

## **Dry Suspension Formulation of Ethanol Extract from Wuluh Starfruit (*Averrhoa bilimbi* L.) with Arenga Gum as Suspending Agent**

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**Abstract.** Wuluh starfruit (*Averrhoa bilimbi* L.) has a sour taste and high-water content which makes the fruit easily damaged and has a short shelf life. It is necessary to process wuluh starfruit to obtain a product with a longer shelf life and tastes better without reducing its benefits. This study aimed to determine whether the wuluh starfruit ethanol extract can be formulated into a dry suspension dosage form with arenga gum as a suspension which at a certain concentration can be an optimal suspending agent. The results showed that the ethanol extract of wuluh starfruit (*Averrhoa bilimbi* L.) could be formulated into a dry suspension. The preparation with 0.5% arenga gum as a suspending agent is the best preparation which has a moisture content of 0.46%, stationary angle of 26.48°, flow time of 4.77 seconds, compressibility index of 18%, particle size distribution of 152.133µm, viscosity 106.00 cP, pH 4.16 and sedimentation volume 0.55. Data were analyzed using SPSS 26.0 using one-way analysis of variance (ANOVA) test and post hoc Multiple Comparisons Tukey HSD test.

Wuluh starfruit (*Averrhoa bilimbi* L.) has a sour taste and high-water content, which makes the fruit easily damaged and has a short shelf life. It is necessary to process wuluh starfruit to obtain a product with a longer shelf life and tastes better without reducing its benefits. The purpose of this study was to formulate the wuluh starfruit ethanol extract into a dry suspension dosage form using arenga gum as a suspending agent. The results showed that the ethanol extract of wuluh starfruit (*Averrhoa bilimbi* L.) could be formulated into a dry suspension. The preparation with 0.5% arenga gum as a suspending agent is the best preparation which has a moisture content of 0.46%, stationary angle of 26.48°, flow time of 4.77 seconds, compressibility index of 18%, particle size distribution of 152.133µm, viscosity 106.00 cP, pH 4.16 and sedimentation volume 0.55. Data were analyzed using SPSS 26.0 using one-way analysis of variance (ANOVA) test and post hoc Multiple Comparisons Tukey HSD test.

**Keywords:** Wuluh starfruit, Dry suspension, Suspending agent, Arenga gum, Galactomannan.

**Abstrak.** Buah belimbing wuluh (*Averrhoa bilimbi* L.) memiliki rasa yang asam dan kadar air yang tinggi sehingga membuat buah mudah rusak dan daya simpannya singkat. Diperlukan pengolahan terhadap buah belimbing wuluh agar diperoleh suatu produk yang memiliki masa simpan lebih lama dan rasa lebih enak tanpa mengurangi manfaatnya. Adapun penelitian ini bertujuan untuk mengetahui buah belimbing wuluh dalam bentuk ekstrak etanol dapat diformulasikan ke dalam sediaan suspensi kering dengan arenga gum yang pada konsentrasi tertentu dapat menjadi bahan pensuspensi yang optimal. Berdasarkan hasil penelitian menunjukkan bahwa ekstrak etanol buah belimbing wuluh (*Averrhoa bilimbi* L.) dapat diformulasikan ke dalam sediaan suspensi kering. Sediaan dengan bahan pensuspensi arenga gum 0,5% merupakan sediaan terbaik yang memiliki hasil kadar air

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0,46%, sudut baring 26,48°, waktu alir 4,77 detik, indeks kompresibilitas 18%, distribusi ukuran partikel 152,133  $\mu\text{m}$ , viskositas 106,00 cP, pH 4,16 dan volume sedimentasi 0,55. Data dianalisis dengan menggunakan SPSS 26.0 menggunakan uji one-way analysis of variance (ANOVA) dan uji post hoc Multiple Comparisons Tukey HSD.

**Kata Kunci:** Buah belimbing wuluh, Suspensi kering, Bahan pensuspensi, Arenga gum, Galaktomanan.

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## 1. Introduction

Wuluh starfruit empirically has often been used as a drug to lower blood glucose levels, hypertension, and lower blood cholesterol levels [1]. This fruit has a sour taste and a moisture content of  $\pm 93\%$ , which makes the fruit easily damaged and has a short shelf life (4-5 days) [2]. It is necessary to process the wuluh starfruit to obtain a product with a longer shelf life, and a better taste without reducing the benefits of the wuluh starfruit as well as the easiness to use, and the more accurate dosage [3]. In this study, it was made into a dry suspension dosage form with arenga gum as a suspending agent.

Dry suspension is a solid mixture that must be reconstituted with a suitable carrier before use [4]. Dry suspensions are preferred over solid forms such as tablets or capsules because the liquid is easier to swallow. One of the components in the manufacture of dry suspension is a suspending agent, which affects the physical stability of dry syrup that has been mixed with solvent [5]. *Suspending agents* are divided into several groups. One of them is a group polysaccharides consisting of gum arabic, tragacanth, sodium alginate, starch, carrageenan, xanthan gum, and guar gum. There is another example of a suspending agent of the polysaccharide group, namely arenga gum.

Arenga gum is a galactomannan obtained from sugar palm fruit. The use of sugar palm fruit as a pharmaceutical excipient is still very limited. The high carbohydrate content of sugar palm fruit allows its use as a suspending agent because it has the ability to form thick solutions in very low concentrations, and is only slightly influenced by pH, ionic and heating power [6].

## 2. Methods

This study used descriptive and experimental methods, using the wuluh starfruit (*Averrhoa bilimbi* L.) as the test material.

### 2.1 Materials

The materials used in this study were wuluh starfruit, sugar palm fruit, sodium alginate, sucrose, lactose, nipagin, and distilled water.

### 2.2 Preparation of Ethanol Extract

The extraction method used in this research is maceration method using ethanol pro analysis as the solvent. The preparation of ethanol extract was performed using 300 g of dry powder of wuluh starfruit put into a vessel. The dry powder was soaked into ethanol pro analysis, left for five days and protected from light while stirring frequently, and then re-macerated with ethanol pro analysis. The macerate was collected in a cup, leave it at room temperature and protected from light. Then the extract was left in the open air until a thick extract was obtained [7].

### **2.3 Arenga Gum Manufacturing Procedure**

1 kg of sugar palm fruit was cleaned of mucus using distilled water, then blended with 10 L of distilled water. After blending it was filtered and put in the refrigerator for 24 hours. Then the supernatant was mixed with ethanol 96% with a ratio of 1:2, and put back in the refrigerator for 24 hours. The obtained aggregates were squirted using a sterilized gauze. The aggregate was moistened with ethanol pro analysis and re-sheared. Gum is put in a desiccator. After the gum is dry, weigh the gum according to the desired weight [8].

### **2.4 Arenga Gum FTIR Test**

Fourier transform infrared (FTIR) spectroscopic analysis was used to characterize the structure of organic and inorganic compounds by analyzing the functional groups of each sample. This test used arenga gum samples. The samples were analyzed using the Shimadzu IR Prestige21 FTIR tool in the range of 4000-400  $\text{cm}^{-1}$  wave numbers with accumulated recordings of 40 scans and 4  $\text{cm}^{-1}$  resolution [9].

### **2.5 Dry Suspension Manufacturing Procedure**

Preparation of dry suspension granules made by wet granulation method. The suspending material, namely arenga gum, was developed with hot distilled water. After the mass expands, it is mixed with other ingredients, and crushed until a homogeneous mass is obtained. Then the mass was sieved through a 12-mesh sieve, then dried in a drying cabinet at 50°C for 3 hours. Sift again with a 14-mesh sieve, and then the granules obtained were weighed and recorded.

### **2.6 Physical Quality Inspection of Dry Suspension Granules Before Reconstitution**

In the dry suspension granules before being reconstituted, a physical quality inspection was carried out which included a moisture content test, stationary angle and flow time test, a specific gravity test, and a particle size distribution test.

### **2.7 Physical Quality Inspection of Dry Suspension Preparations After Reconstitution**

The dry suspension granules after being reconstituted were examined for physical quality including organoleptic, stability, viscosity, pH, and sedimentation volume.

### 3. Results and Analysis

#### 3.1 Ethanol Extract

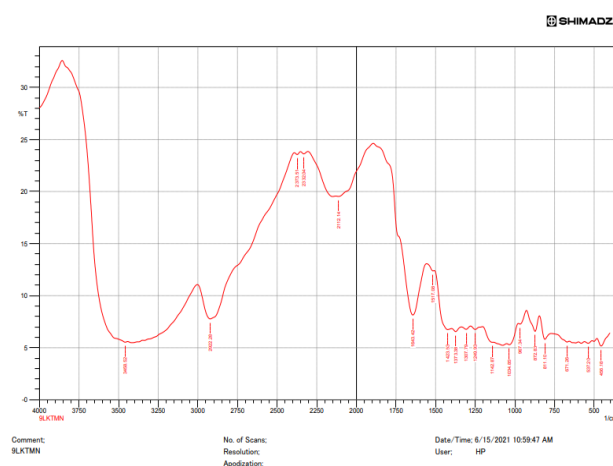
Based on the results of the extraction of 300 g of simplicia powder from the wuluh starfruit (*Averrhoa bilimbi* L.) by maceration method using ethanol pro analysis, it was obtained a thick blackish green extract of 61.60 g. The yield of ethanol extract of wuluh starfruit is 20.53%.

#### 3.2 Arenga Gum

The arenga gum produced in this study was 71.08 g or 7.108% of the initial material.

#### 3.3 Arenga Gum FTIR Test

The functional groups of gums were analyzed using infrared spectrophotometry at wave length of 4000-2000  $\text{cm}^{-1}$ , to confirm that the extracted content was a gum compound, its spectrum must be in the wave length number of 2000-500  $\text{cm}^{-1}$  [10]. The infrared spectrum of the extracted arenga gum can be seen in Figure 1.



**Figure 1** Arenga Gum Infrared Spectrum

Arenga gum produced had absorption peaks at a wave number of  $3458.52 \text{ cm}^{-1}$  indicating the presence of stretching vibrations of the O-H group. The peaks of  $811.10$  and  $872.83 \text{ cm}^{-1}$  corresponded to the presence of anomeric configurations (and conformers) and glycosidic bonds characterized by D-galactopyranose units and D-manopyranose units. The  $1034.85 \text{ cm}^{-1}$  wide band is a stretching vibration of C-O on the C-OH and C-O-C bonds (e.g. glycosidic). The peak of  $1142.87 \text{ cm}^{-1}$  is a bending vibration of C-O, indicating the presence of a pyranose ring. The wide band between  $2800\text{-}3000 \text{ cm}^{-1}$  and  $3100\text{-}3500 \text{ cm}^{-1}$  shows stretching vibrations of C-H and O-H. The peak at wave number  $1643.42 \text{ cm}^{-1}$  indicates the presence of bonds between polysaccharides and water, this peak is also a bending vibration (scissoring) of OH bonds [11].

#### 3.4 Physical Quality Inspection of Dry Suspension Granules Before Reconstitution

##### 3.4.1 Water content test

The water content test aims to see the water content contained in the dry suspension granules. The results of the inspection of the water content of the granules in the four formulas met the requirements for moisture content of 2-5% [12]. The results of the water content test on the granules can be seen in Table 1.

**Table 1** Moisture Content Test Results on Granules

Formula	Wet Granule Weight (g)	Dry Granule Weight (g)	Water Content (%)
F0	30	29.70	1
F1	30	29.76	0.80
F2	30	29.83	0.56
F3	30	29.86	0.46

### 3.4.2 Test of stationary angle and flow time

The stationary angle is the angle between the cone-shaped pile of particles and the horizontal plane. The smaller the stationary angle, the easier the granules flow. The stationary angle test is related to the flow time test which aims to see the flow properties of the granules and the effect on the uniformity of the dry suspension when it is put into the bottle. The flow property of dry suspension granules is classified as good if it forms an angle of 25°- 40° [13]. The results of the stationary angle test for dry suspension granules can be seen in Table 2.

**Table 2** Dry Suspension Granules Stationary Angle Test Results

Replication	Stationary Angle ( $\Phi$ )			
	F0	F1	F2	F3
1	36.50	36.38	25.46	26.56
2	37.91	38.65	25.64	26.34
3	33.20	34.46	26.78	26.56
Average	35.87	36.49	25.96	26.48

The data obtained from the dry suspension granule stationary angle test were then statistically processed using SPSS 26.0 with a 95% confidence level. The normality test shows that the distribution of the data is normal. The homogeneity test also showed a significance value of  $\alpha > 0.05$ , so that it could then use the one-way analysis of variance (ANOVA) test, followed by the post hoc Multiple Comparisons Tukey HSD test. The results of the SPSS post hoc Multiple Comparisons Tukey HSD analysis showed that there was no significant difference ( $p > 0.05$ ) between the stationary angle in each formula. It can be concluded that the stationary angle is F0 with the suspending material *na. alginate* as a comparison has yield angle results that are not significantly different from F1, F2, and F3 with *arenga gum* as suspending agent.

**Table 3** SPSS Analysis Results of Dry Suspension Granules Stationary Angle Test

Tukey HSD

REPLICATION	N	Subset for alpha = 0.05	
		1	
R3	4		30.2500
R1	4		31.2250

R2	4	32.1350
Sig.		.895

The results of the fourth flow time test have different average flow times, namely 3.71 seconds for formulation 0, 5.07 seconds for formulation 1, 6.48 seconds for formulation 2, and 4.77 seconds for formulation 3. These results shows that the four formulas meet the requirements where the resultant flow time is 10 seconds [12].

Table 4 Flow Rate of the Produced Dry Suspension Granule Formulas

Replication	Flow Rate (seconds)			
	F0	F1	F2	F3
1	3.68	5.05	6.54	4.83
2	3.75	5.10	6.46	4.72
3	3.70	5.08	6.45	4.76
Average	3.71	5.07	6.48	4.77

The data from the dry suspension granule flow test results obtained were then statistically processed using SPSS 26.0 with a 95% confidence level. The normality test shows that the distribution of the data is normal. The homogeneity test also showed a significance value of  $\alpha > 0.05$ , so that it could then use the one-way analysis of variance (ANOVA) test, followed by the post hoc Multiple Comparisons Tukey HSD test. The results of the SPSS post hoc Multiple Comparisons Tukey HSD analysis showed that there was no significant difference ( $p > 0.05$ ) between the flow times in each formula. It can be concluded that the flow time of F0 with the suspending agent na. alginate as a comparison has a flow time that is not significantly different from F1, F2, and F3 with arenga gum as a suspending agent.

Table 5 SPSS Analysis Results of Dry Suspension Granule Flow Time Test

#### Tukey HSD

REPLICATION	N	Subset for alpha = 0.05	
		1	
R3	4		4.9975
R2	4		5.0075
R1	4		5.0250
Sig.			.999

#### 3.4.3 Specific gravity test

The measurement of compressive density, real density and compressibility index aims to provide information about the amount of air that enters during the manufacture of a preparation. Based on the results of the specific gravity test, it shows that of the four formulas that meet the requirements of the compressibility index, namely F1, F2, and F3 with a compressibility index requirement of  $< 20\%$  [12].

Table 6 Dry Suspension Granule Type Weight Test Results

Formula	Replication	Real Specific Gravity (g/mL)	Specific Gravity	Compressibility Index (%)
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		Compressed (g/mL)		
F0	1	0.38	0.52	26.92
	2	0.38	0.52	26.92
	3	0.38	0.50	24.00
Average		0.38	0.51	25.94
F1	1	0.45	0.52	13.46
	2	0.45	0.52	13.46
	3	0.45	0.52	13.46
Average		0.45	0.52	13.46
F2	1	0.40	0.47	14.89
	2	0.40	0.47	14.89
	3	0.41	0.47	12.76
Average		0.40	0.47	14.18
F3	1	0.41	0.50	18.00
	2	0.41	0.50	18.00
	3	0.40	0.50	20.00
Average		0.41	0.50	18.66

The results of the compressibility index data on the dry suspension granule density test obtained were then statistically processed using SPSS 26.0 with a 95% confidence level. The normality test shows that the distribution of the data is normal. The homogeneity test also showed a significance value of  $\alpha > 0.05$ , so it could then use the one-way analysis of variance (ANOVA) test, followed by the post hoc Multiple Comparison Tukey HSD test. The results of the SPSS analysis of the dry suspension granule density test can be seen in Table 7.

**Table 7** Statistical Analysis of Dry Suspension Granule Compressibility Index

Tukey HSD

FORMULA	N	Subset for alpha = 0.05		
		1	2	3
F1	3	13.4600		
F2	3	14.0800		
F3	3		18.6667	
F0	3			25.9467
Sig.		.925	1.000	1.000

As shown in Table 7, it can be concluded that the compressibility index of dry suspension granules with 0.3% arenga gum as suspending agent (F1) is significantly different from dry suspension granules with 0.5% arenga gum as suspending agent (F3) and dry suspension granules with Na suspension. alginate (F0), while the dry suspension granules with 0.3% arenga gum as suspending agent (F1) and dry suspension granules with 0.4% arenga gum as suspending agent (F2) were not significantly different.

### 3.4.4 Particle size distribution

The particle size of a dry suspension is of different importance than that of a liquid suspension. In liquid suspension, the particle size greatly affects the stability of the product. In contrast, in dry suspension, it affects the smoothness of the filling process into the bottle which can be shown by the flow rate, besides that it also affects the homogenization process of the product when water is added for use. The results of the particle size distribution test were carried out on formula 3 dry suspension of the ethanol extract of wuluh starfruit (*Averrhoa bilimbi* L.) with 0.5% arenga gum as a suspending agent, presented in graphical form which can be seen in Figure 2. After analysis, the particle size data of the suspension of the dry matter has a normal distribution. The particle radius has an average of 0.00760665 cm or diameter of 152.133  $\mu\text{m}$ . Based on these results indicate that the particle size of the dry suspension has a larger particle size than those required for a liquid suspension of 0.1-100  $\mu\text{m}$  [4], these conditions are desirable so that the flow rate of particles into the container can take place smoothly. When water is added to the dry suspension, it is possible that its particle size is smaller than the particle size requirements of the conventional suspension.

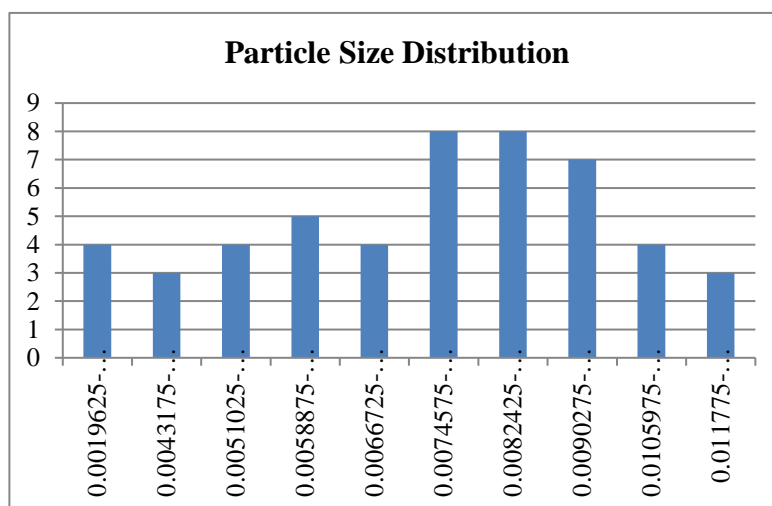


Figure 2 Particle Size Distribution

### 3.5 Physical Quality Check Dry Suspension Preparations After Reconstitution



### 3.5.1 Organoleptic test

The data obtained from the assessment sheet was tabulated and the preference value for each preparation was determined by finding the average result for each panelist at a 95% confidence level. The data on the organoleptic test values of the preparations can be seen in Table 8.

**Table 8 The characteristics of the formulas**

No	Formulation	Preferred Value Interval		
		Color	Flavor	Scent
1	F0	-	-	-
2	F1	2.80-3.45	1.92-2.48	2.57-3.29
3	F2	3.39-4.21	2.15-2.65	3.08-3.72
4	F3	4.03-4.49	2.08-2.78	3.75-4.37

Based on Table 8 above, it shows that the preparations that are preferred by volunteers based on color, taste, and odour are F3 preparations.

1. The color parameter has a preference value interval of 4.03-4.49. For writing the final favorite value, the smallest value is taken, which is 4.03 and is rounded up to 4 (preferred).
2. The taste parameter has a preference value interval of 2.08-2.78. For writing the final value of liking, the smallest value is taken, namely 2.08 and rounded up to 2 (disliked).
3. The aroma parameter has an interval of preference value from 3.75 to 4.37. For writing the final favorite value, the smallest value is taken, namely 3.75 and rounded up to 4 (preferred).

### 3.5.2. Stability test

The suspension stability test was carried out after being reconstituted using distilled water. The suspension test was carried out by accelerated storage using the cycling test method, where the suspension was stored at two extreme temperatures, namely at 4°C in the refrigerator and 40°C in the heating cabinet alternately for 24 hours each for 6 cycles. Based on the results of the stability test showed that there was a change in the four suspension formulations in the sixth cycle.

### 3.5.3. Viscosity test

Viscosity test is carried out in order to find out how much consistency the preparation is and shows the viscosity of a preparation. Viscosity that is too high is not expected because it can cause problems with pouring the suspension from the container and make it difficult for the preparation to be re-dispersed. Viscosity that is too low can interfere with the homogeneity of the unstable mixture so that it will interfere with the number of doses used. In accordance with the viscosity results, the results showed that there was a very significant difference in viscosity between the four dry suspension formulas. Viscosity test showed that formula 0 used the suspending agent sodium alginate has a viscosity of 999.00 centipoise. Viscosity test on formula 1, 2, and 3 with arenga gum as a suspending agent showed that formula 3 had the highest viscosity compared to other

formulas, which was 106.00 centipoise. This is due to the difference in the amount of arenga gum used. Viscosity values that meet the requirements are 37-396 centipoise [14].

**Table 9** Viscosity Test Results

Condition	Replication	Viscosity			
		F0	F1	F2	F3
Before	1	999,00	48.00	54.00	106.00
	2	998.00	47,00	55.00	106.00
	3	999,00	46.00	55.00	105.00
Average		998.66	47,00	54.66	105.66
After	1	980.00	32.00	51.00	103.00
	2	980.00	32.00	50.00	103.00
	3	980.00	31.00	51.00	101.00
Average		980.00	31.66	50.66	102.33

The viscosity data obtained were then statistically analyzed using SPSS 26.0 with a 95% confidence level. The normality test shows that the distribution of the data is normal. The homogeneity test also showed a significance value of  $\alpha > 0.05$ , so it could then use the one-way analysis of variance (ANOVA) test, followed by the post hoc Multiple Comparisons Tukey HSD test. The results of the SPSS analysis of the viscosity test before the stability test can be seen in Table 10 and the results of the SPSS analysis of the viscosity test after the stability test can be seen in Table 11.

**Table 10** SPSS Viscosity Test Analysis Results Before Stability Test

Tukey HSD

FORMULA	N	Subset for alpha = 0.05			
		1	2	3	4
F1	3	47,0000			
F2	3		54.6667		
F3	3			105.6667	
F0	3				998,6667
Sig.		1,000	1,000	1,000	1,000

**Table 11** SPSS Viscosity Test Analysis Results After Stability Test

Tukey HSD

FORMULA	N	Subset for alpha = 0.05			
		1	2	3	4
F1	3	31.6667			
F2	3		50.6667		
F3	3			102.3333	
F0	3				980,000
Sig.		1,000	1,000	1,000	1,000

Based on Table 10 and Table 11, it can be concluded that the viscosity before and after the stability test was carried out by the suspension cycling test method with the suspending agent Na. alginate (F0) was significantly different from the suspension with arenga gum as a suspending agent 0.3% (F1), 0.4% (F2), and 0.5% (F3). The preparation of F3 suspension with 0.5% arenga gum as a suspending agent gave better viscosity results.

### 3.5.4. The pH of the dry suspension produced

The pH test aims to determine the pH of the reconstituted dry suspension preparation. The pH test with a pH meter showed that none of the formulas met the requirements, the optimum suspension pH was 5-6 [14]. This may be caused by the pH extract used was too low. The results of pH test of the preparation can be seen in Table 12.

**Table 12** Preparation pH Test Results

Condition	Replication	pH			
		F0	F1	F2	F3
Before	1	4.3	3.1	3.3	4.2
	2	4.2	3.1	3.3	4.2
	3	4.2	3.1	3.3	4.1
Average		4.23	3.1	3.3	4.16
After	1	4.3	3.0	3.3	4.1
	2	4.2	3.0	3.2	4.0
	3	4.2	2.9	3.2	3.9
Average		4.23	2.96	3.23	4.0

The data obtained from the pH test results were then statistically analyzed using SPSS 26.0 with a 95% confidence level. It was found that the distribution of the data was normal. The homogeneity test also showed a significance value of  $\alpha > 0.05$ , so it could then use the one-way analysis of variance (ANOVA) test, followed by the post hoc Multiple Comparison Tukey HSD test. The results of the SPSS post hoc Multiple Comparison Tukey HSD analysis showed that there was no significant difference ( $p > 0.05$ ) between the pH of the preparation before and after the stability test was carried out with the cycling test method. It can be concluded that the four suspension formulas before and after storage with extreme temperatures at a certain time were not significantly different.

### 3.5.5. Sedimentation volume test

Sedimentation volume aims to provide information about the suspension system formed. The sedimentation volume is the ratio of the final volume of the sediment to the initial volume of the suspension before settling. From the results of the study, it was found that the sedimentation volume of the four dry suspension formulas met the sedimentation volume requirements, namely  $< 1$  to  $> 1$  [15]. The results of the sedimentation volume test can be seen in Table 13.

**Table 13** Sedimentation Volume Test Results

Formula	Total Suspension Volume (ml)		Suspension Volume Total (mL)	Volume Sedimentation
	-	Sediment Volume (mL)		
F0	5	10	0.5	
F1	8	10	0.8	
F2	7	10	0.7	
F3	5.5	10	0.55	

#### 4. Conclusion

The present study proved that wuluh starfruit (*Averrhoa bilimbi* L.) in the form of ethanol extract can be formulated into a dry suspension with arenga gum as a suspending agent. By physical stability, the best dry suspension was F3 with 0.5% arenga gum as a suspending agent with a water content of 0.46%, stationary angle 26.48°, flow time 4.77 seconds, real specific gravity 0.41 g/mL, compressed specific gravity 0.50 g/mL, compressibility index 18%, particle size distribution 152.133 m, viscosity 106.00 centipoise, pH 4.16, and sedimentation volume 0.55.

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