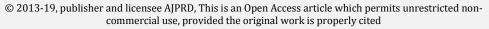
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Research Article

Formulation and Evaluation of Moringa Seed Oil Nanoemulsion Gel

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ABSTRACT

Objective: The objective of this study was to formulate and evaluate the moringa seed oil (MSO) nanoemulsion gel using high-energy emulsification method.

Methods: Nanoemulsion gel formulated by high-energy emulsification method using the comparison of surfactant (tween 80) and cosurfactant (sorbitol) concentration with the variation of moringa seed oil concentration. Evaluation of the stability of the nanoemulsion gel preparation includes centrifugation test, viscosity, pH, organoleptic observation (odor, color, clarity, and phase separation), and particle size measurement during 12 weeks storage at room temperature.

Results: The results showed that all nanoemulsion gel preparations are transparent yellow, characteristic odor, type weights 1.0888–1.1193 g/ml, and stable for 12 weeks storage at room temperature. The smallest particle size produced by the nanoemulsion gel preparation in a formula of the concentration of 5%, which 52.25 nm.

Conclusions: Moringa seed oil can be formulated as a nanoemulsion gel by high energy emulsification method. MSO with a 5% concentration was very stable for 12 weeks storage.

Keywords: Moringa seed oil,nanoemulsion gel, high energy emulsification method, Surfactant, Cosurfactant

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INTRODUCTION

oringa oleifera (Moringaceae) is known as kelor, benzolive, marango, drumstick tree, and sajna. Bioactive compounds such as carotene, vitamin E, vitamin C, vitamin B, vitamin A, phenolics, caortenoids etc, have been reported¹. Fatty oil obtained from the seed kernels of Moringa oleifera is yellowish brown, semi-solid, with a faint odour of bitter almonds. The % composition of mixed fatty acids in seed oil as Palmitic-11.04, Stearic-3.58, Arachidic-3.44, Behenic-7.09, Palmitoleic-2.38, Linoleic-1.83% respectively 17,18. The antioxidant compounds contained in the oil can be used to prevent damage caused by degradation. Antioxidant compounds in the body serve as an antidote to free radicals, so the body is protected from various degenerative diseases and slow the aging process².

A nanoemulsion is considered to be a thermodynamically or kinetically stable liquid dispersion of two immiscible liquid phases such as an oil phase and a water phase. An interfacial tension exists between the two liquids everywhere they are in contact due to differences in attractive interactions between the molecules of the two liquid phases. Amphiphilic surfaceactive molecules or surfactants are added to reduce this interfacial tension³. Droplet size of nanoemulsion falls typically in the range of 50 nm to 500 nm and shows a narrow size distribution⁴. Nanoemulsions can be formed with various formulations such as cream, gel, spray, and foam. Nanoemulsions can also increase absorption bioavailability of drugs, help to stabilize the hydriphobic active substances, and have the efficiency on rapid penetration of some drugs⁵. Nanoemulsion gel has emerged as one of the most interesting topical delivery system as it

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has dual release control system is hydrogel and nanoemulsion. Gel Formulation provides better application property and stability in comparison of ointment and cream. In spite of many advantage of gels a major limitation is in the delivery of hydrophobic drug. So to overcome this limitation an emulsion based approach is being used to that even a hydrophobic moiety can enjoy the unique property of gel⁶.

MATERIAL AND METHODS

Material

Moringa seed oil (MSO) was received as gift sample from House of Alchemy (Medan, Indonesia), Tween 80, sorbitol, carbopol, TEA and distilled water. All other chemicals were of analytical grade.

Apparatus and conditions

Analytical balance (Ohrus), Magnetic Stirrer (Thermo Scientific CIMAREC), Sonicator (Branson), ViscometerNDJ

8-s, pH Meter (Hanna Instrument), centrifuge (Hitachi CF 16 R X II), and Particle Size Analyzer (FRITSCH, Laser Partickle Size Analysette 22).

Formulation of nanoemulsion gel

Formulation of nanoemulsion gel was usedhigh energy emulsification method. MSO was added to the oil phase which has added a mix solution which was a mixture of surfactant and cosurfactant, then stirred with the magnetic stirrer until homogeneous. distilled water was added by means of titration, stirred continuously until nanoemulsion was formed which was marked by the formation of a translucent solution. The percentage of the ingredients of nanoemulsion gel formula (Table 1) was modified from the nanoemulsion gel formula which was carried out in the previous study by Sari, et al⁷. Physicochemical evaluation of MSO nanoemulsion gel Organoleptic test Observations on any changes of color, odor, clarity, and phase separation were made.

Table 1. Formulas of Moringa seed oil nanoemulsion gel

Composition of Gel	Formula (%w/w)					
Karbopol 940	0.5	1 5				
TEA	1	7 6				
distilled water ad	100	20				
Composition of Nanoemulsion	Formula 4(% w/w)	Formula 5(%w/w)	Formula 6(%w/w)			
Moringa Seed Oil	5	10	15			
Tween 80	36	36	36			
Sorbitol	24	24	24			
distilled waterad	100 and De	100	100			

Physicochemical evaluation of MSO nanoemulsion gel

Organoleptic test

Observations on any changes of color, odor, clarity, and phase separation were made.

pH measurement

pH values were measured at 25°C using a digital pH meter. Three measurements were taken for one sample. Before the readings were observed, pH meter was calibrated using pH 7.01, 4.01, and 10.01

Viscosity

The viscosity of nanoemulsion gel was measured using the NDG-8s viscometer at room temperature (25°C±2°C). Viscosity measurements used two spindles speed and experiments were conducted three times⁷.

Nanoemulsion gel particle size measurement

Particle size was measured using FRITSCH, *Laser Particle Size Analysette 22*.

Centrifugation test

The centrifugation test was performed at the beginning and after the preparation was made by measuring one time. The nanoemulsion gel preparation was inserted into centrifugation tube then centrifuged at 3750 rpm for 5 h^8

RESULT AND DISCUSSION

Organoleptic test

Based on data from the result of observation and stability test for 12 weeks indicated that the nanoemulsion gel has a good stability for 12 weeks. Stability of a prepared pharmacy can be seen by any change of color, it's smelt and phase seperation. The characteristics of MSO nanoemulsion gel can be seen in Fig.1 and Table 2

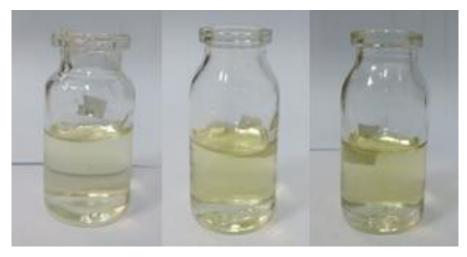


Figure. 1: MSO nanoemulsion gel after 12 weeks storage

Table 2: The effect of storage on MSO nanoemulsion gel

Weeks		Organoleptic													
	Cole	Color			Odor		Clarity		Creaming			Phase seperation			
	F_1	F ₂	F ₃	F ₁	F ₂	F ₃	Fi	F_2	F_3	F ₁	F ₂	F ₃	F ₁	F ₂	F_3
0	Y	Y	Y	S	S	S	T	Т	T		-	-	-	-	-
1	Y	Y	Y	S	S	S	Т	T	Т	0	-	-	-	-	-
2	Y	Y	Y	S	S	S	Т	Т	T	13	-	-	-	-	-
3	Y	Y	Y	S	S	S	T	Т	Т	- 0	-	-	-	-	-
4	Y	Y	Y	S	S	S	T	T	T	7	/-	-	-	-	-
5	Y	Y	Y	S	S	S	Т	Т	Т	-3	-	-	-	-	-
6	Y	Y	Y	S	S	S	T	Т	Т	5-/	-	-	-	-	-
7	Y	Y	Y	S	S	S	TD	eT16	Т	-	-	-	-	-	-
8	Y	Y	Y	S	S	S	Т	T	T	-	-	-	-	-	-
9	Y	Y	Y	S	S	S	Т	T	T	-	-	-	-	-	-
10	Y	Y	Y	S	S	S	Т	Т	Т	-	-	-	-	-	-
11	Y	Y	Y	S	S	S	Т	Т	Т	-	-	-	-	-	-
12	Y	Y	Y	S	S	S	Т	Т	Т	-	-	-	-	-	-

*Y: Yellow, S: Specific, T: Translucent, MSO: Moringa seed oil

Fig. 1 and Table 2 shows that the nanoemulsion gel stored at room temperature remains clear for up to 12 weeks, the color and smell are unchanged and no any creaming. The formation of creaming in the preparation was due to the formation of aggregates from the inner phase which has a greater tendency to rise to the surface. According to Sinko⁹, creaming is the emulsion separation into two layers, wherein the one layer contains drip grains (the dispersed phase) more than the other layers. If the dispersed phase density is smaller than the continuous phase, the sedimentation velocity

becomes negative. In this nanoemulsion gel preparation, there were no any coarse grains from various concentrations of MSO.

pH measurement

Determination of pH value from MSO nanoemulsion gel was using digital pH meter for 12 weeks. Table 3 showed the results of pH value from three formulas for 12 weeks at room temperature.

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Formula Time (week) 0 2 3 4 5 6 7 8 9 10 11 12 F1 6.96 6.96 6.93 6.86 6.76 6.66 6.60 6.46 6.46 6.33 6.23 5.90 5.90 F2 7.00 6.96 6.30 6.93 6.93 6.76 6.73 6.60 6.53 6.43 6.10 6.86 6.10 F3 7.03 7.00 7.00 6.93 6.93 6.90 6.83 6.73 6.56 6.53 6.36 6.20 6.13

Table 3: The effect of storage onpH value of MSO nanoemulsion gel

Viscosity test

Determination of the viscosity of the nanoemulsion gelwas performed using a NDG 8-s viscometer with the corresponding spindle number at room temperature for 12 weeks. Data of viscosity test result and graph of nanoemulsiongel viscosity change can be seen in Fig. 2.

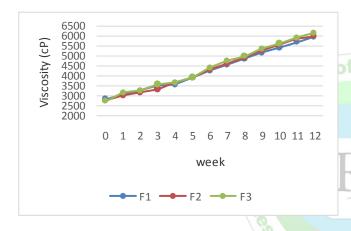


Figure 2: The effect of storage on viscosity of MSO nanoemulsion gel

Based on the viscosity test results in Fig. 2, it was concluded that the higher concentration of moringa seed oil, the viscosity will increase and the longer the storage time, the viscosity will increase. The viscosity of the nanoemulsiongel preparation was carried out at room temperature for 12 weeks in which the room temperature was a low temperature. This suggests that the lower the storage temperature will increase the viscosity of the nanoemulsion gel preparation while storage at room temperature also results in an increase in nanoemulsiongel viscosity. This is consistent with the theory that the storage period will increase the viscosity of the preparation. However, the increase is not so significant.

Nanoemulsiongel particle size measurement

The results of particle size analyzingare shown in Table 4. Based on the centrifugation test results in Fig.3 All formulas are stable, and there is no phase separation, which means that all formulas are stable against the gravitational force

Table 4: The effect of storage on particle size analyzing

Formula	Distribution of particle size (nm)								
	0 week (nm)	6 week (nm)	12 week (nm)						
F1	52.25	129.90	240.73						
F2	65.11	278.96	339.06						
F3	74.09	357.49	416.08						

Determination of particle size is done in the 1st week and week 12. The particle measurement results show that each formula has varying sizes, this is due to the difficulty of homogenizing two different systems, in addition to several other factors, such as duration or speed of stirring, also cause the formula is not homogeneous. Particle sizes from the 5 to 12 weeks increased, because of tween 80 experienced hydrolysis thus reducing the effectiveness of the nanoemulsion globule interface film layer 10. Higher oil contens, lower emulsion viscosity especially, the continuous phase and higher power input during emulsification will increase collision frequency and coalescence frequency. At a higher coalescence frequency, particle size increase rapidly 11.

Centrifugation test

The data of the nanoemulsion gel centrifugation test results can be seen in Fig. 3.



Figure 3: The effect of Centrifugation on MSO nanoemulsion gel

experienced for 1 year. The centrifugation test describes the stability of the dosage because the effect of Earth's gravity equivalent to 1 year⁸. After testing on all three formulas, F1-

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F3 showed no any separation. This shows that these three formulas are stable for 1 year because of the influence of gravity.

CONCLUSIONS

MSO is suitable for preparation of nanoemulsion gel and it gives an o/w emulsion. Best formula consisting with MSO 5%. As this is the preformulation study stage and further studies will include the need for incorporation of the drug to

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