FORMULATION AND EVALUATION OF NANOEMULSION OF BASIL LEAF EXTRACT (OCIMUM BASILICUM L.) WITH TWEEN 80 CONCENTRATION VARIATIONS

anonymous marking enabled

Submission date: 11-Apr-2023 05:14PM (UTC+1000)
Submission ID: 2061375206
File name: ULATION_AND_EVALUATION_OF_NANOEMULSION_OF_BASIL_LEAF_EXTRACT.pdf (1.6M)
Word count: 6510
Character count: 35008

FORMULATION AND EVALUATION OF NANOEMULSION OF BASIL LEAF EXTRACT (*OCIMUM BASILICUM* L.) WITH TWEEN 80 CONCENTRATION VARIATIONS

Redhita, Loviya Ayu,[1,*] Beandrade, Maya Uzia[2], Anindita, Reza[2], Putri, Intan Kurnia[2]

1. Student, Department of Pharmacy, University, STIKes Mitra Keluarga, Jl. Pengasinan Rawa Semut, Margahayu, Bekasi, Indonesia.
2. Lecturer, Department of Pharmacy, STIKes Mitra Keluarga, Jl. Pengasinan Rawa Semut, Margahayu, Bekasi, Indonesia.

*Correspondence: Maya Uzia Beandrade | STIKes Mitra Keluarga | email: maya.uzia@stikesmitrakeluarga.ac.id

Abstract

Introduction: A nanoemulsion is a transparent emulsion system consisting of a mixture of oil and water and surfactant molecules to stabilize it. The nanoemulsion particle size ranged from 10-200 nm. The content of alkaloids, flavonoids, tannins, saponins and eugenol in basil leaves makes them useful as antibacterials. The purpose of this study was to evaluate the physical stability of the basil leaf extract nanoemulsion formulation using the surfactant tween 80 with concentrations of 36%, 37% and 38%.

Method: This study used an experimental design to determine the physical stability of nanoemulsion preparations by making three different concentrations of tween 80 (36%, 37% and 38%) and evaluating organoleptic tests, pH, viscosity, transmittance, particle determination (particle size and polydispersity index) and stability (centrifugation and freeze thawing).

Results: The results of the research on nanoemulsion preparations of basil leaf extract F1, F2 and F3 had average particle sizes of 14,43 nm, 14,73 nm and 15 nm with polydispersity indexes of 0,38, 0,42 and 0,37. The three formulas are brown, clear, have a distinctive smell of basil leaves, and have a transmittance value of above 90%. The results of the centrifugation test on all formulas did not occur in phase separation, and in the freeze thawing test, the results obtained did not change the color, odor, or phase separation. The preparation remained stable during the storage period of 14 days.

Conclusion: Basil leaf extract nanoemulsion with a concentration of 36% in F1 is the best nanoemulsion with clear characteristics of light brown color, characteristic odor of basil leaves, has a pH value of 6.24, viscosity 1333,33-1416,67 cP, transmittance 96,7%, particle size of 14,43 nm and a polydispersity index of 0,38. After centrifugation and freeze-thaw tests, it doesn't change color, smell, or separate into phases.

Keywords: Basil leaves, Nanoemulsion, Tween 80

INTRODUCTION

Emulsions generally degrade as energy levels increase and with time. Reducing the particle size is one of the methods used to solve the problem of emulsions. Nanoemulsions can be used to reduce size (Daud et al., 2017). A transparent emulsion system, consisting of a mixture of oil and water and surfactant molecules to stabilize it (Adi et al., 2019). The particle size of the nanoemulsion ranges from 10–200 nm (Ayuningtias et al., 2017). Nanoemulsions can be used to deliver drugs to the skin. Large surface area, low surface tension, and increased penetration can be achieved by utilizing the interfacial tension of the o/w type emulsion (Nurpermatasari and Ernoviya, 2020). Creaming, flocculation, coalescence and precipitation can be prevented by the presence of nanoemulsions, so that nanoemulsions are more stable than ordinary emulsions (Hanifah and Jufri, 2018). Nanoemulsion technology in Indonesia is currently being developed because it can increase skin permeability for drug penetration (Ayunin, 2017). Oil, water, surfactants and cosurfactants, which are the basic ingredients of nanoemulsions, can be formulated using other ingredients such as plants to enhance the effect of nanoemulsions. Basil is one of the plants that can be used as a mixture of nanoemulsion formulations containing secondary metabolites such as tannins, flavonoids, essential oils and saponins (Utami et al., 2021). Azkawati (2016) explained that basil contains alkaloids, tannins, flavonoids, saponins and eugenol compounds that can be used as antibacterials. Basil (*Ocimum basilicum* L.) leaves have the potential to have antibacterial effects on *Salmonella*, *Escherichia coli*, *Campylobacter jejuni* and *Clostridium aerogenes* (Rubab et al., 2017). Extracts from basil leaves have biological activity that can be used as a deodorant, antidepressant, antipyretic, antidiabetic, antihyperglycemic, as well as antibacterial (Wahid et al., 2020). According to Kindangen et al. (2018), basil (*Ocimum basilicum* L.) leaves have antibacterial properties. The antibacterial results of the ethanolic extract of basil leaves contained in the anti-acne gel had an inhibition zone diameter of 9,7 mm, 14,4 mm and 19,1 mm, which had the ability to inhibit *Staphylococcus aureus* bacteria at concentrations of 0.5%, 1%, and 1,5%. Arianto and Cindy (2019) explained in their research that variations of tween 80 and sorbitol obtained stability during the nanoemulsion experiment within 12 weeks of storage at room temperature, low and high. Kristiani et al. (2019) conducted research on *Ocimum basilicum* L. nanoemulsion with essential oil where at a concentration of 2,5% Salmonella typhi bacteria could be inhibited and obtained nanoemulsion stability with surfactant, namely tween 80 and co-surfactant PEG 400. However, research on nanoemulsion with basil leaf extract (*Ocimum basilicum* L.) using various concentrations of surfactant tween 80 and 36%, 37% and 38% has never been done. Sorbitol was used in this study as a cosurfactant. Based on this, the researchers wanted to formulate and test the stability of the nanoemulsion extract of *Ocimum basilicum* L. by making three variations of the concentration of tween 80 (36%, 37% and 38%), which aim to determine the physical stability of the formulation in the presence of different concentrations of tween 80.

METHOD

A. Research Design

The design of this research is experimental and uses the descriptive analysis method.

B. Research Location and Time

This research was conducted at the Pharmaceutical Technology Laboratory of STIKes Mitra Keluarga in February–March 2022.

C. Population and Sample

The population and sample in this study were basil leaf extract (*Ocimum basilicum* L.) which were obtained from a supplier Herbal Palapa Muda Perkasa located in Kalimulya, Depok City.

D. Procedure

1. Plant Determination

This examination aims to determine whether the basil used is a species of *Ocimum basilicum* (L.). The basil plant samples were determined at the Herbarium Bogoriense, Botanical Division of the BRIN Cibinong Biological Research Center.

2. Extraction By Maceration Method

The maceration method was used to extract 1.500 grams of simplicia powder at room temperature. Maceration with 5 liters of 96 percent ethanol was completed in 3x24 hours. The extract was filtered through filter paper every 24 hours until the filtrate was obtained (Yamlean, 2017). At about 40°C, the filtrate was evaporated with a rotary evaporator to separate the solvent from the solute and get a thick basil leaf extract (Ahmadita, 2017; Ariani et al., 2020).

3. Basil Leaf Extract Phytochemical Screening

a. Alkaloid test

50 mg of the extract was dissolved with a few mL of HCL and then filtered. To the filtrate, 1-2 drops of Mayer, Wagner and Dragendorff reagents were added in different test tubes. The Mayer reagent will be characterized by the presence of a white or yellowish precipitate in the tube. The Wagner reagent will be marked by the presence of a brown or blackish red precipitate in the tube. The Dragendorff reagent will be marked by the presence of an orange precipitate (Kumalasari and Andiarna, 2020).

b. Flavonoid Test

A total of 40 mg of extract was added with 100 mL of hot water. Then boil for 5 minutes, then filtered. The filtrate was measured as much as 5 mL and then 0,05 mg of Mg powder and 1 mL of concentrated HCL were added. If it gives a brown or yellow color after vigorous shaking, it indicates flavonoid positive (Wowor et al., 2022).

c. Tannin Test

A total of 0,1 grams of extract added 10 mL of distilled water. Then filtered, the filtrate added with reagent iron (III) chloride ($FeCl_3$) 1% 5 mL. If it causes a dark blue or green-black color, the identification results show a positive tannin (Kumalasari and Andiarna, 2020).

d. Phenolic Test

A total of 2 mL of the sample was added 3-4 drops of $FeCl_3$. If a green, red, purple, blue or solid black color is formed, the identification indicates a positive phenol (Wowor et al., 2022).

4. Formulation of Basil Leaf Extract Nanoemulsion

The nanoemulsion was made using an aqueous phase consisting of propyl paraben and methyl paraben dissolved in distilled water, and then the aqueous phase was manually stirred with a stirring rod. The solution was then heated on a hot plate, cooled and filtered (Mixture a). Furthermore, the basil leaf extract was dissolved with some sorbitol and some other sorbitol was used to dissolve sunflower oil. The extract that had been dissolved with sorbitol was put into a glass beaker containing sunflower oil that had been dissolved in sorbitol (Mixture b). Put the mixture into a glass beaker containing tween 80, then stir using a mixer. Put mixture b into a glass beaker containing tween 80 and mixture a, then stir using a mixer at 450 rpm for 15 minutes. After thoroughly mixing, the preparation is placed in a sonicator for 30 minutes (Arianto and Cindy, 2019). Table 1 shows the formula for basil leaf extract nanoemulsion.

Table 1. Nanoemulsion Preparation Formula (Arianto and Cindy, 2019)

Material Composition (%)	Formula			Function
	I	II	III	
Basil Leaf Extract	0,5	0,5	0,5	Antibacterial
Sunflower Oil	5	5	5	Oil Phase
Tween 80	36	37	38	Surfactant
Sorbitol	22	22	22	Cosurfactant
Methyl Paraben	0,1	0,1	0,1	Preservative
Propyl Paraben	0,02	0,02	0,02	Preservative

5. Characteristics and Evaluation of Nanoemulsion Preparations

a. Organoleptic Test

Visual observations included color, odor and phase separation of nanoemulsion preparations at room temperature (25°C) for 14 days of storage (Arianto and Cindy, 2019).

b. pH Test

Measurement of the pH of the preparation was carried out using a pH meter. The pH of the nanoemulsion preparation must be in accordance with the pH of the skin, which is 4,5–6,5. A number of nanoemulsions are inserted into the pH meter. Observations were made at room temperature (25°C) for 14 days of storage (Arianto and Cindy, 2019).

c. Viscosity Test

Viscosity testing using a Brookfield LV-801 viscometer at room temperature (25°C) for 14 days. A total of 100 mL of the nanoemulsion preparation was measured, then put on spindle number 4. Place the preparation under the spindle that has been installed. The spindle is lowered to the limit of the spindle submerged in the preparation. The speed of the viscometer is set at the correct rpm. The measurement results will be shown on the screen of the Brookfield viscometer (Zulfa et al., 2019). The viscosity value of a good semisolid preparation is between 500 and 5000 cP (Kusuma *et al.*, 2017).

d. Transmittance Test

The transmittance percentage was measured at 650 nm with a UV-Vis spectrophotometer. It was carried out at the beginning of the observation after the preparation was made by dissolving 100 mL of the basil leaf extract nanoemulsion in 5 mL of distilled water (Huda and Wahyuningsih, 2018). The nanoemulsion preparation looks clear and transparent, as shown by the fact that 90–100% of light can pass through it (Indalifiany et al., 2021).

e. Nanoemulsion Particle Determination Test

The determination of particles from nanoemulsion preparations was measured by a particle size analyzer (Horiba Scientific, Nanoparticle Analyzer SZ-100), which includes particle size and polydispersity index. Testing was conducted at the beginning of the observation after the preparation was made by means of 100 mL of nanoemulsion sample dissolved in 50 mL of distilled water (Maharini et al., 2018). The nanoemulsion particle size ranges from 10–200 nm (Ayuningtias et al., 2017). The polydispersity index value 0,5, including monodispersion, indicates uniform droplet size (Adi et al., 2019).

f. Physical Stability Test

1) Centrifugation Test

The nanoemulsion preparation was put into Eppendorf as much as 1,5 mL, then centrifuged for 30 minutes at a speed of 3000–4000 rpm for 30 minutes at the beginning of the observation after the preparation was made (Winarti et al., 2016). The centrifugation test makes sure that the nanoemulsion preparation is stable and that the phases don't separate (Arianto and Cindy, 2019).

2) Freeze Thawing Test

This test was carried out by storing the nanoemulsion preparation for 24 hours at a cold temperature (4°±2°C), then moving the preparation to room temperature (25°±2°C) for 24 hours (1 cycle). A total of 6 cycles were run for testing. The freeze thawing requirement is stable during storage; there is no change in color, odor or phase separation (Rismarika et al., 2020).

RESULTS

A. The Result Of Plant Determination

The determination test was carried out on March 4, 2022 at the Herbarium Bogoriense, Botany Division of the BRIN Cibinong Biology Research Center. The results of the determination test obtained are correct if the basil plant has the Latin name *Ocimum basilicum* (L.) with the *Lamiaceae* tribe.

B. Basil Leaf Extraction Results

The results of the basil leaf extract are blackish brown, have a distinctive aroma of basil leaves and have a thick texture.

C. Basil Leaf Phytochemical Screening Results

The results of phytochemical screening of basil leaf extract were positive for containing alkaloids with Mayer and Wagner reagents, flavonoids, tannins and phenolics. Meanwhile, with Dragendorff's reagent the results obtained were negative for alkaloids.

D. Results Characteristics and Evaluation of Nanoemulsion Preparations

1. Results of Organoleptic Observations of Nanoemulsion

Tables 2, 3 and 4 show the organoleptic results of the basil leaf extract nanoemulsion having different colors in the three formulas. All formulas have a distinctive odor of basil leaves and no separation occurs.

Table 2. Tween 80 Formulation I Organoleptic Test Results 36%

Replication	Days to-	Color	Odor	Phase Separation
Rep 1	0	Clear, Light brown	Typical basil leaves	Not separating
	7	Clear, Light brown	Typical basil leaves	Not separating
	14	Clear, Light brown	Typical basil leaves	Not separating
Rep 2	0	Clear, Light brown	Typical basil leaves	Not separating
	7	Clear, Light brown	Typical basil leaves	Not separating
	14	Clear, Light brown	Typical basil leaves	Not separating
Rep 3	0	Clear, Light brown	Typical basil leaves	Not separating
	7	Clear, Light brown	Typical basil leaves	Not separating
	14	Clear, Light brown	Typical basil leaves	Not separating

Table 3. Tween 80 Formulation II Organoleptic Test Results 37%

Replication	Days to-	Color	Odor	Phase Separation
Rep 1	0	Clear, slightly dark brown	Typical basil leaves	Not separating
	7	Clear, slightly dark brown	Typical basil leaves	Not separating
	14	Clear, slightly dark brown	Typical basil leaves	Not separating
Rep 2	0	Clear, slightly dark brown	Typical basil leaves	Not separating
	7	Clear, slightly dark brown	Typical basil leaves	Not separating
	14	Clear, slightly dark brown	Typical basil leaves	Not separating
Rep 3	0	Clear, slightly dark brown	Typical basil leaves	Not separating
	7	Clear, slightly dark brown	Typical basil leaves	Not separating
	14	Clear, slightly dark brown	Typical basil leaves	Not separating

Table 4. Tween 80 Formulation III Organoleptic Test Results 38%

Replication	Days to-	Color	Odor	Phase Separation
Rep 1	0	Clear, Dark brown	Typical basil leaves	Not separating
	7	Clear, Dark brown	Typical basil leaves	Not separating
	14	Clear, Dark brown	Typical basil leaves	Not separating
Rep 2	0	Clear, Dark brown	Typical basil leaves	Not separating
	7	Clear, Dark brown	Typical basil leaves	Not separating
	14	Clear, Dark brown	Typical basil leaves	Not separating
Rep 3	0	Clear, Dark brown	Typical basil leaves	Not separating
	7	Clear, Dark brown	Typical basil leaves	Not separating
	14	Clear, Dark brown	Typical basil leaves	Not separating

2. Nanoemulsion pH Test Results

Table 5 shows the results of the nanoemulsion pH test of the basil leaf extract, which was stable and met the skin pH range of 4,5–6,5.

Table 5. pH Test Results

Days to-	pH		
	F1 Tween 80 36%	F2 Tween 80 37%	F3 Tween 80 38%
	Average ± SD	Average ± SD	Average ± SD
0	6,24 ± 0,02	6,36 ± 0,02	6,41 ± 0,02
7	6,24 ± 0,02	6,36 ± 0,02	6,41 ± 0,02
14	6,24 ± 0,02	6,36 ± 0,02	6,41 ± 0,02

3. Nanoemulsion Viscosity Test Results

Table 6 shows the results of the nanoemulsion test results of stable basil leaf extract and meets the viscosity requirement range of 500-5000 cP.

Table 6. Viscosity Test Results

Days to-	Viscosity (cP)		
	F1 Tween 80 36%	F2 Tween 80 37%	F3 Tween 80 38%
	Average ± SD	Average ± SD	Average ± SD
0	1416,67 ± 520,42	2833,33 ± 144,34	3000 ± 250
7	1336,33 ± 386,79	2166,67 ± 144,34	2166,67 ± 144,34
14	1333,33 ± 381,88	1916,67 ± 144,34	2166,67 ± 144,34

4. Nanoemulsion Transmittance Test Result

Table 7 shows the transmittance test results of the basil leaf extract nanoemulsion that met the transmittance requirement range of 90-100%.

Table 7. Transmittance Test Results

Transmittance (%)		
F1 Tween 80 36%	F2 Tween 80 37%	F3 Tween 80 38%
Average ± SD	Average ± SD	Average ± SD
96,7 ± 0,005	97,9 ± 0,003	91,7 ± 0,02

5. Nanoemulsion Particle Determination Test Result

Table 8 shows the particle size test results for basil leaf extract nanoemulsions that meet the requirements for a particle size of 10–100 nm.

Table 8. Particle Size Test Results

Particle Size (nm)		
F1 Tween 80 36%	F2 Tween 80 37%	F3 Tween 80 38%
Average ± SD	Average ± SD	Average ± SD
14,43 ± 0,21	14,73 ± 0,06	15 ± 0,12

Table 9 shows the results of the nanoemulsion polydispersity index test of basil leaf extract meeting the range of requirements for the polydispersity index <0.5.

Table 9. Polidispersity Index Test Results

Polydispersity Index		
F1	F2	F3
Tween 80 36%	Tween 80 37%	Tween 80 38%
Average ± SD	Average ± SD	Average ± SD
0,38 ± 0,04	0,42 ± 0,06	0,37 ± 0,01

6. Physical Stability Test Results of Basil Leaf Nanoemulsion

The stability of basil leaf nanoemulsion was tested using centrifugation and freeze-thawing. The results of the centrifugation test of basil leaf extract nanoemulsion using surfactant tween 80 with concentrations of 36%, 37 and 38% with 3 replications met the requirements for centrifugation, namely no separation. Then the results of the freeze thawing test using surfactant tween 80 with concentrations of 36%, 37% and 38% with 3 replications for 6 cycles met the freeze thawing requirements, namely no change in color, odor and phase separation.

DISCUSSION

The manufacture of basil leaf extract is done using maceration method, using 96% ethanol as solvent. The extract obtained was blackish brown in color, had a distinctive aroma of basil leaves and had a thick texture. The results obtained in accordance with the research of Risnayanti and Dalimunthe (2022) are dark brown-black in color, have a distinctive aroma of basil leaves and have a thick extract texture. The extraction of 1.500 g of dried simplicia of basil leaves resulted in a thick extract of 105,4 g of basil leaves with an extract yield of 7,03%. The yield obtained is in accordance with the requirements of the Indonesian Herbal Pharmacopoeia (2017), which is not less than 5,6%.

The results of the phytochemical test of alkaloids with Mayer's reagent on samples of basil leaf extract (*Ocimum basilicum* L.) were positive for alkaloids, these results were in accordance with previous research conducted by Warsi and Sholichah (2017) when the sample was dripped with Mayer reagent, a white precipitate was formed. In the alkaloid test using Wagner's reagent on a sample of basil leaf extract (*Ocimum basilicum* L.) the results obtained were positive for containing alkaloids, these results were in accordance with previous research conducted by Kumalasari and Andiarna (2020) which formed a blackish red precipitate. Testing of alkaloids with Dragendorff's reagent on samples of basil leaf extract (*Ocimum basilicum* L.) showed negative results, namely the formation of an orange precipitate, where the results were the same as the research conducted by Harianja et al. (2020) negative result no orange precipitate formed. Identification of flavonoids in samples of basil leaf extract (*Ocimum basilicum* L.) positive for flavonoids, these results are in accordance with previous studies conducted by Harianja *et al*. (2020) formed an orange-red and brown solution. Identification of tannins in samples of basil leaf extract (*Ocimum basilicum* L.) positive for tannins, these results are in accordance with previous research conducted by Kumalasari and Andiarna (2020) which formed a blackish green color. The results of the phenol phytochemical test on the basil leaf extract sample (*Ocimum basilicum* L.) were positive for containing phenol, these results were in accordance with previous research conducted by Harianja *et al*. (2020) a greenish black solution was formed.

There are three formula modifications in this study, namely at a concentration of tween 80, consisting of F1 (tween 80, 36%), F2 (tween 80, 37%) and F3 (tween 80, 38%). Each formula was carried out three replications to see the physical stability of the nanoemulsion preparation. There are several test evaluations to see the physical stability of nanoemulsion preparations modified with tween 80 concentration, namely organoleptic tests, pH, viscosity, transmittance, particle determination and stability (centrifugation and freeze thawing tests). In this study, changing the amount of Tween 80 didn't change how stable the nanoemulsion was physically. Because of this, these three formulas were part of a stable and good formulation because they met the test evaluation requirements during storage.

In this study, organoleptically, there were slight differences in color observations, namely formula 1 tween 80 36% was clear light brown, formula 2 tween 80 37% was clear, slightly dark brown and formula 3 tween 80 38% was clear dark brown. The use of basil leaf extract and variations of tween 80 as a surfactant have an effect on the color of the nanoemulsion preparations, where the higher the tween 80 used, the darker the color of the resulting preparation. Meanwhile, the observation of odor and phase separation gave the same results, which had a characteristic odor of basil leaves and no separation occurred.

The pH value of the preparation was determined by using the pH test on the three formulas. Because these topical preparations will be applied to the skin, it is important to consider how safe these preparations are when used (Zulfa, 2020). The pH of the topical preparation is between 4,5-6,5 according to the pH range of the skin. Skin irritation can occur if the pH of the preparation is too acidic and scaly skin can occur if the pH is too alkaline caused by damage to the acid mantle in the stratum corneum (Rahmaniyah, 2018). Based on the test results, the pH value of the nanoemulsion preparations ranged from 6,24-6,41 so that the three formulas were included in the safe skin pH range, which was between 4,5-6,5 for topical use. The pH measurement results show that the higher the concentration of tween 80 used, the higher the pH of the nanoemulsion preparation obtained, namely F1 (tween 80 36%) an average of 6,24, F2 (tween 80 37%) an average of 6,36 and F3 (tween 80 38%) an average of 6,41. This is possible because tween 80 has a pH of 6-8, so the higher the concentration of tween 80, the higher the pH value of the preparation (Handayani et al., 2018). After 14 days, the pH of the three formulas did not change. This shows that the preparation was stable.

Viscosity testing on nanoemulsion preparations was carried out to determine the viscosity. The release of the active ingredient from the preparation is influenced by the viscosity of the preparation (Hidayati, 2020). The viscosity value of semisolid preparations for topical use ranges from 500-5000 cP (Kusuma et al., 2017). The viscosity test on the basil leaf extract nanoemulsion was measured 3 times for 14 days, namely days 0, 7 and 14. On day 0 of the three formulas, the viscosity values ranged from 1416,67 - 3000 cP. On the 7th day of the three formulas the viscosity values ranged from 1336,33 – 2166,67 cP. On the 14th day of the three formulas, the viscosity values ranged from 1336,33 – 2166,67 cP. The three formulas fall into the required range between 500-5000 cP. The results of the viscosity measurement showed that the higher the concentration of tween 80 used, the higher the viscosity of the nanoemulsion preparation. This increase in viscosity is due to the effect of surfactant concentration in the preparation, which induces flocculation between the dispersed particles, increasing the viscosity of the nanoemulsion preparation (Hakim et al., 2018). The viscosity values of the three formulas showed a decrease during storage, but remained within the required range. Improper temperature conditions and storage procedures can cause a decrease in viscosity. This means that the surface tension of the preparation has gone down, which could make the preparation less stable (Mardikasari et al., 2016).

The clarity of the nanoemulsion formulations was assessed using the transmittance test. The percentage of transmittance to ensure the purity of the nanoemulsion preparations as a result of visual observation using distilled water as a comparison. The transmittance value shows the comparison between the light intensity that appears after interacting with the test substance and the light intensity that appears before engaging the test substance (Daud et al., 2017). The preparation has a clear and transparent appearance, as evidenced by the transmittance percentage of 90-100% (Indalifiany et al., 2021). The results of the measurement of the transmittance of the nanoemulsion preparations obtained are F1 (tween 80 36%) an average of 96,7%, F2 (tween 80 37%) an average of 97,9% and F3 (tween 80 38%) an average of 91,7%. The three formulas have transmittance values that are not much different and fall into the required range between 90-100%. The formation of nanoemulsions is indicated by a clear preparation, therefore the higher the clarity value, the better the nanoemulsion preparation (Daud et al., 2017).

The particle determination test was carried out at the beginning of the observation after the preparation was made, namely day 0 of the three formulas. The results of the nanoemulsion preparation particle size obtained are F1 (tween 80 36%) an average of 14,43 nm, F2 (tween 80 37%) an average of 14,73 nm and F3 (tween 80 38%) an average of 15 nm. The particle sizes of the three formulas were in accordance with the requirements ranging from 10-200 nm (Ayuningtias et al., 2017). The smaller the particle size the better for drug absorption and release (Winarti et al., 2016). The measurement results showed that the formulations of formulas 1, 2 and 3 had different sizes but were not significant. This is due to the difficulty of uniforming the two different systems, as well as other parameters such as duration and speed of stirring (Nurpermatasari and Ernoviya, 2020). The particle size of the nanoemulsion is also affected by the surfactant concentration. Surfactants act as emulsifiers, reducing the amount of free energy required to make nanoemulsions by lowering the interfacial tension in the oil-in-water system. The surfactants produce a film layer on the droplet surface in an o/w nanoemulsion system consisting of a nonionic surfactant. Droplets will not enter the dispersion medium because of the film layer (Maharini et al., 2018). The nanoemulsion preparation has an average particle size of about 5-200 nm, so the nanoemulsion preparation has high kinetic stability (Mardikasari et al., 2016). Nanoemulsion preparations with a particle size of less than 90 nm have been shown to be more stable against sedimentation (Pratiwi et al., 2018).

The polydispersity index is a measurement of the molecular mass distribution of the sample (Handalis, 2018). The polydispersity index value shows information about the physical stability of the dispersion system and the uniformity of droplet size (Zulfa, 2020). All formulas that have a polydispersity index value <0,5 indicate that the resulting globules vary. The range of polydispersity index values that are well accepted is 0 (monodispersive particles) to 0,5 (large size distribution) (Adi et al., 2019). The low polyspersity index value indicates that the resulting dispersion system is more stable over time (Maharini et al., 2018). The results of the polyspersity index of nanoemulsion preparations obtained are F1 (tween 80 36%) an average of 0,38, F2 (tween 80 37%) an average of 0,42 and F3 (tween 80 38%) an average of 0,37. The polypersity index value of the three formulas yielded a value of less than 0,5 which indicates that the level of particle size uniformity and will be stable in long-term storage (Maharini et al., 2018).

To determine the mechanical stability of the preparation, a centrifugation test was carried out. The centrifugation test on the basil leaf extract nanoemulsion was carried out at the beginning of the observation after the preparation was made, namely day 0 for each formula. Centrifugation testing can describe the stability of nanoemulsion preparations under the influence of the earth's gravity for a period of one year (Hakim et al., 2018). The purpose of the centrifugation test is to see whether the nanoemulsion preparation is stable by observing the phase separation after centrifugation. This test is needed to determine the impact of product transportation shocks on the physical appearance of a product (Daud et al., 2017). The results obtained from this test, the three formulas remained stable and there was no phase separation. Indicates that the preparation is stable for one year against the force of gravity.

A freeze thawing test was used to see whether the nanoemulsion preparation changes after storage and temperature differences. The results obtained in this test after 6 cycles were consistent. There was no difference between before and after the test. The three formulas remained stable and did not change organoleptically in terms of color, odor and phase separation. This is due to the presence of tween 80 in the preparation, which helps to keep the nanoemulsion preparation stable (Aulia, 2017).

CONCLUSION

From the research that has been done, it can be concluded:

1. Basil leaf extract (*Ocimum basilicum* L.) can be formulated into nanoemulsion preparations with variations in tween 80 concentrations of 36%, 37% and 38%. The three formulas are included in the requirements for nanoemulsion preparations, namely organoleptic, pH, viscosity, transmittance, particle determination and stability. And has good physical stability during the storage period of 14 days.
2. Basil leaf extract nanoemulsion (*Ocimum basilicum* L.) with a concentration of 36% in F1 is the best nanoemulsion with clear characteristics of light brown color, characteristic odor of basil leaves, has a pH value of 6,24, viscosity 1333,33 – 1416,67 cP, transmittance 96,7%, particle size of 14,43 nm and polydispersity index of 0,38. It remains stable without changing color, odor or phase separation after centrifugation and freeze thawing tests.

ACKNOWLEDGMENTS

The author would like to thank STIKes Mitra Keluarga Bekasi for the support during the author's research.

REFERENCES

Adi, A. C., Setiawaty, N., Anindya, A., & Rachmawati, H. (2019). Formulasi dan Karakterisasi Sediaan Nanoemulsi Vitamin A. *Media Gizi Indonesia*, *14*(1), 1–13. https://doi.org/10.20473/mgi.v14i1.1-13

Ahmadita, A. N. F. (2017). *Formulasi Losion Ekstrak Etanol 70% Herba Kemangi (Ocimum americanum L.) Menggunakan Asam Stearat Sebagai Emulgator*. Skripsi. Program Studi S1 Farmasi. Universitas Islam Negri Syarif Hidayatullah. Jakarta.

Ariani, N., Febrianti, D. R., & Niah, R. (2020). Uji Aktivitas Ekstrak Etanolik Daun Kemangi (Ocimum sanctum L.) terhadap Staphylococcus aureus secara In Vitro. *Jurnal Pharmascience*, *7*(1), 107–115. https://doi.org/10.20527/jps.v7i1.8080

Arianto, A., & Cindy, C. (2019). Preparation and Evaluation of Sunflower Oil Nanoemulsion as a Sunscreen. *Herbal Medicine in Pharmaceutical and Clinical Sciences*, *7*(22), 3757–3761. https://doi.org/10.3889/oamjms.2019.497

Aulia, Y. (2017). *Pengaruh Variasi Konsentrasi Tween 80 Dan Sorbitol Terhadap Aktivitas Antioksidan Minyak Alpukat (Avocado oil) Dalam Formulasi Nanoemulsi*. Skripsi. Program Studi S1 Farmasi. Universitas Sumatera Utara. Medan.

Ayunin, K. (2017). *Formulasi Dan Uji Pelepasan Meloxicam Dalam Sistem Nanoemulsi Menggunakan Kombinasi Fase Minyak Palm Oil Dan Virgin Coconut Oil*. Skripsi. Fakultas Kedokteran dan Ilmu-Ilmu Kesehatan. Islam Negeri Maulana Malik Ibrahim. Malang.

Ayuningtias, D. D. R., Nurahmanto, D., & Rosyidi, V. A. (2017). Optimasi komposisi polietilen glikol dan lesitin sebagai kombinasi surfaktan pada sediaan nanoemulsi kafein. *E-Jurnal Pustaka Kesehatan*, *5*(1), 157–163.

Azkawati, E. (2016). *Aktivitas Antibakteri Ekstrak Daun Kemangi (Ocimum basillicum) Terhadap Methicilin Resistant Staphylococcus aureus*. Abstrak. Repositori Riset Kesehatan Nasional. https://doi.org/https://r2kn.litbang.kemkes.go.id/handle/123456789/25971

Daud, N. S., Musdalipah, M., & Lamadari, A. (2017). Formulasi Nanoemulsi Aspirin Menggunakan Etanol 96 % Sebagai Ko-Surfaktan. *Warta Farmasi*, *6*(1), 1–11. https://doi.org/10.46356/wfarmasi.v6i1.66

Departemen Kesehatan Republik Indonesia (2017). *Farmakope Herbal Indonesia Edisi Kedua*. Jakarta: Departemen Kesehatan Republik Indonesia.
https://doi.org/10.1201/b12934-13

Hakim, N. A., Arianto, A., & Bangun, H. (2018). Formulasi dan Evaluasi Nanoemulsi dari Extra Virgin Olive Oil (Minyak Zaitun Ekstra Murni) sebagai Anti-Aging. *Talenta Conference Series: Tropical Medicine (TM)*, *1*(2), 391–397. https://doi.org/10.32734/tm.v1i2.222

Handalis, B. B. N. (2018). *Preparasi Dan Karakterisasi Nanopartikel Emas Ekstrak Daun Singkong Gajah (Manihot Esculenta Crantz.) Dengan Proses Biosintesis High Energy*. Skripsi. Program Studi S1 Farmasi. Universitas Islam Indonesia. Yogyakarta.

Handayani, F. S., Nugroho, B. H., & Munawiroh, S. Z. (2018). Optimization of Low Energy Nanoemulsion of Grape Seed Oil Formulation Using D-Optimal Mixture Design (DMD). *Jurnal Ilmiah Farmasi*, *14*(1), 17–34. http://journal.uii.ac.id/index.php/JIF

Hanifah, M., & Jufri, M. (2018). Formulation and Stability Testing Of Nanoemulsion Lotion Containing *Centella asiatica* Extract. *Journal of Young Pharmacists*, *10*(4), 404–408. https://doi.org/10.5530/jyp.2018.10.89

Harianja, M., Rahman, H., & Wigati, S. (2020). Invitro: Evaluasi Aktifitas Peluruhan Batu Ginjal Ekstrak Daun Kemangi (*Ocimum basilicum*) Menggunakan Spektrofotometer Serapan Atom Invitro: *Jurnal Sains Dan Kesehatan*, *3*(3), 451–457.

Hidayati, S. R. (2020). *Formulasi Dan Uji Stabilitas Nanoemulsi Ekstrak Buah Parijoto (Medinilla speciosa Blume)*. Skripsi. Program Studi S1 Farmasi. Universitas Ngudi Waluyo. Ungaran.

Huda, N., & Wahyuningsih, I. (2018). Karakterisasi Self-Nanoemulsifying Drug Delivery System (SNEDDS) Minyak Buah Merah (*Pandanus conoideus* Lam.). *Jurnal Farmasi Dan Ilmu Kefarmasian Indonesia*, *3*(2), 49. https://doi.org/10.20473/jfiki.v3i22016.49-57

Indalifiany, A., Malaka, M. H., Sahidin, Fristiohady, A., & Andriani, R. (2021). Formulasi Dan Uji Stabilitas Fisik Nanoemulgel Formulation And Physical Stability Test Of Nanoemulgel Containing Petrosia Sp . *Jurnal Farmasi Sains Dan Praktis (JFSP)*, 112–123.

Kindangen, O. C., Yamlean, P. V. Y., & Wewengkang, D. S. (2018). Formulasi Gel Antijerawat Ekstrak Etanol Daun Kemangi (*Ocimum basilicum* L.) Dan Uji Aktivitasnya Terhadap Bakteri *Staphylococcus aureus* Secara In Vitro. *Pharmacon*, *7*(3), 283–293.

Kristiani, M., Ramayani, S. L., Yunita, K., & Saputri, M. (2019). Formulasi dan Uji Aktivitas Nanoemulsi Minyak Atsiri Daun Kemangi (*Ocimum basilicum* L.) Terhadap *Salmonella typhii*. *Jurnal Farmasi Indonesia*, *16*(1), 14–23.

Kumalasari, M. Li. F., & Andiarna, F. (2020). Uji Fitokimia Ekstrak Etanol Daun Kemangi. *Indonesian*

Journal for Health Sciences, *4*(1), 39–44.

Kusuma, S., Mita, S., & Mutiara, A. (2017). Antimicrobial lotion containing red Piper betle leaf (Piper crocatum Ruiz and Pav) ethanolic extract for topical application. *National Journal of Physiology, Pharmacy and Pharmacology*, *8*(130–138), 1.

Maharini, I., Utami, D. ., & Fitrianingsih, F. (2018). The Influence of Tween 80 in the Formulations of Nanoemulsion Virgin Coconut Oil. *International Conference on Pharmaceutical Research and Practice*, 75–78.

Mardikasari, S. A., Jufri, M., & Djajadisastra, J. (2016). Formulation and in vitro penetration study of topical dosage form of nanoemulsion from genistein of *Sophora japonica* Linn. *J Ilmu Kefarmasian Indonesia*, *14*, 190–198.

Nurpermatasari, A., & Ernoviya, E. (2020). Formulasi dan Evaluasi Nanoemulsi Ketokonazole. *Jurnal Dunia Farmasi*, *4*(3), 138–148. https://doi.org/10.33085/jdf.v4i3.4698

Pratiwi, L., Fudholi, A., Martien, R., & Pramono, S. (2018). Uji Stabilitas Fisik dan Kimia Sediaan SNEDDS (Self-nanoemulsifying Drug Delivery System) dan Nanoemulsi Fraksi Etil Asetat Kulit Manggis (Garcinia mangostana L.). *Traditional Medicine Journal*, *23*(2), 84–90.

Rahmaniyah, D. N. K. (2018). *Perbandingan Formula Sistem Nanoemulsi dan Nanoemulsi Gel Hidrokortison Dengan Variasi Konsentrasi Fase Minyak Palm Oil*. Skripsi. Program Studi S1 Farmasi. Universitas Islam Negeri Maulana Malik Ibrahim. Malang.

Rismarika, Maharini, I., & Yusnelti. (2020). Pengaruh konsentrasi PEG 400 sebagai kosurfaktan pada formulasi nanoemulsi minyak kepayang. *Chempublish Journal*, *5*(1), 1–14. https://doi.org/10.22437/chp.v5i1.7604

Risnayanti, & Dalimunthe, G. I. (2022). Formulasi Foot Spray Ekstrak Daun Kemangi (*Ocimum africanum* L.) Sebagai Penghilang Bau Kaki Serta Uji Aktivitas Antibakteri. *Jurnal Farmasi, Sains, Dan Kesehatan*, 1(2), 115–123.

Rubab, S., Hussain, I., Ali, B., Ayaz, K., Unar, A., Abbas, K. A., & Khichi, Z. H. (2017). Biomedical Description of *Ocimum basilicum* L. *JIIMC*, *12*(1), 59–67.

Sugiyono. (2013). *Metode Penelitian Kuantitatif, Kualitatif, dan R&D* (19th ed.). Bandung: Alfabeta.

Utami, P. W., Syaflida, R., & Siregar, I. B. (2021). Pengaruh ekstrak daun kemangi (*Ocimum basilicum* L) terhadap *Staphylococcus aureus* di rongga mulut. *Jurnal Kedokteran Gigi Universitas Padjadjaran*, *33*(1), 38–43. https://doi.org/10.24198/jkg.v32i1.29968

Wahid, A. R., Ittiqo, D. H., Qiyaam, N., Hati, M. P., Fitriana, Y., Amalia, A., & Anggraini, A. (2020). Pemanfaatan Daun Kemangi (*Ocinum sanctum*) Sebagai Produk Antiseptik Untuk Preventif Penyakit Di Desa Batujai Kabupaten Lombok Tengah. *Jurnal Pengabdian Masyarakat Berkemajuan*, *4*(1), 500–503. https://doi.org/10.31764/jpmb.v4i1.2841

Warsi, & Sholichah, A. R. (2017). Phytochemical screening and antioxidant activity of ethanolic extract and ethyl acetate fraction from basil leaf (*Ocimum basilicum* L.) by DPPH radical scavenging method. *IOP Conference Series: Materials Science and Engineering*, *259*(1). https://doi.org/10.1088/1757-899X/259/1/012008

Winarti, L., Martien, R., Suwaldi, & Hakim, L. (2016). An experimental design of SNEDDS template loaded with bovine serum albumin and optimization using D-optimal. *International Journal of Pharmaceutical and Clinical Research*, *8*(5), 425–432.

Wowor, M. G. G., Tampara, J., & Saogo, S. P. (2022). Skrining Fitokimia dan Uji Antibakteri Masker Peel-Off Ekstrak Etanol Daun Kalu Burung (*Barleria prionitis* L.) Phytochemical Screening and Antibacterial Test of Peel-Off Mask with Ethanol Extracts of Kalu Burung Leaves (*Barleria prionitis* L .). *Jurnal Ilmiah Sains*, *22*(1), 75–86.

Yamlean, P. V. Y. (2017). Formulasi Dan Uji Antibakteri Sediaan Sabun Cair Ekstrak Daun Kemangi (*Ocymum Basilicum* L.) Terhadap Bakteri *Staphylococcus Aureus*. *Pharmacon*, *6*(1), 76–86. https://doi.org/10.35799/pha.6.2017.19731

Zulfa, A. (2020). *Formulasi Dan Evaluasi Sediaan Nanoemulsi Topikal Minyak Atsiri Sereh Wangi (Cymbopogon Nardus L.) Yang Berpotensi Sebagai Antiaging*. Skripsi. Program Studi S1 Farmasi.

Universitas Islam Indonesia. Yogyakarta.

Zulfa, E., Novianto, D., & Setiawan, D. (2019). Formulasi Nanoemulsi Natrium Diklofenak dengan Variasi Kombinasi Tween 80 Dan Span 80 : Kajian Karakteristik Fisik Sediaan. *Media Farmasi Indonesia*, *14*(1), 1471–1477.

FORMULATION AND EVALUATION OF NANOEMULSION OF BASIL LEAF EXTRACT (OCIMUM BASILICUM L.) WITH TWEEN 80 CONCENTRATION VARIATIONS

ORIGINALITY REPORT

24%	13%	13%	10%
SIMILARITY INDEX	INTERNET SOURCES	PUBLICATIONS	STUDENT PAPERS

PRIMARY SOURCES

1	jmk.stikesmitrakeluarga.ac.id Internet Source	3%
2	vdocuments.net Internet Source	1%
3	journal.uad.ac.id Internet Source	1%
4	journal.unimma.ac.id Internet Source	1%
5	oamjms.eu Internet Source	1%
6	media.neliti.com Internet Source	1%
7	Ronaldo Panggabean, Nofita, Ade Maria Ulfa. "The Hepatoprotective Effects of Basil Leaf (Ocimum sanctum L.) Extract on Paracetamol Induced Liver Damage in Male Rat", Biomedical Journal of Indonesia, 2021 Publication	1%

8	china.iopscience.iop.org Internet Source	<1 %
9	Ilma Nur Khafifa, Ayu Shabrina, M Fatchur Rochman. "STABILITY AND SUNSCREEN ACTIVITY OF NUTMEG SEED OIL EMULGEL WITH CARBOPOL 940 VARIATION AS GEL BASE", Jurnal Farmasi Sains dan Praktis, 2022 Publication	<1 %
10	Shara Sasmita, Indah Anggraini Yusanti, Sofian Sofian. "The effectiveness of basil extract on the mildew saprolegnia within in vitro", JPBIO (Jurnal Pendidikan Biologi), 2021 Publication	<1 %
11	eudl.eu Internet Source	<1 %
12	www.researchgate.net Internet Source	<1 %
13	repository.radenintan.ac.id Internet Source	<1 %
14	link.springer.com Internet Source	<1 %
15	www.iosrjournals.org Internet Source	<1 %
16	Theresia Setiyani, Dewi Klarita Furtuna, Fatma Ria. "Pengaruh Ekstrak Daun Kemangi (*Ocimum basilicum* L.) Terhadap Pertumbuhan	<1 %

Streptococcus pneumoniae Secara *In Vitro*", Herb-Medicine Journal: Terbitan Berkala Ilmiah Herbal, Kedokteran dan Kesehatan, 2023
Publication

17	Submitted to Universitas Jenderal Soedirman Student Paper	<1 %
18	e-journal.uajy.ac.id Internet Source	<1 %
19	A H Kusumawati, A Munawaroh, L Fikayuniar. "Formulation and Physical Evaluation of Body Lotion Preparation of Kacip Fatimah (labisia pumila) Ethanolic Extracts as Antioxcidant", IOP Conference Series: Materials Science and Engineering, 2021 Publication	<1 %
20	Submitted to Universitas Sam Ratulangi Student Paper	<1 %
21	fmipa.unsrat.ac.id Internet Source	<1 %
22	"Influence of Leadership and Academic Culture on Academic Quality Assurance in Nursing Vocational Schools in the Era of Industrial Revolution 4.0", International Journal of Recent Technology and Engineering, 2019 Publication	<1 %

23	ijpsr.com Internet Source	<1%
24	www.id-press.eu Internet Source	<1%
25	aip.scitation.org Internet Source	<1%
26	public.pensoft.net Internet Source	<1%
27	www.thefreelibrary.com Internet Source	<1%
28	Ary - Samsiar. "a Nanoemulsi Fraksi Daun Pelawan (Tristaniopsis merguensis Griff) Sebagai Antidiabetes", Stannum : Jurnal Sains dan Terapan Kimia, 2021 Publication	<1%
29	Pratiwi Liza. "Novel antimicrobial activities of self-nanoemulsifying drug delivery system (SNEDDS) ethyl acetate fraction from Garcinia mangostana L. peels against Staphylococcus epidermidis: Design, optimization, and in vitro studies", Journal of Applied Pharmaceutical Science, 2021 Publication	<1%
30	Win Yee Lim, Eric Wei Chiang Chan, Chia Wei Phan, Chen Wai Wong. "Emulsion formulated using Hibiscus tiliaceus L. extract and flaxseed	<1%

	oil for topical application", Industrial Crops and Products, 2022 Publication	
31	Yan Yi Sim, Kar Lin Nyam. "Application of Hibiscus cannabinus L. (kenaf) leaves extract as skin whitening and anti-aging agents in natural cosmetic prototype", Industrial Crops and Products, 2021 Publication	<1 %
32	download.atlantis-press.com Internet Source	<1 %
33	www.fulltxt.org Internet Source	<1 %
34	Lailiana Garna Nurhidayati. "FORMULASI DAN UJI SIFAT FISIK SEDIAAN NANOEMULSI NATRIUM DIKLOFENAK DENGAN KOMBINASI TWEEN 80 DAN TRANSKUTOL", Sainteks, 2020 Publication	<1 %
35	ejurnal.seminar-id.com Internet Source	<1 %
36	smujo.id Internet Source	<1 %
37	vdocuments.mx Internet Source	<1 %
38	www.atlantis-press.com Internet Source	<1 %

39	Naelaz Zukhruf Wakhidatul Kiromah, Nining Sugihartini, Laela Hayu Nurani. "Development and characterization of clove oil microemulsion", Pharmacia, 2023 Publication	<1%

Exclude quotes	Off	Exclude matches	Off
Exclude bibliography	Off		