Ad-Dawaa' Journal of Pharmaceutical Sciences
ISSN: 2654-7392, E-ISSN: 2654-6973
Vol. 6, No. 1, June 2023, p. 67 – 76
DOI: https://doi.org/10.24252/djps.v6i1.38469

Optimization and Analytical Validation Test of Flavonoid In Capsule Self Nano-Emulsifying Drug Delivery System Containing Extract Parang Romang Leaves

Magfirah[1], Indah Kurnia Utami[2], I Dewa Ayu Nurllya K.D[3], Niluh Puspita Dewi[2]

[1]Department of Pharmacy, Pharmaceutical Technology, STIFA Pelita Mas Palu, Indonesia
[2]Department of Pharmacology, Clinical Pharmacy, and Community, STIFA Pelita Mas Palu, Indonesia
[3]Student, Bachelor in Pharmacy, STIFA Pelita Mas Palu, Indonesia

Article history:
Submited: 13-6-2023
Revised: 24-6-2023
Accepted: 18-7-2023

Corresponding author e-mail: magfiralukman@gmail.com

Cite this article: Magfirah, Utami, I.K., Nurllya, I.D.A.K.D., Dewi, N.P. (2023). Physical Characteristics and Shelf Life Estimation of Instant Powder Drink Made From The Combination of Yellow Sweet Potatoes and Red Beans. Ad-Dawaa' J. Pharm. Sci. 6(1): 67-76.

ABSTRACT

Introdiction: Parang romang contains alkaloids, flavonoids, tannins, and saponins. The secondary metabolites of the leaf extract have low solubility in water. The self-nano emulsifying drug delivery system (SNEDDS) is one solution to increase the solubility of the extract. **Aims** This study aims to obtain the best formula and ensure that the assay analysis method used can provide accurate and reliable results so that they can be trusted. **Methods**: Parang romang leaf extract was added to the optimum mixture of tween 20/80, propylene glycol, and olive oil and then characterized including% transmittance, drug loading, emulsification time, particle size, index polydispersity, zeta potential, dissolution test, Scanning electron microscopy (SEM) and validation of the analysis method including linearity, precision, accuracy, LOD, and LOQ. **Result:** The results showed the ratio of surfactant, co-surfactant, and olive oil which produced the optimum formula for SNEDDS, namely 26 ml; 17.5 ml; and 6.5 ml,% transmittance 85.90, drug loading 45.36, emulsification time for AGF, AIF, and aqua dest was 15-16 seconds, particle size 404.1 nm, index polydispersity 0.840, zeta potential -31.4 mV, dissolution test 92.13%, SEM in the form of spherical chunks, and the result of the validation test of the analytical method with a linearity of r = 0.9902, precision 0.436%, accuracy 103.738%, LOD 1.87 µ / ml and LOQ 5.57 µ / ml. SNEDDS parang romang leaf extract using olive oil as the oil phase, Tween 80 as a surfactant, and propylene glycol as a cosurfactant provide optimal nanoemulsion characteristics and validation of the analytical method meets the requirements. **Conclusion:** Formula 1 SNEDDS parang romang leaf extract using olive oil as the oil phase, Tween 80 as a surfactant, and propylene glycol as a cosurfactant is an optimal formula that provides good nanoemulsion characteristics and validation of analytical methods meets the requirements.

KEYWORDS: Optimization, SNEDDS, ethanol extract of parang romang leaves, validation of analytical methods.

INTRODUCTION

SNEDDS is an isotropic mixture consisting of surfactant, co-surfactant, and oil in spontaneous emulsion form. (Rahayu et al., 2019). The components of SNEDDS include oil as a carrier, surfactants as an emulsifier that lowers the tension between oil and air, and co-surfactants as an emulsifier that maintains the protection of the film between oil and water (Zulfa et al., 2020). Plants used for traditional

medicine, one of which is Boehmeria virgata (G.Forst) Guill or machetes, including plants belonging to the Boehmeria genus with the Urticaceae tribe, are empirically used in the treatment of the Makassar tribe of South Sulawesi as an anti-tumor drug (Magfirah & Christin, 2020). Parang Romang leaves have various benefits that are proven to be active against cancer cells, intestinal inflammation, fractures, and hematemesis and are used in mixing ingredients to protect the skin (Rusdi et al., 2018).

Parang romang (Boehmeria virgata) contains secondary metabolites in the form of alkaloids, terpenoids, phenolics, and flavonoids. However, secondary metabolites of those leaves extract have low solubility in water. Hence, to be the formula, a self-nano emulsifying drug delivery system (SNEDDS) is one of the solutions to increase the extract solubility and bioavailability (Magfirah & Utami, 2021). However, several studies suggested the limitation of herbal Extracts, such as poor aqueous solubility, poor stability, and poor oral bioavailability. To overcome the issues, extract ethanol parang romang leaf was formulated as an oral *self-nano emulsifying drug delivery system* (Magfirah, 2023). Some approaches for enhancing the solubility characteristic of slightly water-soluble drugs included reducing particle size. Several studies have reported the self-nano emulsifying drug delivery system (SNEDDS) has recently increased the dissolution, solubility, and bioavailability of slightly water-soluble drugs such as andrographolide isolated from Andrographis paniculata. It is important to ensure the quality of the dosage form by evaluating the extract ethanol parang romang leaf load in the SNEDDS and optimizing the formula(Nastiti et al. 2021)

A Spectrophotometry UV-Vis method was developed and validated before the load determination of extract ethanol parang romang leaf in the SNEDDS formulation. Method validation is an attempt to evaluate and document evidence of a procedure is feasible to use and yields comparable results. As a result, analytical methods can be used if they meet predetermined parameters. These validation parameters include linearity, the limit of detection, the limit of quantification, accuracy, and precision. In vitro, diffusion study has been conducted to be a tool for evaluating the absorption of SNEDDS in this formulation to the systemic circulation. To obtain reliable measurement results during testing, we need a valid method for determining the active compound in the preparation. The suitability of an analytical procedure must be performed to verify the system in a series of analytical validation methods (Syukri et al, 2020). The current study reported that the validated A Spectrophotometry UV-Vis method had been developed to quantify herbal extract (Yulianto et al. 2017).

This study aims to determine the preparation of self-nano-emulsifying drug delivery system Parang Romang leaf extract to

produce the optimum formulation using the simple lattice design method and to validate the analytical method for determining extract ethanol parang romang leave especially flavonoid concentration in SNEDDS preparations during a diffusion study using by Spectrophotometry UV-Vis method.

MATERIAL AND METHODS

Chemicals and Instrument

Parang romang (*Boehmeria virgata*) plant was taken in Ojoloboku village, Moncongloe district, Gowa regency, South Sulawesi province. The plants were identified at UPT. Sulawesi Biological Resources Tadulako University. The materials used are Alcohol(Merck), Aquadest, Aluminum (III) chloride 1.2% (Merck), Tween 20(Merck), Tween 80 (Merck), olive oil (Bertoli), Propylene glycol (Merck), Acetic acid (Merck), NaOH 0.2 N (Merck), HCL (Merck), Sodium chloride (Merck), DiSodium hydrogen phosphate (Merck), tissue, label paper.

The instrument used is a 40 mesh sieve (Retsch), Erlenmeyer (Pyrex), Magnetic stirrer (Thermo Scientific), Rotary vacuum evaporator (Heidolph), Spectrophotometry UV-Vis Evolution 201 (Thermo Scientific) and Waterbath (Memmert)

Table 1. The composition of the formula

Formula	Tween 20/80 (ml)	PG (ml)	Olive oil (ml)
1	30	15	5
2	20	25	5
3	10	35	5
4	5	40	5
5	40	5	5
6	45	0	5
7	0	45	5

Extraction

Parang romang (*Boehmeria virgata*) leaves (800 grams) were extracted using maceration with 96% ethanol solvent for 3 days. The extract was filtered using filter paper and the obtained filtrate was then concentrated using a rotary vacuum evaporator at a temperature of 60ºC and evaporated using a water bath to obtain the concentrated extract (Magfirah & Utami, 2021).

Preparation of SNEDDS

All SNEDDS formulas contain parang romang leaf ethanol extract 100 mg put into 50 ml each of a mixture of tween 20, tween 80, propylene glycol, and olive oil, the composition of the formula can be seen in Table 1, then mixed at 45 ℃ for 30 minutes using hotplate magnetic stirrer, then ultrasonicated for 1 hour at 45 ℃. The resulting mixture was stored for 24 hours at room temperature to see its homogeneity.

The selected formulas were characterized including emulsification time, %Transmittan determination, drug loading testing, particle size, particle size distribution, zeta potential, dissolution test, and Scanning Electron Microscopy (Magfirah & Utami, 2021). Then followed by method validation including linearity, precision, accuracy, LOD, and LOQ (Syukri et al, 2020).

Characterization of SNEDDS

Emulsification time

The SNEDDS formula (0.1 ml) was added with aqua dest, artificial gastric fluid, and artificial intestinal fluid and until the final volume of 50 ml was homogenized with a hotplate magnetic stirrer at 120 rpm at 37℃ for 15 minutes (Ujilestari et al., 2018).

%Tansmittan

A total of 0.5 mg of SNEDDS was added to 10 ml of distilled water and then a magnetic stirrer for 30 seconds. Measurement of %Transmittan by spectrophotometry at 510 nm (Suryani et al., 2019).

Drug loading

A total of 1 ml of SNEDDS was filled into a 10 ml definite flask to the mark with 10 ml of ethanol and then read the absorbance using spectrophotometry at 510 nm (Magfirah & Utami, 2021).

Particle size, polydispersity index, zeta potential.

Particle size, polydispersity index, and zeta potential were observed by a particle size analyzer (PSA) (Horiba Scientific SZ-100 Horiba, Kyoto. Japan). A total of two drops of nanoemulsion were mixed into 5 ml of distilled water as much as 3 ml was taken and put into a cuvette for analysis (Magfirah & Utami, 2021).

Scanning Electron Microscopy (SEM)

The particle morphology of SNEDDS was analyzed by SEM at an accelerated voltage of 20 kV (Magfirah & Utami, 2021).

Dissolution test

SNEDDS formula and pure parang romang leaf extract with dissolution test equipment at 900 ml aqua dest with a rotation speed of 50 rpm at 37℃. Take 5 ml for 5-30 minutes. After that, the concentration of SNEDDS and parang romang leaf extract was measured using a Spectrophotometer UV-Vis (Baloch, J. et.al. 2019)

Weight uniformity test

Weighed 20 capsules and each capsule was weighed. Calculate the weight of the contents of the capsules and the average weight of each capsule (Reddy et. al, 2022)

Validation Methods

Linearity

A total of 50 mg of quercetin standard solution was dissolved into 50 ml of ethanol to obtain a stock solution concentration of 1000 ppm, after which a dilution of quercetin was made at a concentration of 1.563; 3.125; 6,250; 12,500; 25 ppm. Then add 1 ml of 1.2% aluminum (III) chloride and measure its absorbance at a wavelength of 510 nm (Tuldjanah M, et.al.2022) The absorbance results obtained are then plotted into the line equation (y = ax + b). Linearity is obtained from the correlation coefficient (r) ((Yulianto et al. 2017).

Accuracy and precision

Accuracy and precision were carried out using the spike method to make preparations with a concentration range of 80%; 100%; 120%. Each concentration was made in 3

replications and then the absorbance was read using a UV-Vis spectrophotometer at a wavelength (λ) of 510 nm. The absorbance results were used to calculate the recovery price with a standard deviation (SD) and a relative standard deviation (RSD) ≤ 2 .7%. Compare the percent recovery value with the standard acceptance percent gain (AOAC, 2016). The analysis is carried out on the same day (interday) and will then be analyzed on 3 different days in a row (intraday) (Wulansari & Lubada, 2020)

LOD and LOQ

The detection limit and the quantity limit can be statistically calculated by linear regression line from the calibration curve by making a working standard solution with concentrations of 1.5 ppm, 0.5 ppm, 10.75 ppm, 1 ppm, 1.25 ppm, pipetted 1 ml with the addition of 1 ml Potassium acetate 120 mM, and 1 ml Aluminum (III) chloride incubated up to 30 minutes (Alwi, H. 2017), using spectrophotometry at 510 nm. LOD and LOQ calculations from the standard deviation (SD) and the slope of the standard curve regression equation (S), with the following calculations: LOD = 3.3 (SD/S) and LOQ = 10 (SD/S) (Wulansari & Lubada, 2020)

RESULTS AND DISCUSSION

SNEDDS is a mixture of oils, surfactants, co-surfactants, and also active medicinal compounds that will produce oil in water (o/w) emulsion in a water medium with mild agitation (Izham et al., 2019). The SNEDDS

Optimization and analytical validation test formulation of the extract increases the bioavailability of the extract, so that the therapeutic effectiveness is enhanced (Suryani et al., 2019). Parang romang leaves (Boehmeria virgata (G.forst) Guill. Has been used by the Makassar tribe as a cancer cure (Manggau, 2018). However, the herbal extract has low oral bioavailability, to increase the bioavailability of herbal extract to make SNEDDS (Magfirah, and i. K. Utami, 2021). In this study, SNEDDS was designed using the simple lattice design method with the application of design expert 12 to obtain 7 variations of surfactants, co-surfactants, and oils. Optimization was carried out by mixing surfactant, co-surfactant, and oil to obtain the selected optimization formulation based on emulsification time, % transmittance, drug loading and characterized based on particle size, polydispersity index, zeta potential, SEM (scanning electron microscopy), dissolution test and weight uniformity. Based on the results of the formulation optimization carried out, formula 1 was obtained as the best formula which was then continued for characterization. The results of formulation optimization can be seen in table 2.

Percent transmittance is used to observe the emulsification of the process by measuring the transmittance during the emulsification process (Ujilestari et al., 2018). In Table 2, % Transmittan 85.90% has a clear dispersion with % Transmittan close to 100% indicating that it meets the requirements of nanoemulsion (Syukri et al., 2019). High surfactant concen

Table 2. Composition and characterization SNEDDS of ethanol extract parang romang leaves

Formula	Composition SNEDDS			Characterization SNEDDS				
	Tween 20/80 (ml)	PG (ml)	Olive oil (ml)	% T	Drug loading (ppm)	Emulsification time AGF	AIF	Aquadest
1	30	15	5	85,90	45,36	15	16	15
2	20	25	5	65,60	35,16	35	36	35
3	10	35	5	45,60	22,76	35	36	35
4	5	40	5	25,80	15,89	45	46	45
5	40	5	5	65,90	25,16	25	26	25
6	45	0	5	66,90	25,96	35	36	35
7	0	45	5	15,90	5,98	40	36	40

ration followed by co-surfactant and olive oil, the resulting concentration % Transmittan is close to 100% because the surfactant has lipophilic and hydrophilic parts which can reduce interfacial tension so that it affects the formation of nanoemulsions and nanoemulsion droplet size (Huda & Wahyuningsih, 2018)

Drug loading aims to evaluate drug levels in SNEDDS (Baloch et al., 2019). If the level is higher, it is expected that the drug can reach target cells in the body without being influenced by enzymes in the digestive tract because SNEDDS can bind and protect drugs from first-pass metabolism (Cherniakov et al., 2015). In Table 2, the drug loading value is 45.36 ppm, this shows a high drug loading value, which means that SNEDDS can reach target cells in the body. Particle size, polydispersity index, and zeta potential aim to determine the size range and stability of SNEDDS with PSA (particle size analyzer).

Table 3. Results optimization SNEDDS of ethanol extract parang romang leaves

Characterization SNEDDS	Result ± SD
Particle size	404,1 ± 11,45
Polidipersitas indeks	0,8 ± 0,13
Zeta potensial	-31 ± 1,17

Emulsification time is a very important parameter to describe the stability of the system and prepare emulsification in gastric fluid from the characteristics of SNEDDS (Ujilestari et al., 2018). The emulsification time of SNEDDS when in contact with aqua dest media, artificial gastric fluid, and artificial intestinal fluid shows the spontaneity of the emulsification time <1 minute. Tween 20 and tween 80 are non-ionic surfactants with low toxicity and better emulsification ability which allows rapid dispersion when in contact with biological fluids (Ujilestari et al., 2018). Hydrophilelipophile balance value of 15 which can reduce the interfacial tension of water and oil to help speed up the emulsification time (Venkatesh & Mallesh, 2013). Propylene glycol is commonly used in nanoemulsion formulations to increase water and oil in mixing due to partitioning between the two phases. The results of the characterization of Formula 1 as the best formula can be seen in Table 3.

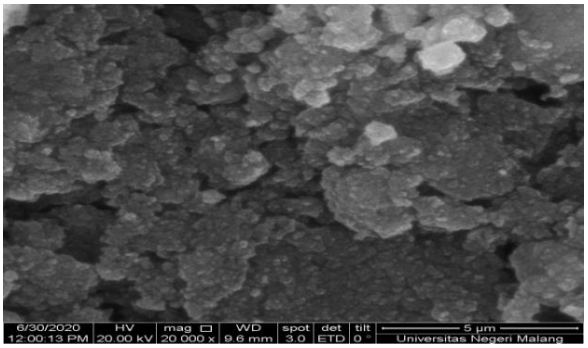

Figure 1. SEM results of SNEDDS formulation of parang romang leaf extract.

formulation using an electron microscope

Table 4. Dissolution test results

Abs SNEDDS	Abs Extract	% SNEDDS	% Extract
0.073	0.055	88.78	4.20
0.073	0.056	89.71	8.88
0.073	0.055	90.18	4.20
0.074	0.055	92.99	4.20
0.075	0.055	98.13	4.20
0.074	0.055	92.99	4.20
Average soluble content		92.13	4.98
Terms		No less than Q+5%	

Based on Table 3. The obtained particle size, polydispersity index, and zeta potential are 404.1; 0.8; -31. The nanoemulsion particle size range is 100-500 nm the smaller the nanoemulsion droplet size, the larger the surface area provided for drug absorption (Magfirah & Utami, 2021). The size of the globules determines the rate and rate of drug release as well as the absorption and bioavailability of SNEDDS (Ujilestari et al., 2018). The polydispersity index shows a value close to 1 indicating a wide droplet size range (De Azevedo Ribeiro et al., 2015). Zeta potential for nanoemulsion preparations is > +30 mV and > -30 mV positive results indicate that the particles are dispersed in a positive charge and vice versa (Kazi et al., 2020).

Observation of particle morphology aims to determine the shape and size of each SNEDDS (Kazi et al., 2020). Morphological results from the SNEDDS formula for ethanol extract of parang romang leaves with a magnification of 20,000 times with a particle size of 5µm. From the observations, it can be seen in Figure 1 that the morphology of the shape of the SNEDDS particle size is in the form of lumps in the form of a regular ball.

Dissolution test to determine conformance to dissolution requirements. The concentration of the active substance dissolved in the preparation is 92.13% and the extract is 4.98%. The concentration of the dissolved active substance in the Indonesian Pharmacopoeia edition IV has complied with the requirements where the dissolved active substance content is not less than Q+5%, this indicates that the active substance SNEDDS of parang romang (Depkes, 1995). The result can be seen in

Table 5. Test results of weight uniformity

Average Weight	Requirement
A1 = 701-716/716 x 100 = 2.09 %	± 7.5 – 15%
A2 = 700-716/716 x 100 = 2.23 %	
A3 = 699-716/716 x 100 = 2.37 %	

Table 4.

The weight uniformity test recognizes the amount of deviation in weight per capsule and dose per capsule. Testing of 20 capsules on the average weight of capsules with a weight difference of 2.09% - 2.37% met the requirements for the average weight of the capsule contents ±7.5% and ±15% (Depkes, 1979). It can be seen in Table 5.

Method validation aims to obtain specific, accurate analytical methods. The results of linearity, precision, accuracy, LOD, and LOQ are (r) 0.9902; 0.436%; 103.738%; 1.876 /ml, and 5.576 /ml. Shows that these values have good linearity, precision, and accuracy of LOD and LOQ with values ranging from 0.99, 2%, 85-115%, and 5709µ/ml (Wulansari & Lubada, 2020). The result can be seen in Table 6.

CONCLUSION

The result optimization and characterization obtained formula 1 as the best formula with a transmittance percentage of 85.90%; drug loading of 45.36 ppm; emulsification time of 15-16 seconds, a particle size of 404.1 nm, polydispersity index of 0.840 and zeta potential of -31.4 mV.

Table 6. Validation of analytical methods SNEDDS parang romang

Average Weight	Requirement
Linearity = (r) 0,9902	≥ 0.99
Precision = 0,436 %	≤ 2%
Accuracy = 103,738 %	85 – 115 %

The validation of the analytical method meets the requirements of the validation test of the spectrophotometric analysis method including linearity with a value of r = 0.9902, precision 0.436%, accuracy 103.738%, LOD 1.87 and LOQ 5.57 µ/ml.

CONFLICT OF INTEREST

The author declares that there is no conflict of interest regarding this research.

REFERENCES

Alwi, H. 2017. Validasi metode analisis flavonoid dari ekstrak etanol kasumba turate (*Carthamus tinctorius* L.) secara spektrofotometri. [Doctoral dissertation]. Makasar: Universitas Islam Negeri Alauddin Makasar.

Baloch, J., Sohail, M. F., Sarwar, H. S., Kiani, M. H., Khan, G. M., Jahan, S., Rafay, M., Chaudhry, M. T., Yasinzai, M., & Shahnaz, G. (2019). Self-nano emulsifying drug delivery system (snedds) for improved oral bioavailability of chlorpromazine: In vitro and in vivo evaluation. *Medicina (Lithuania)*, *55*(5).

Cherniakov, I., Domb, A. J., & Hoffman, A. (2015). Self-nano-emulsifying drug delivery systems: An update of the biopharmaceutical aspects. *Expert Opinion on Drug Delivery*, *12*(7), 1121–1133.

De Azevedo Ribeiro, R. C., Barreto, S. M. A. G., Ostrosky, E. A., Da Rocha-Filho, P. A., Veríssimo, L. M., & Ferrari, M. (2015). Production and characterization of cosmetic nanoemulsions containing Opuntia ficus indica (L.) Mill extract as a moisturizing agent. *Molecules*, *20*(2), 2492–2509.

Depkes, R. (1979). *Farmakope Indonesia* (III). Dirjen POM.

Depkes, R. (1995). *Farmakope Indonesia* (IV). Dirjen POM.

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.

Izham, M. N. M., Hussin, Y., Aziz, M. N. M., Yeap, S. K., Rahman, H. S., Masarudin, M. J., Mohamad, N. E., Abdullah, R., & Alitheen, N. B. (2019). Preparation and characterization of self nano-emulsifying drug delivery system loaded with citraland its antiproliferative effect on colorectal cells in vitro. *Nanomaterials*, *9*(7). https://doi.org/10.3390/nano9071028

Kazi, M., Shahba, A. A., Alrashoud, S., Alwadei, M., Sherif, A. Y., & Alanazi, F. K. (2020). Bioactive self-nanoemulsifying drug delivery systems (bio-snedds) for combined oral delivery of curcumin and piperine. *Molecules*, *25*(7).

Lukman, M., & Christin, V. (2020). Analisis profil bobot badan tikus dan gejala toksis pada pemberian ekstrak etanol daun parang romang (*Boehmeria virgata*) terhadap tikus putih (*Rattus novergicus*). *Jurnal Farmasi Galenika (Galenika Journal of Pharmacy) (e-Journal)*, *6*(1), 1–6. https://doi.org/10.22487/j24428744.2020.v6.i1.13928

Magfirah, & Utami, I. kurnia. (2020). *formulasi nanoemulsi ekstrak etanol daun parang romang (boehmeria virgata)* (pertama). Magama.

Magfirah, & Utami, I. kurnia. (2021). Optimization and characterization of formulation self-nanoemulsifying drug delivery system ethanol extract of romang parang leaves (*Boehmeria virgata*). *Asian Journal of Pharmaceutical and Clinical Research*, *14*(1), 207–212.

Manggau, M. A. (2018). In vitro study of the alkaloid anticancer compound from makassar medicinal plants *Boehmeria virgata* Linn. *Int. J. Pharm. Sci. Rev. Res., 49(1)*(13), 77–81.

Tuldjanah, M., Fajarizki, G. R.,Tandi, J., & Magfirah. (2022). Penetapan kadar metabolit sekunder ekstrak etanol daun alpukat (*Persea americana* Mill.) secara spektrofotometri uv-vis. *Farmakologika Jurnal Farmasi.*21(2). 31-42

Nastiti, C. M. R. R., & Riswanto, F. D. O. R. (2021). Analytical method validation and formula optimization of topical nanoemulsion formulation containing resveratrol. I*ndones. J. Chem*. 21 (5), 1287 - 1297

Rahayu, N. Y., Budiharjo, A., Pangastuti, A., Artanti, A. N., Prihapsara, F., & Harini, M. (2019). Optimation formula of snedds dosage from ethanol extract of turmeric (*Curcuma domestica*) with waste oil of eel (*Anguilla spp*.) as a carrier. *IOP Conference Series: Materials Science and Engineering*, *578*(1). 25-30

Reddy, N. N., & Fazal ul Haq, S. M. 2022. Formulation and evaluation of hydrogel based oral controlled drug delivery system for saquinavir. *Saudi J Med Pharm Sci*, 8(10): 575-584

Rusdi, M., Hasan, T., Ardillah, A., & Evianti, E. (2018). Perbandingan metode ekstraksi terhadap kadar flavonoid total dan aktivitas antioksidan batang *Boehmeria virgata*. *Ad-Dawaa' Journal of Pharmaceutical Sciences*, *1*(1), 16–24.

Suryani, Zubaydah, W. O. S., Sahumena, M. H., Adawia, S., Wahyuni, R., Adjeng, A. N. T., Nisa, M., Kasmawati, H., Ihsan, S., Ruslin, & Aswan, M. (2019). Preparation and characterization of self-nanoemulsifying drug delivery system (SNEDDS) from *Moringa oleifera* L.and *Cassia alata* L. leaves extracts. *AIP Conference Proceedings*, *2199*.

Syukri Y., Nugroho, B. H., & Sirin, M. 2020. Determination of andrographolide content in self nano emulsifying drug delivery system (snedds) for in vitro diffusion study using validated hplc. AIP Conference Proceedings,2229.

Syukri, Y., Fitriani, H., Pandapotan, H., & Nugroho, B. H. (2019). Formulation, characterization and stability of ibuprofen-loaded self-nano emulsifying drug delivery system (snedds). *Indonesian Journal of Pharmacy*, *30*(2), 105–113.

Ujilestari, T., Martien, R., Ariyadi, B., Dono, N. D., & Zuprizal. (2018). Self-nanoemulsifying drug delivery system (Snedds) of *Amomum compactum* essential oil: design, formulation, and characterization. *Journal of Applied Pharmaceutical Science*, *8*(6), 14–21. https://doi.org/10.7324/JAPS.2018.8603

Venkatesh, M., & Mallesh, K. (2013). Self-nano emulsifying drug delivery system (snedds) for oral delivery of atorvastatin- formulation and bioavailability studies. *Journal of Drug Delivery and Therapeutics*, *3*(3).

Wulansari, S. A., & Lubada, E. I. (2020). Validasi metode analisa pada penetapan kadar koenzim q10 dalam sediaan mikroemulsi menggunakan metode spektrophotometer uv-vis. *Jurnal Kimia Riset*, *5*(1), 29-33.

Yulianto, A. N.,Muctharidi., Rusdiana, T., & Subarnas, A. (2017). Validation of uv-vis spectrophotometry methode for analysis of apigenin in celery extract (*Apium graveolens* L.). *Pharmaciana, 7(2),159-*

Zulfa, E., Novianto, D., & Setiawan, D. (2014). Formulasi nanoemulsi natrium diklofenak dengan variasi kombinasi tween 80 dan span 80 : kajian karakteristik fisik sediaan. *Media Farmasi Indonesia*, *14*(1), 1471–1477.