compounds or their salts (sodium bicarbonate, sodium carbonate, and potassium bicarbonate) when in contact with water (Aslani and Fattahi, 2013). The released CO2 provides a fresh sensation like soda taste and overrides the bitterness of the active ingredients. These advantages have a positive impact on the development of Moringa-based functional drinks (Ali et al., 2018).

The main components of the granule formula are the active ingredient and the effervescent agents. The effervescent agents consist of acid and alkaline sources. Other excipients that are commonly used include fillers, binders, disintegrants, sweeteners, flavors, dyes, and surfactants (Jassim et al., 2018). The combination of acid and other sources widely used in previous studies is citric acid and sodium bicarbonate. Citric acid has an advantage over other acid sources due to its solubility in water and provides an acceptable sour taste to the preparation. Furthermore, sodium bicarbonate dissolves completely in water, it is not hygroscopic, can be consumed safely, is widely used in food products as baking soda, and gives a fresh sensation like sparkling water (Grajang and Wahyuningsih, 2019). Moreover, it is a weak base compound. The solution which contains 0.85% of sodium bicarbonate produces a solution pH of 8.3. This solution also produces 52% carbon dioxide to the whole solution when reacting with water in effervescent preparations. The presence of gas from this reaction, will mask the bitter taste and enhance the dissolution process (Herlina et al., 2020). The combination of citric acid and sodium bicarbonate as an effervescent agent has been proven to produce carbonated wine powder with water content, pH, and dispersion time according to the requirements (Sandrasari and Zaenal Abidin, 2006). Also, the ratio of citric acid and sodium bicarbonate affects the physical characteristics of the granules. According to previous studies, the amount of citric acid determines the particle size, specific gravity, flow characteristics, and moisture content of the granules. Therefore, an increase in the citric acid proportion causes the particle size to increase, easier to flow, and the moisture content also increases (Grajang and Wahyuningsih, 2019). This moisture increase is
due to the hygroscopic characteristics of citric acid (Wati and Saryanti, 2019). Also, sodium bicarbonate proportion determines the physical properties of the granule, such as flow and moisture content. The results showed the use of sodium bicarbonate in large concentrations helps to absorb moisture in the granules (Aslani and Jahangiri, 2013). In addition, the ratio of citric acid and sodium bicarbonate has a significant impact on the granule characteristics after reconstitution, such as dispersion time, pH, and sensory acceptance (Budi et al., 2011).

The development of effervescent granules with the active ingredient of Moringa leaves powder has not been previously reported. Hence, the granules were developed with three molar ratios of citric acid to sodium bicarbonate, namely formulation 1 (1:3.11), 2 (1:3), and 3 (1:2.93). The result from the previous study implied that the molar ratio of citric acid and sodium bicarbonate is an essential factor to determine the characteristics of effervescent granules. The difference in the ratio affected the physical properties, the characteristics after reconstitution, and sensory analysis. The molar ratio of the effervescent agent in this study contained sodium bicarbonate in a higher proportion than citric acid, hence the intense effervescent reaction, rapid dissolution of the granules, and shorter effervescent time will be obtained. The citric acid-sodium bicarbonate ratio of 1:3 causes the acid to completely neutralized, and the pH of the solution tends to neutralize (Taymouri et al., 2019). The slight increase of citric acid proportion in formulation 3 and the slight decrease of citric acid proportion in formulation 1 was designed to evaluate the effect of remaining acid or base in the solution on the disintegration time, pH of dispersion, and the taste of dispersion. Therefore, this study aims to develop Moringa leaves effervescent formula in the form of granules with physical characteristics which fulfill the requirements and are accepted by consumers, as well as to optimize the ratio of citric acid and sodium bicarbonate as an effervescent agent.

MATERIALS AND METHODS
Equipment and materials

The equipment used includes a blender (Panasonic), mixer (Electrolux), analytical balance (Mettler Toledo), oven (Memmert), a set of sieves and vibrator (Retsch D-42759 HAAN), tapped density tester (Pharma Test D-63512 Hainburg), moisture content analyzer (Mettler Toledo), climatic chamber (Binder), standard funnel, stopwatch, stative, Stormer Serial 80202 viscometer (Arthur H Thomas Co. Philadelphia USA), and pH meter (Horiba Scientific).

The study materials were included in the category of pharmaceutical grade (p.g) or food-grade (f.g), and the plant used was Moringa leaves (Moringa oleifera) harvested from Bogo Village, Bojonegoro, East Java. Furthermore, the part of the plants namely Moringa leaves was determined at the Center for Information and Development of Traditional Medicine (PIPOT) Faculty of Pharmacy, the University of Surabaya with a letter of determination no 1412/D.T/X/2019. Also, the chemicals used include citric acid pg (Weifang Ensign Industry Co., Ltd), sodium bicarbonate FG (Chruch and Dwight Co., Inc USA), xanthan gum pg (Shandong Fufeng Fermentation Co., Ltd), maltodextrin DE15-20 pg (Zhucheng Dongxiao Biotechnology Co., Ltd), sodium benzoate pg (Kalama Chemical), sucrose fg (PT. Brataco, Indonesia), stevia pg obtained from PT. Jammu Iboe-Indonesia, Poloxamer 188 p.g (Merck Millipore), melon flavored powder (KH. Roberts, Indonesia), and strawberry flavored powder (KH. Roberts, Indonesia).

Method

Preparation of Moringa leaves simplicia

Moringa leaves were harvested from Bogo Village, Bojonegoro, East Java. The fresh leaves were washed, wet sorted, drained, dried by aerating in the shade at 25-35 °C and humidity was not more than 50%. Furthermore, drying was carried out until the moisture content was < 10% (Suganthi et al., 2019); (Departemen Kesehatan Republik Indonesia, 2008). Subsequently, the dried leaves were stored in a tightly closed container before the continuation of the particle size reduction process (Ali et al., 2017). Formulation into powder form aimed to maintain content stability for a long time before...
being processed further (Singh and Singh, 2014). In addition, the dried leaves were reduced in particle size and sieved using a 100 mesh sieve (Ali et al., 2017).

**Production of Moringa leaves effervescent granule**

The granules were developed by the wet granulation method. This was carried out separately between the acidic and basic components (Budi et al., 2011). In this study, three molar ratios of effervescent agents (citric acid and sodium bicarbonate) were optimized, namely 1:3.11 (formulation 1), 1:3 (formulation 2), and 1:2.93 (formulation 3). The components are presented in Table 1.

**Table 1. Formulation of Moringa leaves effervescent granule with citric acid and sodium bicarbonate as effervescent agent**

| Material                  | Formulation 1 (Gram) | Formulation 2 (Gram) | Formulation 3 (Gram) |
|---------------------------|-----------------------|-----------------------|-----------------------|
| Moringa leaves dried powder | 2                     | 2                     | 2                     |
| Citric Acid               | 2.23                  | 2.27                  | 2.30                  |
| Sodium Bicarbonate        | 2.77                  | 2.73                  | 2.70                  |
| Xanthan Gum               | 0.5                   | 0.5                   | 0.5                   |
| Sucrose                   | 10.5                  | 10.5                  | 10.5                  |
| Stevia                    | 1.6                   | 1.6                   | 1.6                   |
| Sodium Benzoate           | 0.05                  | 0.05                  | 0.05                  |
| Poloxamer 188             | 0.125                 | 0.125                 | 0.125                 |
| Maltodextrin              | 2.15                  | 2.15                  | 2.15                  |
| Melon Flavor              | 2.075                 | 2.075                 | 2.075                 |
| Strawberry Flavor         | 1                     | 1                     | 1                     |
| Purified water            | 2.0 mL                | 2.0 mL                | 2.0 mL                |

**Description:**
The total amount of agent (citric acid and sodium bicarbonate) of each formulation was 5 grams. The molar ratio of citric acid to sodium bicarbonate in the formulation 1-3 was 1:3.11, 1:3, and 1:2.93 respectively. The weight ratio of citric acid and sodium bicarbonate from these three formulations were 4:5 (formulation 1), 5:6 (formulation 2), and 6:7 (formulation 3)

The granules production was carried out at a controlled temperature and humidity of 20-25°C, RH 50-60% respectively. In the initial stage, a mixture of all formulation components was prepared except citric acid (acid source) and sodium bicarbonate (base source). Furthermore, the leaves powder, xanthan gum, sucrose, stevia, sodium benzoate, poloxamer 188, maltodextrin, melon flavor, and strawberry flavor were mixed using a Y-cone tumbling mixer (1 kg of capacity) on 100 rpm for 5 mins to obtain mixture 1. Mixture 1 was weighed for acidic and basic components according to the ratio of citric acid and sodium bicarbonate in the formula (Budi et al., 2011). The ratios of the acid components weighed from mixture 1 for the three formulas were 4/9 parts (formulation 1), 5/11 parts (formulation 2), and 6/13 (formulation 3). Subsequently, weighed mixture 1 was added with citric acid and stirred until homogeneous in a Y-cone tumbling mixer (1 kg of capacity) on 100 rpm for 5 mins to obtain mixture 2. The ratio of the base components which have been weighed from mixture 1 to the three formulas was 5/9 parts (formulation 1), 6/11 parts (formulation 2), and 7/13 (formulation 3). Hence, the weighed mixture 1 was added with sodium bicarbonate and stirred until homogeneous in a Y-cone tumbling mixer (1 kg of capacity) on 100 rpm for 5 mins to obtain mixture 3. The mixing process has been validated in the previous study, hence the homogeneity of the mixture can be assured.

Mixtures 2 and 3 in each formulation were separately granulated using the wet granulation method. The fluid added was purified water, and the process was continued until a granule mass was obtained. Furthermore, the granules were sieved through mesh No. 10 sieves and dried in the oven.
The drying was carried out at a temperature of 50°C for approximately 3 hours until a moisture content of 3-5% was reached. The dried granules for both acidic and basic components were sieved using a mesh no. 16 sieve (Okoye et al., 2013). Subsequently, the granules of the acid and base components were mixed using a Y-cone tumbling mixer at 100 rpm for 5 mins, according to the ratio in each formula. In the next stage, the granules were filled in the primary packaging (sachets) as much as 30 grams per sachet.

**Evaluation of granule physical characteristics**

**Organoleptic evaluation**

Moringa leaves effervescent granules were organoleptically observed including shape, color, smell, and taste.

**Evaluation of flowability and angle of repose**

Evaluation of flowability and angle of repose aims to predict the flow characteristics of the granules. This was determined using the funnel method (Thoke et al., 2013). The granules which weighed as much as 100 grams were put in a funnel with a closed bottom hole. At the time of measurement, the bottom hole was opened, while the time required for all the granules to flow down and form a heap was determined using a stopwatch. Flowability was calculated using the following formula:

\[
\text{Flowability} = \frac{W}{t} \tag{1}
\]

Description:  
\(W = \) powder weight (g)  
\(t = \) the time needed by the powder to flow (s)

The granule heap formed on a flat plane was measured for the angle of repose, with the formula:

\[
\alpha = \arctan \frac{h}{r} \tag{2}
\]

Description:  
\(\alpha = \) angle of repose(°)  
\(h = \) heap height (cm)  
\(r = \) heap radius(cm)

**Evaluation of bulk density, tapped density, hausner ratio, and compressibility index**

Measurement of bulk and tapped density was carried out by weighing 100 grams of granules and pouring them into a 250 mL measuring cup. The cup was tilted at a 45° angle when the granules were poured. Subsequently, the measuring cup was erected and shaken rapidly to even out the granule surface. The volume read was used to calculate the bulk density. The bulk density was calculated by the following formula:

\[
\text{Bulk density} = \frac{W_0}{V_0} \tag{3}
\]

Description:  
\(W_0 = \) granule weight  
\(V_0 = \) granule volum
to 500 taps. Furthermore, the volume in the measuring cup was observed, until three consecutive observations showed a constant volume (Vt). Tapped density was calculated by the following formula:

\[
\text{Tapped density} = \frac{W_0}{V_t}
\]  

\[(4)\]

Description:  
\[W_0 = \text{granule weight}\]
\[V_t = \text{granule volume after compression}\]

The results of the bulk and tapped density were used to calculate the derived parameters of the granule flow characteristics, namely the Hausner ratio and compressibility index (Allen and Ansel, 2014). The Hausner ratio was calculated through the ratio between the bulk density and the tapped density, meanwhile, the compressibility index was determined by the following formula:

\[
\text{Compressibility index} = \frac{\text{Bulk density} - \text{Tapped density}}{\text{Tapped density}} \times 100\%
\]  

\[(5)\]

Evaluation of granule particle size distribution

Evaluation of the granule particle size distribution was carried out with a standard sieve set. The sieves and collection pans were orderly arranged from top to bottom as follows, 20, 30, 50, 60, 80, 100, and collection pans. Furthermore, the sample used was 100 grams of effervescent granules. At the initial stage, each sieve and collection pan were weighed, and the arrangement was placed on the “Retsch Vibrator. Subsequently, the granules were placed on the top sieve, closed, and tightened. A set of sieves was vibrated at a frequency of 60 Hz for 20 mins and weighed. Based on the data obtained, the weight of the granules contained in each sieve and the collection pan was determined. In addition, the weight data distributed on each sieve was processed into a particle size distribution curve (Okoye et al., 2013).

Evaluation of the granule moisture content

Moisture content evaluation was carried out by weighing 5 grams of the granules and placing them on the moisture content analyzer. The instrument was run for 15 mins with an annealing temperature of 100°C. The previous validation exhibited that for these granules 15 mins had been reached a constant weight. The moisture content was indicated on the instrument in percentage (Sanzida, 2018).

Evaluation of Moringa leaves effervescent granule characteristics after reconstitution

Dispersion time and foam height

Dispersion time was carried out by dispersing 30 grams of granules (1 sachet) into 150 mL of purified water in a beaker glass. The time required until all the granules were homogeneously dispersed in the purified water was recorded as the dispersion time. The dispersion time requirement for the granules was 5 mins (Pandey et al., 2013). In addition, the foam height was measured after the granules were dispersed to determine the dispersion ability.

Evaluation of preparation pH

The pH measurement of the reconstituted effervescent preparation (30 grams) with 150 mL purified water was carried out using a Cyberscan 510 pH meter (Giyatmi and Lingga, 2019).
Evaluation of flow properties and viscosity

The viscosity and flow properties after reconstitution were determined using a Stormer (cup and bob) viscometer. Glycerin has been applied as standard fluid to determine the viscometer constant and load correction. A total of 400 mL of the dispersed granules was placed into a cup and the time needed by Bob to travel 100 rounds at a certain additional load was observed. The laminar flow of the dispersed granules was maintained by providing relaxation time to the sample after the shearing stress was applied. Based on these data, the preparation viscosity was calculated. The flow properties of the preparation were determined by plotting the load vs rpm and rpm vs viscosity. Based on the rheogram obtained, the flow properties of the granule preparation were determined (Noval et al., 2020).

Organoleptic and sensory evaluation

The reconstituted granules were subjected to organoleptic as well as sensory evaluation including color, taste, smell, and consistency. The test was carried out on 40 trained panelists. Each panelist was given their reconstituted formula to be tasted with the tongue. For every change of formula, panelists need to first rinse their mouths with 10 mL of purified water. The panelists gave an assessment on a scale of 1-5 for each parameter (color, taste, smell, and consistency. The details on the scale given are as follows, 1= strongly dislike, 2= dislike, 3= quite like, 4= like, 5 = strongly like (Husni et al., 2017).

Data Analysis

Evaluation of physical characteristics including organoleptic, specific gravity, flowability, angle of repose, Hausner ratio, and particle size distribution was descriptively carried out. The results were compared with the physical quality requirements of granules contained in the compendia (Indonesian Pharmacopoeia VI and USP 40), as well as the optimal results in previous studies. Meanwhile, the effect of differences in the ratio of citric acid and sodium bicarbonate as an effervescent agent on the reconstituted characteristics (dispersion time, pH, viscosity, and foam height) was carried out by statistical analysis using a one-way ANOVA test at a 95% confidence level. The test was continued with LSD when there were significant differences in each parameter of the three formulations. The organoleptic and sensory evaluation results based on the panelists’ assessment were tabulated to determine their preferences. Furthermore, panelists' assessment scores for each parameter, namely color, taste, smell, and consistency were analyzed statistically with the Kruskal-Wallis test at a 95% confidence level (Rahmawati and Adi, 2017).

RESULTS AND DISCUSSION

Moringa leaves were used as a nutritious ingredient in effervescent preparations. The leaves were dried until the simplicia moisture content was < 10%. This drying process was an effort to maintain the nutritional stability, and to suppress the moist content of Simplicia to prevent the growth of bacteria, fungi, and molds (Clement et al., 2017). Based on a previous study, drying by aeration at room temperature is an economical method as it maintains the nutrient content and manages a high amount of bioactive content. This method also maintains the highest phenolic content compared to other drying techniques (Ali et al., 2017). Further processing of dried leaves simplicia was milled, and particle size was reduced to produce the powder. Processing into powder form is an effort to increase functionality and effectiveness. The leaves powder can be added to various food products, beverages, or developed into pharmaceutical preparations as nutraceutical products (Chinwe et al., 2013). The granules were prepared by the wet granulation method. The process for acidic and basic components was carried out separately to avoid premature reactions during granulation (Budi et al., 2011). The granules of the three formulas were dark green and have a distinctive aroma as shown in Figure 1. The evaluation results on physical characteristics are shown in Table 2.
Measurement of bulk density aims to determine the material bulk. This parameter described the volume that will be occupied by a certain number of granules, hence have an impact on determining dosage pack and the process of filling into the primary package (Van den Ban dan Goodwin, 2017). Furthermore, determination of compressibility index and Hausner ratio were used to indirectly ascertain the flow characteristics. The compressibility index measured the bond strength between particles and bond stability, while the Hausner ratio measured interparticulate friction (Khandelwal and Shah, 2016). Lower values of the compressibility index and Hausner's ratio indicated better granule flow characteristics than those with higher values of both parameters (Saw et al., 2015). Based on the compressibility index and Hausner ratio, it was discovered that the granules of formula 1 were included in the good flow characteristics. Meanwhile, formulas 2 and 3 showed very good flow characteristics (compressibility index 10% and Hausner ratio in the range of 1.00-1.11). The difference in the ratio of citric acid to sodium bicarbonate affected the flow characteristics in terms of compressibility index value and Hausner ratio. Therefore, increasing the proportion of citric acid in the effervescent agent increased the flow characteristics (Grajang and Wahyuningsih, 2019). This is because citric acid has high specific gravity and good flow characteristics (Sun et al., 2020).

Flow characteristics can also be predicted through evaluation of the flowability and angle of repose. The evaluation results showed the three formulas included in the category could flow well (4-10 g/s) (Augsburger and Hoag, 2008). Also, the angle of repose showed all the granules could flow well (31-35°). Based on the results obtained, it was concluded that the granules developed in this study were able to flow well. Good flow characteristics have an impact on the weight diversity and uniformity of dosage content (Murphy et al., 2020). Also, the wet granulation process proved efficient in influencing particle shape, surface morphology, and powder size. In addition, the spherical shape of the particles, a smooth surface, and the uniform distribution of the particle size have an impact on improving the flow characteristics compared to powders (Murphy et al., 2020).
Another parameter that determined the granule characteristics was the moisture content. Effervescent granules easily absorb moisture from the air. Changes in moisture parameters affect the flow characteristics and dissolution rate of the active ingredients (Mutahar et al., 2008). In general, the requirement for the moisture content of granules or powders was around 3-5% (Crouter and Briens, 2013). Based on the evaluation results, the three formulations had a moisture content that fulfills the requirements. This amount of moisture was needed to form a particle bond, hence a compact and spherical granule was obtained. Furthermore, moisture has a significant impact on the real density and granule flow characteristics. A very high moisture content caused the formation of a stronger bond bridge between the particles. This phenomenon can be physically observed as sticky particles or cake forms. This condition has an impact on the decrease in granule flow characteristics (Jung et al., 2018). A very high granule moisture content harms the microbiological stability preparation. This is because a certain amount of moisture in the product activates microbial growth (Rezaei and Vander Gheynst, 2010).

Particle size evaluation was carried out to determine the distribution of granules as well as the range with the largest proportion, and percentage of fines. The evaluation results showed the largest proportion was in the particle size of > 850 μm, while 40-50% were in the size range of 300-600 μm and 600-850 μm. According to previous studies, granules with a narrow size distribution improved the flow characteristics. Also, they showed an increase in the uniformity of granule content due to a narrow particle size distribution, which was in the range of 410-710 μm (Santl et al., 2012). The proportion of fines in the three formulas < 10% showed the risk of static charge during the filling process in the primary packaging can be minimized. The static charge during this process caused the fines to be thrown and not filled into the package, which also affected flow characteristics. This is very crucial especially when fines proportion mostly consists of active ingredients (Shah et al., 2008).

Evaluation of granule characteristics after reconstitution aims to observe the effect of different ratios of effervescent agents on dispersion ability and suspensions formed. The evaluation results of the reconstituted granule characteristics are tabulated in Table 3. Also, the reconstituted granules

### Table 2. Evaluation results on physical characteristics of Moringa leaves effervescent granules

| Parameter                  | Formulation 1 | Formulation 2 | Formulation 3 | Specification |
|----------------------------|---------------|---------------|---------------|---------------|
| Organoleptic               | Granules, dark green, typical Moringa aroma | Granules, dark green, typical Moringa aroma | Granules, dark green, typical Moringa aroma | Granules, dark green, typical Moringa aroma |
| Bulk density (g/mL)        | 0.57±0.00     | 0.60±0.01     | 0.58±0.00     | -             |
| Tapped density (g/mL)      | 0.64±0.00     | 0.66±0.01     | 0.62±0.01     | -             |
| Compressibility index (%)  | 10.42±0.30    | 9.90±0.50     | 7.67±0.45     | 1-15% (Allen and Ansel, 2014) |
| Hausner Ratio              | 1.12±0.15     | 1.11±0.20     | 1.08±0.10     | 1.00-1.18 (York et al., 2001) |
| Flowability (g/s)          | 8.71±0.40     | 9.32±0.26     | 8.78±0.48     | 4-10 g/s (Augsburger and Hoag, 2008b) |
| Angle of repose (°)        | 33.69±0.00    | 33.67±1.78    | 32.64±1.73    | 25-35° (York et al., 2001) |
| Moisture content (%)       | 4.07±0.09     | 4.01±0.04     | 4.17±0.02     | 3-5% (Crouter and Briens, 2013) |
| % fines                    | 1%            | 0%            | 0%            | <10% (Allen and Ansel, 2014) |

*Description: Results are expressed as mean ± SD value of three replications.*

The development of Moringa ...(Rani et al.,)
produced a green suspension with a sweet taste and a typical Moringa aroma. Meanwhile, evaluation of dispersion time served to predict the effervescent reaction velocity until all the granules were dissolved or completely dispersed in water. Granules are dissolved or completely dispersed when all the fragments are evenly distributed and the gas release stops (da Cunha-Filho et al., 2014). The dispersion time of the three formulas fulfilled the requirement of < 5 mins (Grajang and Wahyuningsih, 2019). The statistical analysis results using the one-way ANOVA test showed a significant difference (p<0.05) in the time dispersion of the three formulations. The dispersion time of formulation 1 was significantly different from 2 and 3, but there was no difference in dispersion time between formulation 2 and 3 based on LSD test results (p>0.05). Formula 1 with a ratio of citric acid to sodium bicarbonate (1:3.11) showed a faster dispersion time due to the higher porosity of formula 1. Particle cavities help the granule disintegration process because liquids enter more easily and assist the granules to disintegrate (Dewi et al., 2014). The results showed the greater the proportion of acid component in the effervescent agent, the longer the dispersion time. This is because, at a higher proportion of acid, sodium bicarbonate decreases. The decrease in sodium bicarbonate causes a reduction in porosity and the carbonation rate (Herlina et al., 2020). The stoichiometric reaction of acid-base in formulation 1 implied that sodium bicarbonate remained after the reaction. The greater amount of base compared to the amount of acid caused a more intense effervescent reaction, hence the rapid dispersion and dissolution of the granules have been observed (Giyatmi and Lingga, 2019). The acid-base ratio in formulation 2 showed that citric acid and sodium bicarbonate were completely reacted. Moreover, in formulation 3 slight amount of citric acid remained in the final effervescent reaction. The amount of citric acid in the terminate reaction didn’t play a significant role to enhance the dispersion time compared to formulation 2. Hence, the dispersion times of formulations 2 and 3 were not significantly different.

Table 3. Evaluation results of Moringa leaves effervescent granule characteristics after reconstitution

| Parameter                          | Formulation 1                          | Formulation 2                          | Formulation 3                          | Specification                  |
|------------------------------------|----------------------------------------|----------------------------------------|----------------------------------------|---------------------------------|
| Organoleptic                       | Suspension is the green, sweet taste, typical Moringa aroma | Suspension is the green, sweet taste, typical Moringa aroma | Suspension is the green, sweet taste, typical Moringa aroma | Suspension is the green, sweet taste, typical Moringa aroma < 5 minutes (Grajang and Wahyuningsih, 2019) |
| Dispersion time (seconds)          | 207.33±2.52                           | 227.33±3.06                           | 234.67±5.51                           | < 5 minutes                     |
| Foam height(cm)                    | 6.00±0.00                              | 6.33±0.58                             | 6.67±0.29                             | -                              |
| pH                                 | 5.89±0.03                              | 5.92±0.01                             | 5.74±0.01                             | 5.00-6.00 (Taymouri et al., 2019) |
| Viscosity(cps)                     | 326.18±1.94                           | 333.03±6.64                           | 328.13±2.57                           | 200-400 cps (Wagoner et al., 2020) |
| Flow characteristics               | Dilatant                               | Dilatant                               | Dilatant                               | -                              |

The average foam height of the granule ranged between 6.00-6.67 cm. Foam is a collection of thousands of tiny bubbles due to chemical reactions or mechanical treatment such as stirring. The bubbles are also known as foam accumulate quickly and move to the surface. The evaluation results showed the difference in the ratio of effervescent agents had no significant effect (p>0.05) on the foam height after reconstitution. This height correlated with the dispersion time of the granules. The
granules which are quickly and completely dispersed often reach saturation conditions and stop producing foam. Meanwhile, a longer dispersion time will cause an increase in the foam height (Widyaningrum et al., 2015). This condition is under the results that formulation 1 with the fastest dispersion time produced the lowest foam height. Based on these data, it was concluded that the foam produced was directly proportional to the granule dispersion time.

The granules after reconstitution showed the preparation pH range of 5.74-5.92. The pH of the resulting preparation (4.0-9.0) was not too acidic and was under the pH range of the antioxidant component stability in Moringa leaves (Arabshahi-D et al., 2007). The slightly acidic preparation pH also provided a fresh taste to the effervescent granule dispersion (Dewi et al., 2014). The effervescent preparation pH is one of the parameters affecting consumer acceptance. High acidic preparation pH irritates the stomach, while high alkaline pH causes a bitter and uncomfortable taste (Grajang and Wahyuningsih, 2019). The difference in the ratio of citric acid and sodium bicarbonate as effervescent had a significant impact on the preparation pH (p<0.05). There was a significant difference in the pH of formula 3 compared to formulations 1 and 2 (p<0.05) based on the results of the LSD test. The pH of the effervescent formulation 3 was lower than the other two. Increasing the ratio of citric acid and sodium bicarbonate (6:7) caused a significant decrease in pH. This is because the increment of acid proportion has a role in increasing the concentration of unreacted ions [H3O+] in the solution (Budi et al., 2011). The pH of the effervescent granule preparations for the three formulations was less than 6.00 because the unreacted citric acid produced a more acidic solution pH. The resulting acidity increases consumer taste perceptions (Jassim et al., 2018). Therefore, based on the previous studies, it was concluded that there was an interaction between the proportions of citric acid and bicarbonate in determining the effervescent pH (Budi et al., 2011).

Evaluation of the viscosity and flow properties of the granules after reconstitution was carried out to predict consistency, ease of pouring, and dispersion stability. The evaluation results of the sample viscosity with the Stormer Cup and Bob viscometer which is manually operated showed that the dispersion viscosity ranged between 326-333 cps at shearing stress of 100 grams. The statistical analysis effect of the effervescent agent ratio on the preparation viscosity after reconstitution with ANOVA showed that there was no effect of ratio difference on the preparation viscosity (p>0.05). This was due to the interaction of acid and base components as effervescent agents that do not trigger changes in viscosity, moreover, the difference in proportions between formulations was not too large. Factors that have more influence on the preparation viscosity include the concentration of the suspending agent, thickener, and temperature (Herlina et al., 2020). The reconstitution results showed the characteristics of the dilatant flow. The increase in RPM on the stormer viscometer caused a rise in the preparation viscosity. This was due to the high proportion of solid particles (Moringa leaves powder) in the preparation, hence the increase in stirring speed drain the solid particles and caused the amount of water in the system to be insufficient. The flow properties rheogram of the three formulas are shown in Figure 2.

Organoleptic and sensory evaluations including color, aroma, taste, and texture were carried out to determine the panelists' acceptance. Furthermore, the evaluation was carried out on 40 trained panelists, the assessment was performed on a preference scale. The liking scale was divided into 5 scores with the following descriptions, 1 = strongly dislike, 2 = dislike, 3 = quite like, 4 = like, and 5 = strongly like. The assessment was carried out on each parameter including color, taste, smell, and texture. Assessment of color parameters is important because it relates to the level of product visual acceptance. Aroma parameters are related to consumer perceptions of the food and beverage product taste. Meanwhile, the assessment of taste can be performed in the presence of chemical responses by the taste buds. Taste is an essential parameter for consumers to accept or reject a food product. The texture or mouthfeel of a food ingredient plays a role in product acceptance and the panelists' ability to chew or swallow the product (Andarini et al., 2018).
Figure 2. Shearing stress vs rate of shear rheogram of Moringa leaves effervescent granule after dispersion

The distribution of panelists' acceptance on the parameters of color, aroma, taste, and texture of the three Moringa leaves effervescent granule formulations is shown in Figure 3. The evaluation results showed that the percentage of panelists who stated quite like to like from the color parameter was 62.50%-65.00%. Meanwhile, in terms of aroma, taste, and texture parameters, the percentage of panelists who stated that they quite like to like was around 72.50%-85.00%. The average score tabulation of the panelists' acceptance in terms of color, aroma, taste, and texture is shown in Table 4. The mean value of the panelists' acceptance scores (mean rank) for these four parameters ranges from 3.00 to 3.50, indicating that the preparation was acceptable to the panelists. This is because the value was in the range of quite like (score = 3) to like (score = 4). The color showed a lower average value than the other parameters. This condition was caused by the result of granule reconstitution which showed a dark green color. In the next development, it is necessary to improve the color parameters by adding dyes (FD&C) such as yellow or light green to increase the brightness of the granule color.

Figure 3. Evaluation results of panelist acceptance of Moringa leaves effervescent granules
The development of Moringa leaves effervescent granules (Rani et al.,)

Statistical analysis of panelist acceptance was carried out using the Kruskal-Wallis test at a 95% confidence level. It showed that there was no significant difference (p>0.05) in panelists' responses to the color, aroma, taste, and texture of the effervescent granule preparations. In general, based on the results, it was concluded that the three formulas were well received. Formulation 3 acceptance was better than the others in terms of color, aroma, taste, and texture based on the average score.

### Table 4. Distribution of panelists' preference level for Moringa leaves effervescent granules

| Parameters | Formulation 1 | Formulation 2 | Formulation 3 | p-value |
|------------|---------------|---------------|---------------|---------|
| Color      | 3.00          | 3.03          | 3.05          | 0.988   |
| Aroma      | 3.48          | 3.40          | 3.43          | 0.957   |
| Taste      | 3.45          | 3.23          | 3.63          | 0.192   |
| Texture    | 3.23          | 3.18          | 3.23          | 0.948   |

### CONCLUSION

Moringa leaves effervescent granule formula developed in this study had good flow characteristics, uniform particle size distribution, and fulfills quality requirements. Furthermore, formulation 3 showed better flow characteristics than the other formulas. The evaluation results of the reconstituted granules showed the three formulas were rapidly dispersed within 207-229 seconds, the preparation pH ranged between 5.73-5.91 (neutral pH), the viscosity was between 326-333 cps, and showed dilatant flow characteristics. Also, results of the organoleptic and sensory evaluation showed the three formulas were acceptable to the panelists. In addition, formulation 3 (citric acid ratio sodium bicarbonate 6:7) was the most acceptable in terms of color, taste, aroma, and texture.

### ACKNOWLEDGMENTS

The authors are grateful to the LPPM (Research institutions and community service) Faculty of Pharmacy, University of Surabaya, and the Bogo Village Government for participating in this study.

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