Formulation and Evaluation of Effervescent Granul from African Leaf Extract (*Vernonia amygdalina* Del.)

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A B S T R A C T

African leaf plants (Vernonia amygdalina Del.) contain many secondary metabolites, one of which is flavonoids. African leaves are commonly used in traditional medicine. This study aims to determine the extract of African leaf plants can be formulated in the preparation of effervescent granules and is suitable for the criteria for good granules. African leaves were macerated using 96% ethanol as solvent and evaporated to obtain a thick extract. Then, the African leaf extract obtained was formulated into effervescent granules with a concentration of 10%. The production of effervescent granules is based on the principle of the wet granulation method. The results showed that the evaluation of the effervescent granules of African leaf extract was only suitable for the requirements of the haussner factor, porosity, particle size distribution, moisture content, angle of repose, dispersion, and pH but did not suitable for the requirements of compressibility and flowability. The flavonoid content in the African leaf extract is 96,2276% while the effervescent granule is 94,634%.94,634%.

Keywords: African leaf, *Vernonia amygdalina* Del., Effervescent Granules

Introduction

Indonesia is a country with a very abundant flora, people can use plants as a traditional medicine to minimize unwanted side effects. Traditional medicine that has been used for generations by ancestors aims to save time and cost, improve health, will be safe when consumed appropriately, and have side effects that can be estimated (Supardi & Susyanty, 2010). As many as 3.4 billion of the world's population including Indonesia are very dependent on traditional medicine because 88% of the population uses drugs made from natural ingredients for the continuation of their primary health (Sarker & Lutfun, 2007).
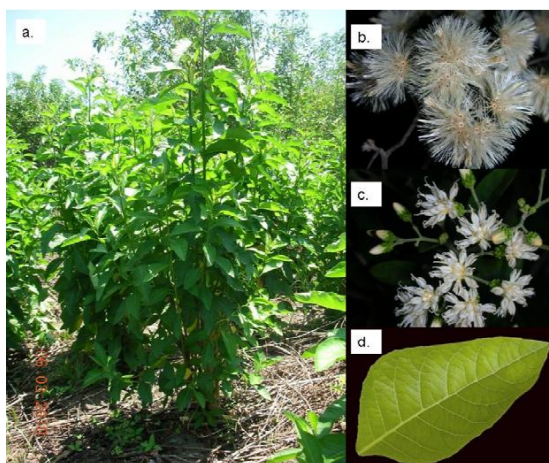

Figure 1. Vernonia amygdalina plant (Yeap et al., 2010)

One of the plants used as a folk remedy is the African leaf plant (*Vernonia amygdalina* Del). African leaf or also known as bitter leaf (Fatimah & Sundu, 2020), is a plant that contains nutrients and chemical compounds that are used in traditional medicine and can be used for various diseases, such as cancer drugs, heart disease treatment, lowering cholesterol, preventing stroke, regulating blood sugar, lowering fever, malaria, hepatitis, coughing,

anti-inflammatory, antioxidants and others (Saraswati et al., 2019; Solikhah et al., 2021; Yeap et al., 2010). African leaves contain flavonoid compounds, alkaloids, tannins, saponins, terpenoids, and lactones sesquiterpenes (Saraswati et al., 2019; Solikhah et al., 2021).

So far African leaf extract has been formulated in the dosage form of direct felt tablets (Wahyudi & Sari, 2021), nanoparticle suspension (Wirasti et al., 2020), burn ointment (Lahagina et al., 2019), lotion (Zamzam & Indawati, 2020), cream (Nainggolan et al., 2018), gel (Meilani & Kusumastuti, 2019), and others.

In society, the use of African leaves is still very simple, in the form of simplicity powder preparations or decoctions of African leaves that are drunk as infusion preparations. This method is less effective for minimizing the bitter taste of African leaf steeping. In this study, African leaves were processed into extracts which were then formulated into effervescent granules. Effervescent granules are coarse to coarse granules or powders and contain medicinal elements in a dry mixture, usually consisting of sodium bicarbonate, citric acid, and tartaric acid. When water is added, the acid and base will react to release carbon dioxide, resulting in foam. A solution with the resulting carbonate can mask the salt flavor or other undesirable tastes of the medicinal substance (Lachman et al., 1994). Based on the above background, this study aims to formulate and evaluate effervescent granules from African leaf extract (*Vernonia amygdalina* Del.)..

Methods:

The materials used in this study were African leaves (*Vernonia amygdalina* Del.) (Kab. Agam, West Sumatra), ethanol 96% (Novalindo), citric acid (Novalindo), tartric acid (Novalindo), sodium bicarbonate (Novalindo), lactose (Novalindo), aspartame (Novalindo), PVP K30 (Novalindo), quercetin (Sigma Aldrich), methanol p.a (Merck), acetic acid (CH_3COOH) (Merck), Aluminum chloride ($AlCl_3$) (Merck).

African Leaf Extract Preparation

Put 500 grams of dry powder of simplisia in the macerator, and add 5 L of 96% ethanol. Soak for the first 6 hours while stirring occasionally, then let stand for 18 hours. Separate the macerate by filtration. Repeat the filtering process three times with the same amount of solvent until macerates 1, 2, and 3 are obtained. Collect all the macerat, then vaporize it with a vacuum vaporizer or low-pressure vaporizer can also use a rotary evaporator (Heidolph) until a viscous extract is obtained. Then determine the percentage of extract amendments.

PreEffervescent Granule Manufacturing

Effervescent granules are made by the method of separate granulation between the acid part and the base part so that no premature effervescent reaction occurs. The effervescent granule formula can be seen in Table I. Half of the formula (African leaf extract, citric acid, tartaric acid, lactose, aspartame, and PVP solution) is mixed with all the ingredients until it is done. The mixture is sifted through 16 mesh and dried in a 40°C oven for 18 hours (acidic part). A portion of the formula (African leaf extract, sodium bicarbonate, lactose, aspartame, and PVP solution) is mixed with all the ingredients until it is smoothed/formed into a dough. The mixture is sifted through 16 mesh and dried in a 40°C oven for 18 hours (alkaline part). Mix the granules of the acidic part and the base part into a container through a 16-mesh sieve. Then transfer it into a tightly closed container and protected it from sunlight. After that, evaluate the granules.

Table I. Formulation of each granule for 200 grams

Material	Formula (%)	
	F0	F1
African leaf extract	-	10
Citric acid	6,5	6,5
Tartaric acid	8,5	8,5
Sodium bicarbonate	15	15
Aspartame	1	1
PVP K30	10	10
Lactose ad	200	200

Evaluation of Effervescent Granules

Organoleptic Test

The effervescent granule odor test is placed on the palm of the hand and smelled the aroma. Test the form, the resulting shapes are as much as possible the same as each other. Color test, effervescent granules are observed color directly by looking at the physical shape directly, wherever possible it looks homogeneous.

Powder particle size measurement with vibrator sieve (Symore ZN48)

Weigh each sieve and then weigh 100 grams of sample, placed on the top sieve, then sifted for 10 minutes. Re-weigh each sieve with the granule until a difference in the weight of the granule on each sieve is obtained. Based on the data obtained, a graph of the granule frequency distribution curve can be made (Halim, 2012).

Flowability study

Bulk density and tapped density (Al-Mousawy et al 2019)

Two types of density were determined (bulk density (BD) and tapped density (TD). In a 100 ml measuring cylinder, an appropriate amount of granules was weighted and put; then the initial volume was recorded. After that, the measuring cylinder was tapped at the height of 2.5 cm at 2-second intervals until no further change was noted in the volume . From the equation below, bulk density and tapped density were calculated.

$$BD = \frac{granules\ weight}{Packing\ Volume}$$

$$TD = \frac{granules\ weight}{Votapped\ voleme\ of\ the\ packing}$$

Where BD is the bulk density and TD is the Tapped density.

Carr's index for ibuprofen granules was measured to evaluate the bulk density and tapped density. The values of carr"s indexs of African leaf extract granules wew compares with references as shown in table 2.

$$Carr'sindex = \frac{\{(TD-BD)x\ 100\}}{TD}$$

Hausner's ratio of ibuprofen granules was calculated by using the equation below. Hausner's ratio which is less than 1.25 shows good flowing properties more than higher ones. Hausner's ratios which are from 1.25 to 1.6 show moderate flowing properties. Hausner's ratio which is more than 1.6 will show more cohesive powders [9].

$$Hausner\ ratio = \frac{tappe\ density}{bulk\ density}$$

Table 2 : Flow properties and compressibility index

Flow characters	Carr"s Index
Excellent	1 -10
Good	11-15
Fair	16-20
Passable	21-25
Poor	26-31
Very poor	32-37
Very, very poor	>38

The angle of repose determination (Halim, 2012; Wells, 2004)

Weigh 30 grams of granules and put into a flow tester that has been coated with graph paper. At the same time open the lid of the funnel and the granules are allowed to flow freely onto the available graph paper. Then there will be a pile of cone-like powder. Measured the height of the pile (h) and the diameter (d) of the powder so that its radius is calculated (r), then the angle of its stacking is a stationary angle that can be calculated in the following way:

tg θ = h/r atau cos θ = r/s, dimana s = hipotonus

or the tangent angle is equal to the height of the stack divided by the radius of the stack. The values of the angle of repose were compared with references as shown in table 3.

Table 3 : Flow properties of the angle of respose

The angle of repose value	Flow properties
< 20	Excellent
20-30	Good
30-34	Passable
> 40	Very poor

Determination of water content (Voight, 1994)

Evaluation of water content was carried out using *an infrared moisture balance* tool (Ohaus MB45).

Dispersion time test

Put 100 mL of cold water with a temperature of 15-25ºC into a 250 mL beaker. After that, 5 grams of *effervescent* granules are put into the water. When the granule is dispersed in water and completes the reaction within < 5 minutes, it shows a perfectly dispersed preparation (Gopalan & Gozali, 2018).

pH test

The solution in the dispersion test was measured using a pH measuring device (Ionix) and the measurement results were said to be good if the pH of the effervescent solution was close to neutral (Gopalan & Gozali, 2018).

Determination of Total Flavonoid Levels of *Effervescent* Granules from African Leaf Extract

Quercetin as much as 10 mg was carefully weighed and then dissolved in 100 mL of methanol p.a for 1000 ppm. Next is created a concentration series to obtain a standard curve. Take absorbance measurements at a maximum absorption wavelength of 415.5 nm using a *double beam* UV-Vis Spectrophotometer (Shimadzu UV-1800).

An *effervescent* granule of 100 mg african leaf extract equivalent to 10 mg extract was carefully weighed then dissolved with methanol p.a in a measuring flask of 100 ml. Then pipette 0.6 ml into a measuring flask 10 ml then add sodium acetate 1 M 0.1 ml, $AlCl_3$ 10% 0.1 ml and aqua dest to the limit mark. Take absorbance measurements at a maximum absorption wavelength of 415.5 nm using a *double beam* UV-Vis spectrophotometer

(Shiamdzu UV-1800). The sample solution was prepared in 3 replications and the total flavonoids were calculated using the linear regression equation of the quercetin calibration curve.

RESULTSAND DISCUSSION

African leaves contain various phytochemical compounds, one of which is flavonoids. Phytochemical compounds usually have to be extracted from plants using a solvent through an extraction process. One of the extraction methods that can be used is maceration. The extraction results were converted into the yield percentage. The extraction results showed that the yield of the extract obtained was 11.6230%.

Effervescent Granule Evaluation:

Effervescent granules are granules or powders that are coarse to very coarse and contain medicinal elements in a dry mixture, usually consisting of sodium bicarbonate, citric acid, and tartaric acid, when added with acidic water and the base reacts to liberate carbon dioxide to produce foam (Ansel, 1989). The manufacture of effervescent granules uses the wet granulation method separately so that the initial effervescent reaction does not occur.

The granule organoleptic test can be observed that the granules are coarse, dark green to yellowish, and have a characteristic odor.

Figure 2: African leaf extract effervescent granule

The particle size distribution test aims to determine the range of granule particle size and particle size distribution which can be seen from how many granules are left in each mesh number. This test was carried out using a vibrator sieve. The importance of this test is because the particle size can affect the flow rate of a powder. The finer the particle size, the lower the flow rate. This is because the cohesiveness between particles is getting bigger. Granules that are distributed more at a certain size mean the quality of the granules or the uniformity of particle size is good (Halim, 2012: Lachman et al., 1994).

From the results obtained, the granules are very coarse powders because no more than 20% passes through a 60-mesh sieve. The granule particle size distribution data is made in the form of a frequency distribution curve graph which can be seen in Figure 1. The frequency distribution curve is the relationship between the average particle size and frequency. It can be seen on the graph that the high frequency on a 60-mesh sieve is said to be large and the average granule size is > 250.
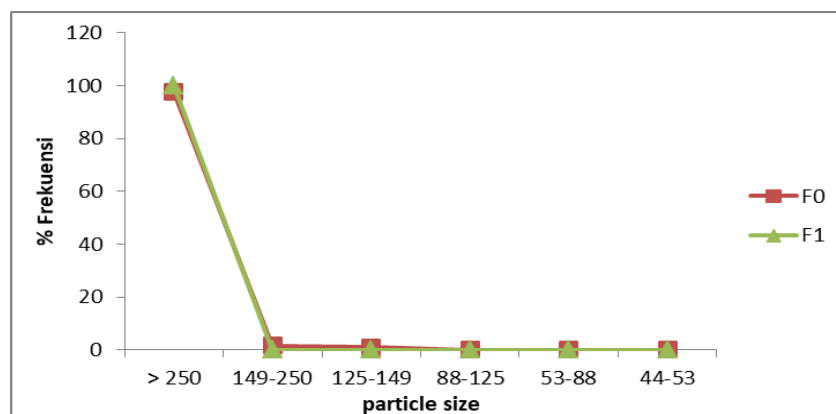

Figure 1. Particle frequency distribution curve

Based on the test of effervescent granules of African leaf extract (*Vernonia amygdalina* Del.) it does not meet the criteria for good granules because it only meets the requirements for testing the Haussner factor 1.2408 g/mL, porosity 34.6050%, particle size distribution >250, moisture content 4.0066 % ± 0.84, angle of repose, dispersion 4.2033 min ± 0.09 and pH 5.57 ± 0.36. Whereas in the granule carr's indexs 19.4126% is categorized as fair. This could be due to the consistency of the granules containing 10% extract which made the granules slightly moist, so it was necessary to add a lubricant such as Mg Stearate or talc to improve the flowability.

Table 4 : flow properties of of effervescent granules of African leaf extract (*Vernonia amygdalina* Del.)

No	Parameter	Result	
		FO	F1
1	Bulk density	0.4954	0.4857
2	tapped density	0.5559	0.6027
3	Carr's index	10.8832	19.4126
4	Hausner ratio	1.1221	1.2408
5	Angle of repose	20.4741 ± 1,36	14.5088° ± 0.55
6	dispersion	3.2133 min ± 0.17	4.2033 min ± 0.09
7	pH	7,1833 ± 0.15	5.57 ± 0.36

Determination of Total Flavonoid Content of Effervescent Granules from African Leaf Extract.

Analysis of flavonoid granules using UV-Vis double beam spectrophotometer (Shimadzu UV-1800) by measuring absorbance at a maximum wavelength of 415.5 nm. After measuring the calibration curve, the regression equation for the line $y = 0.04177x + 0.04304$ is obtained with a value of r = 0.99926. The results of the determination of flavonoid levels in effervescent granules of African leaves were obtained at 94.634% ± 1.83.

From the results of the assay, there was a slight decrease in the concentration of the extract but it was still in the high range because it was in the range of> 90%. This is because the effervescent granule formulation uses a long heating method in the granule drying process so that it more or less affects the flavonoid content in the granules and can also be influenced by the level of accuracy during the sample pipetting process.

CONCLUSION

Based on the formulation research and evaluation of effervescent granules from African leaf extract (*Vernonia amygdalina* Del.) that has been carried out, it can be concluded that:

1. African leaf extract (*Vernonia amygdalina* Del.) can be formulated in effervescent granule preparations.

2. The preparation of effervescent granules from African leaf extract (*Vernonia amygdalina* Del.) did not meet the criteria for a good granule because it only met the requirements for testing the haussner factor, porosity, particle size distribution, moisture content, angle of repose, dispersion and pH. Meanwhile, in the flowability test, the granules are difficult to flow and the compressibility test is categorized as quite good.

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