



UNIVERSITI PUTRA MALAYSIA

***FABRICATION OF NANO LOADED JACKFRUIT LEAVES EMULSION BY
ULTRASOUND METHOD***

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BY

NIK SYAZWANI BINTI NIK HARON

**This thesis submitted in fulfilment of the requirement for the degree of Bachelor of
Engineering (Process and Food) from the Faculty of Engineering, Universiti Putra
Malaysia**

ABSTRACT

Fabrication of Nano Loaded Jackfruit Leaves Emulsion by Ultrasound Method

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Jackfruit leaves have recently attracted attention for its therapeutic characteristics and cosmeceutical applications. It is believed that the artocarpin compound in the jackfruit leaves possess anti – inflammatory effect that capable in wound healing. Besides, it is reported that the jackfruit leaves also contain high amount of antioxidant properties which will be beneficial to the skin (Haswani Maisarah, 2018). However, delivering the beneficial attributes of the bioactive ingredients into formulation is challenging due to poor solubility and low bioavailability of bioactive substances. Nanoemulsion system is an emulsion containing two phases (oil & aqueous) and surfactant with droplet size in between 20-200 nm. This system have been used widely in the cosmetic and pharmaceutical area as creams, lotions, moisturizers and also foundation. Since human skin pore size are approximately in the range of 40–80 μm , the nanoemulsion system is the best approach to improve the efficiency of the bioactive extract in the jackfruit leaves. Therefore, nanoemulsion has been chosen to ensure an effective transdermal delivery of bioactive ingredients in jackfruit leaves extract for skin application. This study aimed to formulate an oil in water (O/W) nanoemulsions containing Jackfruit leaves extract in a cream form as well as to evaluate its physicochemical properties and the stability of nanoemulsions during eight (8) weeks of storage at room temperature and 45°C by high energy method which

is ultrasound. In this work, Central Composite Design (CCD) was used to determine the optimal composition of the nanoemulsions. The optimum formulation of the nanoemulsions produces were at 80 % water content, 0.93 O/S, and 25 minute ultrasonication time and 0.05 % of extract of jackfruit leaves with the optimum particle size of 99.6 nm. The nanoemulsions possessed a pH compatible with skin pH (4.5-6.0); droplet size varying from 99.6 nm to 127.8 nm; low viscosity of 1.72 mPa.s to 2.52 mPa.s and turbidity of 486 NTU to 521 NTU. Under the optimal conditions, the average droplet size obtained was 99.6 nm. The optimal nanoemulsions composition was observed to be stable under an accelerated stability study during storage at room temperature and 45 °C for 60 days. The optimal formulation exhibit suitable physicochemical properties and stability against phase separation and coalescence which is desirable for skin products.

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TABLE OF CONTENT

ABSTRACT	i
ACKNOWLEDGMENT	iii
APPROVAL SHEET	iv
TABLE OF CONTENT	vi
LIST OF FIGURES	ix
LIST OF TABLES	xi
CHAPTER 1	1
INTRODUCTION	1
1.1 Background	1
1.2 Problem Statement	4
1.3 Hypothesis	5
1.4 Objectives	5
CHAPTER 2	6
LITERATURE REVIEW	6
2.1 Introduction	6
2.2 Jackfruit (Artocarpus Heterophyllus)	7
2.2.1 Introduction to Jackfruit	7
2.2.2 Jackfruit Parts and Benefits	9
2.2.3 Jackfruit Leaves as Medicinal Ingredient	13
2.3 Nanoemulsion	15
2.3.1 Introduction of Nanoemulsions	15
2.3.2 Nanoemulsions for Cosmetic Application	16
2.4 Selection of Oil Phase	18
2.4.1 Caprylic Capric Triglycerides	18
2.5 Selection of Surfactant	20
2.6 Ultrasound Method	22
2.7 Physical Properties of Nanoemulsions	25
2.7.1 Particle Size	25
2.7.2 Viscosity	25
2.7.3 pH	26
2.7.4 Turbidity	27
2.7.5 Stability Studies	27

CHAPTER 3	28
METHODOLOGY	28
3.1 Introduction	28
3.2 Material of Experiment	28
3.3 Extraction by Maceration Method.	29
.....	30
.....	30
3.4 Solubility of Extract.	30
3.5 Preparation of Emulsion	30
3.6 Preparation of Nanoemulsions: Ultrasonic Method	32
3.7 Verification of the Model	32
3.8 Analysis of Nanoemulsion Physicochemical Properties	33
3.8.1 Particle Size	33
3.8.2 Viscosity	33
3.8.3 pH Determination	33
3.8.4 Turbidity	34
3.8.5 Stability Test	34
.....	35
.....	35
.....	35
.....	35
CHAPTER 4	36
RESULT AND DISCUSSION	36
4.1 Introduction	36
4.2 Solubility of Jackfruit Leaves Extract	36
4.3 Statistical Analysis	37
4.3.1 Analysis of Variance (ANOVA)	37
4.3.2 Optimization of Nanoemulsion Formulation	39
4.3.3 Validation of Responses	43
4.4 Analysis of Physicochemical Properties during Storage	45
4.4.1 Particle Size	45
4.4.2 Viscosity	46
4.4.3 pH	47
4.4.4 Turbidity	49
4.5 Stability Analysis	51
CHAPTER 5	53

CONCLUSION AND RECOMMENDATIONS	53
5.1 Conclusion	53
5.2 Recommendations for Future Studies	55
APPENDIX A	57
APPENDIX B	59
APPENDIX C	60
APPENDIX D	62
REFERENCES	64



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LIST OF FIGURES

Figure 2. 1: The characteristic of cultivars found in Hawaii. (Crane et al., 2002).....	7
Figure 2. 2: Various name of jackfruit. (Trindade et al., 2006)	8
Figure 2. 3: Availability in different countries. (Baliga et al., (2011).	8
Figure 2. 4: Production of jackfruit in 2005- 2011	8
Figure 2. 5: Difference between jackfruit clones.	9
Figure 2. 6: The leaves of jackfruit. (Kim Winkinson, 2000).....	10
Figure 2. 7: The fruit of jackfruit. (Kim Winkinson, 2000).....	10
Figure 2. 8: Different parts of the jackfruit plant	11
Figure 2. 9: Constituents of jackfruit plant. (Trindade et al., 2006).	12
Figure 2. 10: The use of jackfruit parts in medicine. (Haq 2006).....	12
Figure 2. 11: Pharmacological activities of jackfruit plant. (Arora & Parle, 2016)...	14
Figure 2. 12: Nanoemulsions appearance versus microemulsions.....	15
Figure 2. 13: Properties of Nanosystems determining skin absorption and potential routes of penetration. (Nastiti et al., 2017).....	17
Figure 2. 14: Ranking of companies that use Nano-related products.	18
Figure 2. 15: Chemical structure of caprylic/capric triglyceride. (Jaworska et al., 2014)	20
Figure 2. 16: Representative nanoemulsions of water-insoluble drugs that are generated by ultrasonication. (Sivakumar et al., 2014).....	24
Figure 2. 17: Nanoemulsions process mechanism, advantages and disadvantages. (Yukuyama et al., 2016).....	24
Figure 3. 1: leaves powder and ethanol in incubator shaker	29
Figure 3. 2: Filtration of obtaining the extract	30

Figure 3. 3: Evaporation of leaves filtrate.....	30
Figure 3. 4 : Summary of process design.....	35
Figure 4. 1: Response surface plot showing the effect of O/S ratio and water content towards particle size.....	41
Figure 4. 2: Response surface plot showing the effect of water content and sonication time towards particle size.....	43
Figure 4. 3: Changes in particle size of nanoemulsions during storage for 8 weeks at room temperature and 45°C.....	46
Figure 4. 4: Changes in viscosity of nanoemulsions during storage for 8 weeks at room temperature and 45°C.	47
Figure 4. 5: Changes in pH of nanoemulsions during storage for 8 weeks at room temperature and 45°C	48
Figure 4. 6: Changes in turbidity of nanoemulsions during storage for 8 weeks at room temperature and 45	50
Figure 4. 7: Physical stability after storage of 2 month at room temperature, and 45°C	52
Figure C. 1: Front view of rotary evaporator	60
Figure C. 2 : Front view of ultrasound machine	60
Figure C. 3 : Position of sample during ultrasonication	61
Figure C. 4: Front view of a rheometer.....	61
Figure D. 1: 20 Formulation of nanoemulsions by ultrasound method	63

LIST OF TABLES

Table 3. 1: Constraints of variables proportions	31
Table 3. 2: CCD design obtained from Design Expert software.....	31
Table 4. 1: ANOVA of the fitted linear equation for particle size of nanoemulsions	38
Table 4. 2: Predicted and actual response for optimized formulation	44
Table A. 1: Particle size of nanoemulsions during 8 weeks storage at room temperature and 45°C	57
Table A. 2: pH of nanoemulsions during 8 weeks storage at Room temperature and 45 °C	57
Table A. 3: Turbidity of nanoemulsion during 8 weeks storage at Room temperature and 45°C	58
Table A. 4: Viscosity of nanoemulsion during 8 weeks storage at Room temperature and 45°C.	58
Table B. 1 Regression coefficients result of the final reduced models.....	59
Table B. 2 Comparison between experimental and predicted response.....	59

CHAPTER 1

INTRODUCTION

1.1 Background

Medicinal plant products are widely used all over the world. Besides, their potential phytochemical component is being actively studied for various purposes such as for their pharmacological activities. Belonging to the Moraceae family, some important species are *Artocarpus heterophyllus*, *Artocarpus altilis*, *Artocarpus hirsutus*, *Artocarpus lakoocha* and *Artocarpus camansi* (Hari & Divya, 2014). Jackfruit (*Artocarpus heterophyllus* Lam.) trees can be found widely in India, Bangladesh, and in many parts of Southeast Asia (Swami et al., 2012). A study by Hari & Divya, (2014) found that different parts of the *Artocarpus* species such as seeds, fruits, leaves, roots, and barks can be used as a traditional remedy to cure various diseases and used in various medicinal practices. From the journal (“*Artocarpus heterophyllus* Lam . (Moraceae): Phytochemistry , Pharmacology and Future Directions , a mini-review,” 2017) stated the plant is reported to possess antibacterial, anti-inflammatory, antidiabetic, antioxidant and immunomodulatory properties. Besides, the important compound such as sapogenins, cycloartenone, cycloartenol, β -sitosterol, and tannins can be found in the leaves and stem of the jackfruit. The compounds presence is traditionally used in healing fever, boils, wounds and skin

diseases. However, up to my knowledge the use of jackfruit leave extract in cosmetic is still not widely studied.

In Malaysia, the cosmetic field has become an important field to be studied as the consumers are becoming more environmentally conscious. The demand for natural pharmaceutical and cosmetic product greatly increasing as they want products that are safe to be consumed. However, to produce a product with the beneficial bioactive ingredients is a great challenge due to poor solubility and low bioavailability of bioactive substances which may affect their effective transdermal (Che Sulaiman et al., 2016). Therefore, according to H. Chen et al., (2006) to enhance the permeation of the bioactive ingredients, some formulation such as emulsion and gels have been studied. Thus, nanotechnology is one of the method that can be practice to produce the formulation. A study by Shen et al., (2011) found that Nano emulsion is the most significant colloidal Nano system available as this system are able to increase the solubility of bioactive ingredients.

Nanoemulsions (NE) is emulsion with the droplet size of 20 nm - 200 nm and it has smaller droplet than the optical wavelengths of the visible spectrum, which make it appear visibly different from micro-scale (Ghotbi et al., 2014). Hence, Sugumar et al., (2014) stated NE will appear nearly transparent due to its small size. Besides, a smaller size particle will have an improved stability compared to larger size particle such as an emulsion. A typical NE contains water, oil, and surfactant with the different ratio to produce a stable NE. Nanoemulsions can be used in the pharmaceutical field in the form of drug delivery system for topical, ocular, intravenous, internasal and oral (Gupta et al., 2016). In addition, Yukuyama et al., (2016) reported that Nano emulsion possesses the characteristic such as small droplet size for uniform permeation to the skin, large surface area, drug carrier properties, firm formation to the skin, high

stability and pleasant aesthetic characteristics and skin feel. Due to its characteristic, the NE is a suitable system to be applied in the cosmetic industry.

There are two primary methods in the preparation of NE which are the high energy method and low energy method (Solans, Izquierdo, Nolla, Azemar, et al., 2005). For low energy method, the Phase Inversion Temperature (PIT) method is the most widely used method in the industry. For high energy method, NE can be achieved by using high-pressure homogenizer and ultrasound method. In order to produce a NE, one of the high energy method that can be used is ultrasonic agitation (Ghotbi et al., 2014). By using ultrasound method, the particle size of emulsion is broken down by the action of vibration of the sonicator probe. Therefore, the ultrasound method will be used to produce emulsions with nanosized which are in the range of 20 - 200 nm.

1.2 Problem Statement

For many years, various plants are being used as traditional remedies to prevent, treat and cure various illnesses and diseases before modern medicine takes place. Realizing the natural benefits of the plant, various product with plant - based emerged as the demands for organic products are increasing rapidly. In addition, World Health Organization (WHO) reported that between 25-40% of pharmaceuticals drugs are derived from plants. This is due to the reason that the bioactive compound in the plants can provide various advantages when it is being consumed. However, up to my knowledge, there is very little information have been reported about the approach can be used to maximize the benefits of the jackfruit leaves phytochemicals in the area of transdermal application. The compounds present in the leave is reported to have the ability in healing fever, boils, wounds and skin diseases. Therefore, in this study, the extract from the jackfruit leaves is chosen as the bioactive ingredients that will be incorporated in the formulation to form nanoemulsion that will increase the efficiency of the product.

Moreover, there are various products in Malaysia that use jackfruits as their base. However, there is still no published work in the study of the formation of Nano loaded jackfruit leave emulsion for cosmetic application. This emulsion will be in the Nanoemulsion (NE) form which is the droplet size is in the range of 20 nm - 200 nm (Ghotbi et al., 2014). A study by Teo et al., (2010) found that the small particle size of nanoemulsion will provide advantages such that it improved the penetration of active ingredients to the skin compared to the macrosized emulsion. Therefore, in this work, high energy method which is ultrasound will be applied to produce nanoemulsion that are suitable to be used as a transdermal application in the future.

1.3 Hypothesis

The optimal composition of the nano loaded jackfruit leaves emulsion will be produced by using the high energy method which is ultrasound; and the changes during storage period of the physicochemical properties in term of particle size, pH, turbidity, viscosity and, stability will be analysed. The stable nanosized emulsion will have higher efficiency for skin application compare to the macrosized emulsion.

1.4 Objectives

The main objective of this study are as follow:

- I. To extract jackfruit leaves by maceration method.
- II. To prepare and analyse the nano loaded jackfruit leaves.

CHAPTER 2

LITERATURE REVIEW

2.1 Introduction

This chapter will discuss in details the information in the research background as described in Chapter 1. In order to give more understanding of the study, various literature have been reviewed. This chapter includes Section 2.2 Jackfruit, Section 2.3 Nanoemulsions and Section 2.4 Selection of oil phase. In the meanwhile, Selection of surfactant and Ultrasound method were discussed in section 2.5 and Section 2.6 respectively. The physicochemical properties of nanoemulsions were discussed in Section 2.7.

2.2 Jackfruit (*Artocarpus Heterophyllus*)

2.2.1 Introduction to Jackfruit

Artocarpus heterophyllus belongs to the family Moraceae with large fruit about 49 kg (Moke et al., 2017). The family of Moraceae consists of about sixty genera and nearly 1400 species, including *Artocarpus*, *Morris*, and *Ficus*. Figure 2.1 shows the characteristic of cultivars found in Hawaii.

Cultivar and origin	Growth habit and rate	Fruit weight	Fruit shape	Yield per tree	Season and months	Comments
'Black Gold' Australia	Open, spreading, fast	10 kg (22 lb)	Long, tapered	55–90 kg (120–200 lb)	Late, Sept.–Oct.	Tree easily pruned to maintain small tree (~2.5 m [8 ft])
'Dang Rasmi' Thailand	Open, spreading, fast	8–9 kg (18–20 lb)	Uniform oblong	(75–125 kg (165–275 lb)	Mid, July–Aug.	Vigorous tree; annual pruning needed to maintain moderate size (~3.3 m [11 ft])
'Gold Nugget' Australia	Dense, spreading, fast	3–5.5 kg (7–12 lb)	Round	60–80 kg (132–176 lb)	Early, May–June	Thinning number of fruit recommended; tree easily pruned to maintain small tree (~2.5 m [8 ft])
'Honey Gold' Australia	Sparse, spreading, slow-moderate	4.5–5.5 kg (10–12 lb)	Blocky	35–50 kg (77–110 lb)	Mid, July–Aug.	Thinning number of fruit recommended; tree easily pruned to maintain small tree (~2.5 m [8 ft])
'Lemon Gold' Australia	Moderately dense, spreading, moderate	6 kg (13 lb)	Blocky	30–45 kg (66–100 lb)	Mid, July–Aug.	Vigorous tree; annual pruning needed to maintain moderate size (~3.5 m [12 ft])
'NS1' Malaysia	Dense, upright, moderate	4–5.5 kg (9–12 lb)	Blocky	90 kg (200 lb)	Early, May–June	Thinning number of fruit recommended for young trees; moderately vigorous tree; annual pruning to maintain moderate size (~3 m [10 ft])

Figure 2. 1: The characteristic of cultivars found in Hawaii. (Crane et al., 2002)

The tree is 8–25 m (26–82 ft.) in height a stem diameter of 30–80 cm (12–32 in) that is acknowledged by its large fruit. The trees are mostly planted at various region such as Southeast Asia, India, Burma, China, Sri Lanka, Malaysia, Indonesia, Thailand, and the Philippines (Elevitch & Manner, 2006). Jackfruit is often being mistaken with the Cempedak. The difference between this fruit is jackfruit has a larger size, more latex, and thinner rind. In different countries, jackfruits are known by various name (Arung et al., 2006) and different availability which show in Figure 2.2 and Figure 2.3 below.

S/No	Name	Language	Country/Region
1	Fakihat Al Jaka	Arabic	Middle East, North Africa
2	Bōluóni	Chinese	China
3	Jackfruit, Nangka	Dutch	Netherlands, south Africa
4	Jackfruit	English	England, USA, New Zealand
5	Jacquier, Jaquier	French	France
6	Jackbaumfrucht, Jackfrucht	German	Germany
7	καρποί [karpoi]	Greek	Greece, Cyprus
8	katahal	Hindi	India
9	Catala	Italian	Italy
10	Paranitsu	Japanese	Japan
11	Baramil	Korean	Korea
12	Jaca	Portuguese	Portugal
13	Árbol de jack, Panapén, Yaca	Spanish	Spain

Figure 2. 2: Various name of jackfruit. (Trindade et al., 2006)

Country	Month of the year	Major Varieties found
Australia	June-April	Black gold, Golden nugget, cheena, cochun, kappa, naben, Mutton
Bangladesh	June-August	Topa, Hazari, Chala, Goal, Khaja
Brazil	January-March, August-October	Jaca-dura, Jaca-Mole
India	April-July	Khujja, ghila, hazari, gulabi, vanka, chanpa, handia, Safeda
Indonesia	August-January	Nagakasalak, Tabouey, Kandel
Malaysia	April-August, September-December	J-31, J30, Nagakabitulang, Na29, Na31
Philippines	March-August	J-01, J-02, TVC, Torres
Sri Lanka	February-November	Vela, varaka, peniwaraka, kuruwaraka
Thailand	January-May, October-December	Dang Rashm, Golden pillow
USA (Florida)	May-August, September-October	Fair childfirst, Sweet Fairchild, Black gold, Golden nugget, Dang Rashm, J-31, J-30, Tabouey

Figure 2. 3: Availability in different countries. (Baliga et al., (2011).

In Malaysia, the planted area and production has expanded. The good weather condition and management of the crop aided the increment (Ismail & Kaur, 2013).

Figure 2.4 below shows the production of jackfruit in 2005-2011.

Year	Planted Area (Hectare)	Productive Area (Hectare)	Average Yield (Mt/ha)	Production (MT)	Production Value (RM)
2005	3,133	1,754	10.0	17,624	44,137,080
2006	3,123	1,748	10.7	18,712	34,381,429
2007	3,201	1,836	10.0	18,415	34,986,420
2008	3,340	1,957	13.7	26,748	61,519,278
2009	3,359	1,975	10.1	19,934	45,847,554
2010	3,516	2,015	9.7	19,516	44,483,050
2011p	3,534	2,012	9.7	19,614	44,707,339

Source: Perangkaan Agromakanan 2011, Ministry of Agriculture and Agro-based Industry Malaysia

Figure 2. 4: Production of jackfruit in 2005- 2011

New jackfruit clones were produced in 2011. These clones were J32 known as Mantin, J33 known as Tekam Yellow, and J35 known as Mastura. Figure 2.5 shows the difference between jackfruit clones (Ismail & Kaur, 2013)

Characteristics	Mantin	Tekam Yellow	Mastura
Reg. Number	J32	J33	J35
Origin	Negeri Sembilan	Rawang, Selangor	Crossbreed of CJ1 & CJ6 varieties
Average Weight (per fruit)	15 - 20 Kg	16 - 20 Kg	15 - 25 kg (The weight can reach more than 40 kg with good management)
Skin Colour of Ripe Fruits	Yellowish green	Yellowish green	Yellowish green
Shape	Oblong	Oblong	Oblong with tapered shoulder
Bulb	Thick & firm	Thin	Thick
Colour of Bulb	Orange	Yellow	Golden yellow
Taste/Texture	Mildly sweet, moist, a bit hard	Very sweet, crunchy & soft	Sweet & Firm
Production of Latex	High	High	Low

Source: Farmers' Organization Authority, 2011

Figure 2. 5: Difference between jackfruit clones.

2.2.2 Jackfruit Parts and Benefits

The fruit has the properties such as juicy, sweet-smelling and tasteful which can be eaten fresh or preserved in various ways. It also has oblong-cylindrical in shape, typically 30–40 cm in length with weight around 50 kg have been reported by (Morton 1987). Then, the seed usually can be boiled, cooked in the dish and added as the baking ingredient. Besides, the waste from the leaves and fruit may function as food for cattle, pigs, and goats (Elevitch & Manner, 2006). Seeds are round in shape with 2–3 cm in length by 1–1.5 in diameter with light brown colour. As the flowers grow, the colour become darker compare to the younger flowers. The flower own male and female spikes which the male are more dense and fleshy while the female is larger and rounded in shape. Elevitch & Manner, (2006) also state that the leaves are in dark green in colour, shiny, firm and large which up to 16 inches in length. Then, it is deeply lobed

as it premature. Figure 2.6, Figure 2.7, and Figure 8 show the different parts of the jackfruit plant.



Figure 2. 6: The leaves of jackfruit. (Kim Winkinson, 2000)



Figure 2. 7: The fruit of jackfruit. (Kim Winkinson, 2000)

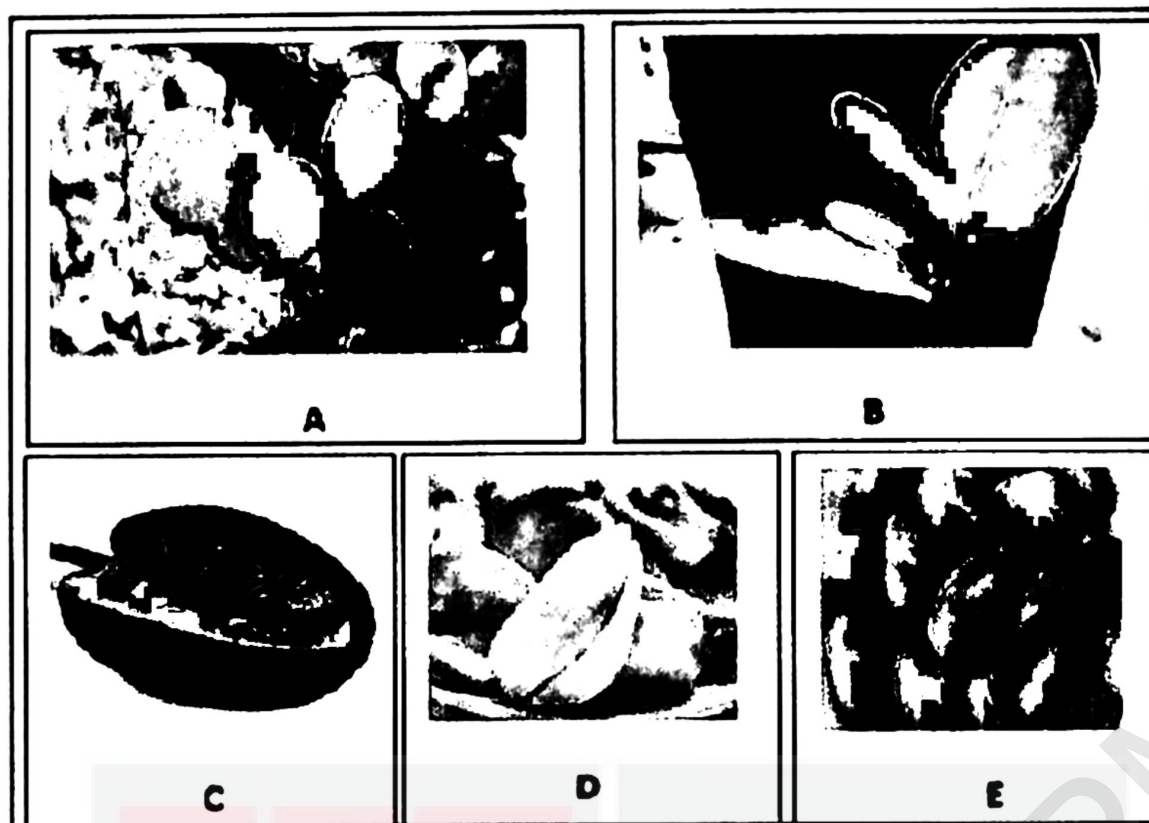


Figure 2. 8: Different parts of the jackfruit plant

A few jackfruit products can be found in the market such as jackfruit in can and jackfruit flavours. Besides, diverse range of products can be found in the market which are using jackfruit powders as the ingredients (Omar et al., 2011). According to (Moke et al., 2017) various parts of the plant such as the bark, roots, leaves, and fruit have the abilities as a traditional medicine to cure various illnesses.

In South-East Asia, Indonesia, Western part of Java and India. *Artocarpus* species are widely used as food and traditional folk medicines (Hari & Divya, 2014). This is due to the reason that certain diseases can be avoided by the action of the bioactive compound in the fruits (Galaverna and others 2008). According to Azizur Rahman et al., (1999) various beneficial sources such as carbohydrates, minerals, carboxylic acids, dietary fibre, and vitamins can be found in the jackfruit plant. There is a huge number of found flavonoid can be found in the jackfruit plant (Wei et al., 2005).

Apart from that, Rama Rao et al., (1973) found that various chemical compounds are present in the jackfruit plant such as colourings, morin, dihydromorin, cynomacurin, artocarpin, isoartocarpin, cyloartocarpin, artocarpesin,

oxydihydroartocarpesin, artocarpetin, norartocarpetin, cycloartinone, and artocarpanone. Rich source of phenolic compounds in the jackfruit plant, provide various benefits to be used in the application of food and improve the health benefits (Jagtap et al., 2010). The Figure 2.9 shows the constituents of jackfruit plant and Figure 2.10 show the use of jackfruit parts in medicine.

Sr.	Phytoconstituents	Plant Part
1	Carbohydrates- Starch, sugar, Dietary fiber	Fruit, Seed
2	Minerals- Calcium, Magnesium, Phosphorus, Potassium, Sodium, Iron	Seed, Fruit
3	Fatty Acids- Capric, Myristic, Lauric, Palmitic, Oleic, Stearic	Fruit
4	Organic Acids- Malic acid, Citric acid	Fruit
5	Carotenoids- 2-carotene, 1-carotene, 1-zeacarotene, 2-zeacarotene, Dicarboxylic Carotenoids	Seed, Fruit
6	Flavonoids- Artocarpine, Artocarpetin, Artonins A, Morin, Dihydromorin, Artocarpanone, Artocarpesin	Fruit
7	Lectin- Jacalin	Seed
8	Volatiles- Isopentylisovalerate, Butyl isovalerate, Butyl Acetate	Seed, Fruit
9	Tannins	Stem, Leaf
10	Vitamins- Vitamin A, Thiamine, Riboflavin, Vitamin E	Fruit

Figure 2. 9: Constituents of jackfruit plant. (Trindade et al., 2006).

No.	Plant part	Use
1	Roots	An extract of roots is used in treating skin diseases, asthma and diarrhea.
2	Leaves	An extract from leaves and latex cures asthma, prevents ringworm infestation and heals cracking of feet. Leaf extract is given to diabetics as a control measure. Heated leaves are reported to cure wounds, abscesses and ear problems and to relieve pain. An infusion of mature leaves and bark is used to treat gallstones. A tea made with dried and powdered leaves is taken to relieve asthma. The ash of jackfruit leaves burned with maize and coconut shells is used alone or mixed with coconut oil to heal ulcers.
3	Flowers	Crushed inflorescences are used to stop bleeding in open wounds.
4	Fruits	Ripe fruits are laxative.
5	Pulp	The jackfruit pulp and seeds are nutritious tonic and useful in overcoming the influence of alcohol on the system.
6	Seed	The seed starch is given to relieve biliousness. Roasted seeds are regarded as an aphrodisiac. Increased consumption of ripe jackfruit kernels alleviates vitamin A deficiency. Extract from fresh seeds cures diarrhea and dysentery. Extract from seeds (or bark) helps digestion.
7	Bark	An extract from bark and rags (nonedible portion of ripe fruits) or roots helps cure dysentery. The bark is made into poultices. Ash produced by burning bark can cure abscesses and ear problems.
8	Latex	Mixed with vinegar, the latex promotes healing of abscesses, snakebites and glandular swellings.
9	Wood	The wood has a sedative property; its pith is said to aid abortion.

Figure 2. 10: The use of jackfruit parts in medicine. (Haq 2006)

2.2.3 Jackfruit Leaves as Medicinal Ingredient

Moke et al., (2017) claimed jackfruit contains several useful compounds like flavonoids, sterols, and prenylflavone that beneficial for abundant of pharmacological properties. Besides, Wetprasit et al., (2000) state that saponin, cycloartenone, cycloartenol, β -sitosterol can be found in the jackfruit leaves. From phytochemical studies, it is found that compound such as flavonoids, terpenoids, steroids, phenols, glycosides, and saponin also exist in the jackfruit leaves (Moke et al., 2017).

Fernando et al., (1991) reported the leaves possesses the antibacterial and antidiabetic properties. Several illnesses include high blood pressure, diabetes, cancers, anaemia, asthma, dermatosis, and diarrhoea can be cure from the *Artocarpus heterophyllus* leaves and stem barks (Fang et al., 2008). Moreover, ulcers and the wound can be cured by using the leaves of jackfruit (Khan et al., 2003). Besides, the production of collagen by the presence of vitamin C also able to strengthen the skin structure (Babitha et al., 2004). The properties such as anti-allergic, antibacterial, anti-inflammatory, antioxidant, antifungal and immunomodulatory in the plant make them applicable in curing fever, boils, wounds, skin diseases, and snake bite (Prakash et al., 2015).

According to Hari & Divya, (2014) cycloartenone, cycloartenol, β -sitosterol and tannins found in the leaves are beneficial to function as wound healing, antisyphilitic, and as an agent to destroy parasitic worms. Jagtap et al., (2011) state that several studies found that the leaves of jackfruit have the wound healing properties, anti-inflammatory and can act as cosmetic agent. Figure 2.11 shows the pharmacological activities of jackfruit plant.

Pharmacological Activities	Plant Parts	Extracts	Phytoconstituent
1 Anti-infective			
Anti-Fungal	Leaf, Seed	Alcoholic extract	Chalcone
Anti-viral	Seed	Alcoholic extract	Oxyresveratrol
Anti-bacterial	Bark of stem and root, leaves, fruit	Alcoholic extract	Isoprenyl Flavones
2 Anti-cancer	Wood	Alcoholic extract	Isoprenoid Flavonoids
3 Inflammatory diseases			
Anti-inflammatory	Fruit	Ethyl acetate extract	Flavonoids
Wound Healing	Leaf	Alcoholic extract	Lectin
4 Anti-Diabetic	Leaf	Aqueous extract	Prenyl Flavonoids
5 Miscellaneous			
Anti-oxidant	Fruit, Seed	Alcoholic and aqueous extract	Prenyl Flavonoids
Anti-malarial	Bark of Root and stem	Aerial part extract	Prenylated Flavones
Anti-diarhoeal	Bark of Root and stem	Artocarpusintegnifolia extract	Prenylated Flavones
Anti-carcinogenic	Leaf	Alcoholic extract	Flavonoids
Anti-platelets	Root, Seed	Alcoholic extract	Flavonoids
Anti-tubercular	Root, Stem	Dichloromethane extract	Prenylated Flavones
Anti-atherosclerosis	Fruit, Root	Ethyl acetate extract	Flavonoids
Anti-arthritis	Leaf, Fruit	Ethyl acetate extract	Flavonoids

Figure 2. 11: Pharmacological activities of jackfruit plant. (Arora & Parle, 2016)

2.3 Nanoemulsion

2.3.1 Introduction of Nanoemulsions

A Nanoemulsions (NE) is a mixture of oil, surfactant, and water with the characteristics of low viscosity, transparent, high water content products (Salim et al., 2012). NE has the properties such as small particle size and low interfacial tension. This is due to the presence of the combination of surfactants in the formulation (Heuschkel et al., 2008). According to Salim et al., (2012) NE contains oil, surfactant, and water in specific proportions. Thus, suitable oil and surfactant must be chosen in order to improve the topical permeation. To achieve desirable particle size, the ratio of emulsion component must be conducted properly to produce a stable NE. Thus, the stability of NE can be achieved by proper choice of system components, composition and preparation method (Cheng et al., 2014).

Several Nanosystems such as microemulsions (ME), nanoemulsions (NE), solid lipid nanoparticles (SLN), nanostructured lipid carriers (NLC), liposomes and vesicles are reported to enhance the skin permeation (Roberts et al., 2017). The advantage of Nanosystems is enhancing the solubility and bioavailability of the molecules. Besides, others characteristics of NE are it prevent the drugs from degradation under physiological condition and able to solubilize hydrophobic compound (Musa et al., 2013). Figure 2.12 shows the Nano emulsion appearance versus microemulsion appearance.

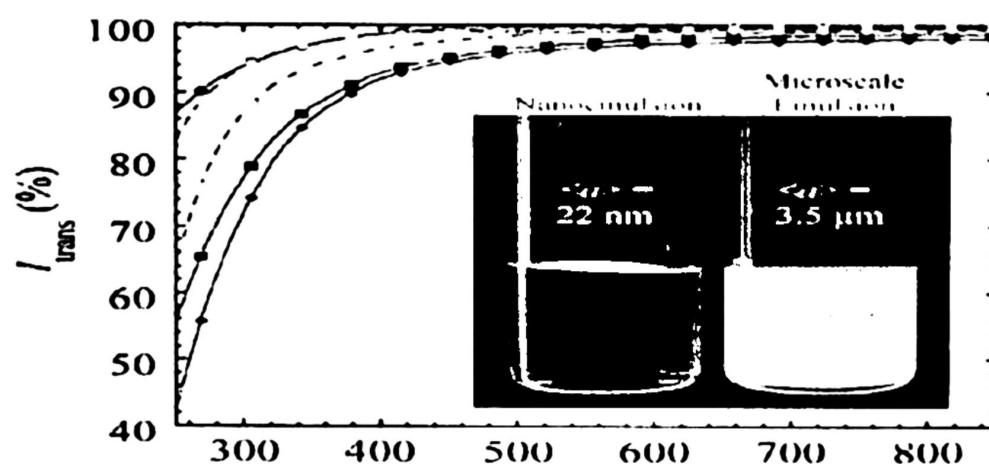


Figure 2. 12: Nanoemulsions appearance versus microemulsions

2.3.2 Nanoemulsions for Cosmetic Application

Cosmeceuticals are cosmetic products with biologically active ingredients which function to care and repair the skin condition (Gao et al., 2008). While Nano cosmetics are the term used for application of nanotechnology in the cosmetics field. There is various nanomaterial use in cosmetics such as liposomes, nanosomes and solid lipid nanoparticle (Raj et al., 2012). The instability and bioavailability of neutraceutical can be overcome by using NE as drug delivery systems. Therefore, there is a growing interest to use NE in the food, cosmetic and pharmaceutical industries (Peshkovsky et al., 2013). NE systems have been used widely in the field of dermatological, cosmetic and transdermal for skin delivery.

Rocha-Filho et al., (2017) found that NE able to support the penetration of active ingredients to the skin which increases the concentration in the skin compared to liposomes. The advantage of using NE in cosmetic is no irritation to the skin, high permeation ability and high drug-loading capacity (Salim et al., 2012). It also allows the effective transport of active ingredients to the skin (Arunkumar et al., 2011). Nastiti et al., (2017) state that the skin acts as the barrier to protect the body from penetration of any molecules from the surrounding and to prevent excessive loss of water from the body. Stratum corneum (SC) is the main skin barrier which constructed by layers of flattened corneocytes surrounded by lipid bilayers composed primarily of ceramides. Figure 2.13 shows the properties of Nanosystems determining skin absorption and potential routes of penetration.

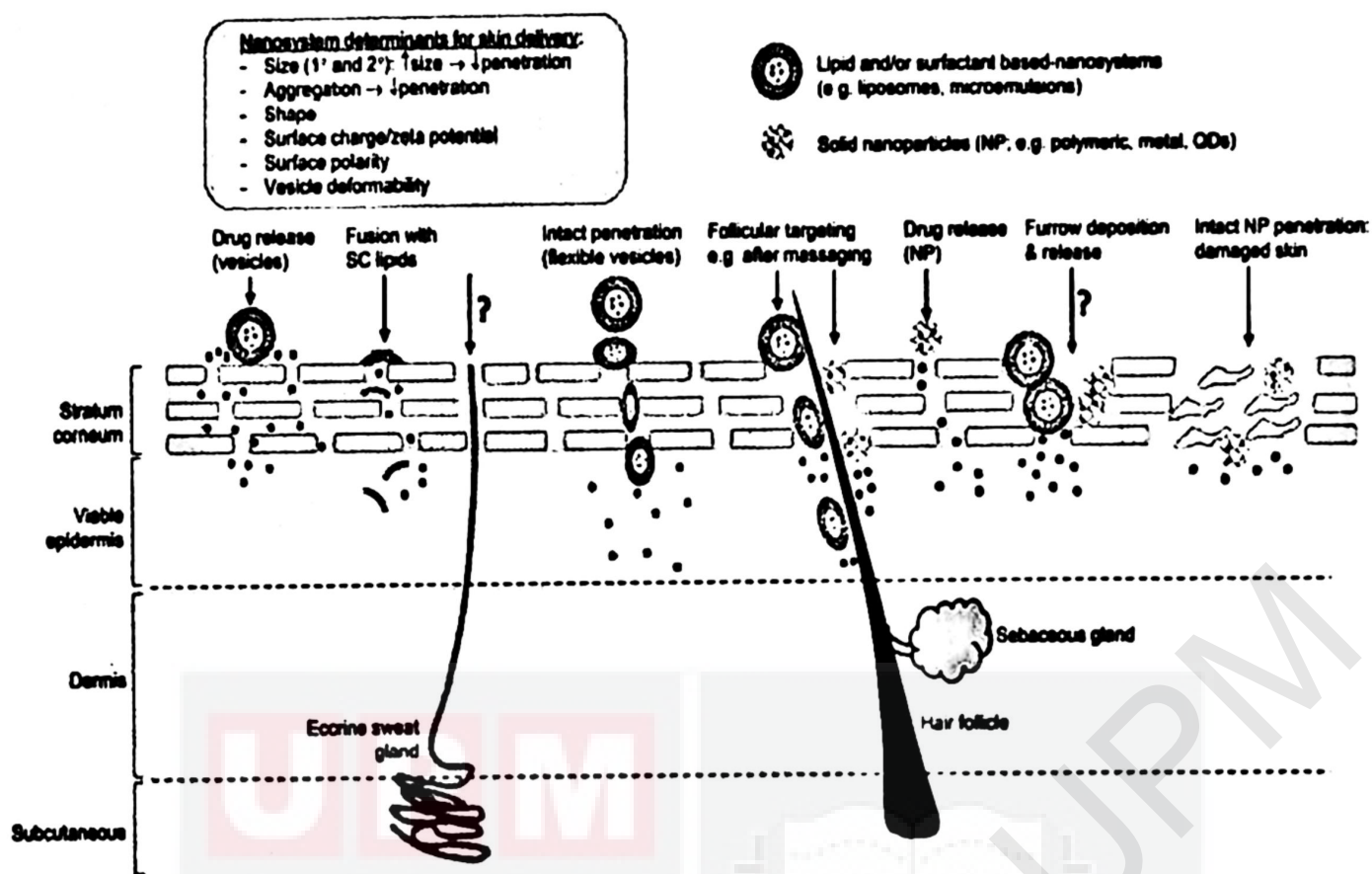


Figure 2. 13: Properties of Nanosystems determining skin absorption and potential routes of penetration. (Nastiti et al., 2017)

Since NE is less sensitive to temperature, pH changes, and dilution, it makes them suitable to use in food and cosmetic sectors (Gupta et al., 2016). The small size of NE ensures a close contact with the skin barrier. Thus, the amount of active compound penetrate to the skin layer can be maximized which make NE suitable for cosmetics (De Azevedo Ribeiro et al., 2015). Shah et al., (2010); Sharma et al., (2010) state that NE is suitable to be formulated as cream and moisturizer since there is no creaming and flocculation is observed which make them more attractive and stable product to be formulated as cosmetic. Moreover, according to L. Yang, (2017) the pleasant aesthetic properties and skin feel of the O/W Nano emulsion is due to its characteristic of the absence of thickener, good fluidity and the transparent nature.

Today, a majority of the cosmetics manufacturers are using nanomaterial in producing their products. The world's largest cosmetics company, L'Oreal investing its revenue about \$17 billion in producing their Nano-related product into the market (Özgün, 2013). Besides, Cosmetics giant Estee Lauder also entered the Nano-Market

in 2006 with a range of products containing “Nanoparticles”. Others manufacturers include Freeze 24/7, DDF (Doctor's Dermatologic Formula), and Colorescience (Raj et al., 2012). Figure 2.14 shows the ranking of companies that use Nano-related products.

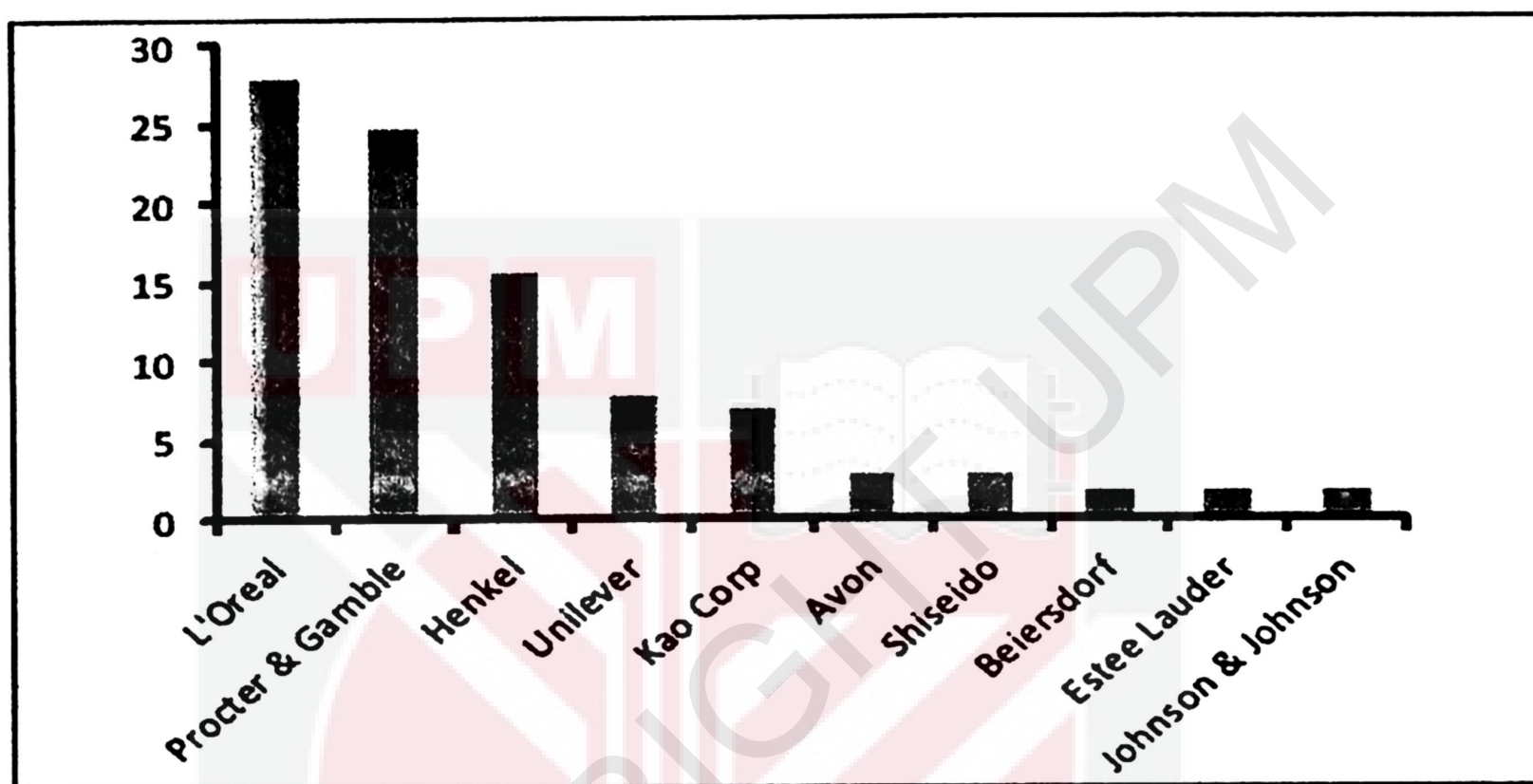


Figure 2. 14: Ranking of companies that use Nano-related products.

2.4 Selection of Oil Phase

2.4.1 Caprylic Capric Triglycerides

The oil phase function is to facilitate the dissolution of the bioactive compound. The components that can act as an oil phase are fatty acids, esters of fatty acids and alcohols, medium chain triglycerides (MCT), triacetin, terpenes and other penetration enhancers. They can be used as single components or mix together to be an oil phase (Nastiti et al., 2017). According to De Azevedo Ribeiro et al., (2015). The of oil phase must be chosen wisely as their properties such as molecular weight, type of chain and

polarity will affect our final products in term of oiliness, sensory properties, and spread ability.

According to Nandi et al., (2005) the by-products from hydrolysed triglycerides by re-esterification of glycerol and fatty acid distillates which is improved by fractional without additional processing will produce the Medium Chain Triglycerides (MCT). MCTs are used in most application because it has distinctive characteristics such as absorption ability, transport and metabolism ability that make it extraordinary than typical fats and oils (Medical & Applications, 1981). MCTs suitable to be incorporate in cosmetic and toiletries industries as they are able to function as a solubiliser and carrier for oil and water-soluble components.

The well-received ingredients to be used in the cosmetic and pharmaceutical application are triglycerides which is the ester. In skin care, they function as the moisturizer, solubiliser, improve penetration to skin and lipophilic vehicle (Jaworska et al., 2014). The example of triglycerides which is approved to be used as cosmetic ingredients are vegetable oil and artificial ester including Caprylic / Capric triglycerides. Caprylic / Capric triglycerides have are fully saturated triglycerides with exceptional soothing ability, easy to spread and prevent water loss from the skin. Besides, they are not toxic and non-skin irritant.

De Azevedo Ribeiro et al., (2015) further state that in formulating cosmetic products, the characteristic of dry, non-oily touch and skin conditioning properties make Caprylic / Capric Triglyceride (MCT), Ethylhexyl Palmitate and C12–15 Alkyl Benzoate often to be chosen in cosmetic ingredients. Schwarz et al., (1995) reported that MCT can successfully integrate into skin lipid more readily Long chain Triglycerides (LCT) which is less polar to the water. Hippalgaonkar et al., (2010) state that the advantages of using Medium chain triglycerides such as Caprylic/Capric

triglycerides are they are resistant to oxidation high and own good solubility properties.

As they are less reactive to chemically, they are often favourable to be used as oil phase in drug delivery system such as to transport ketoprofen (Paolino et al., 2002) and in NE of oil in water (O/W) containing lidocaine (Sadurní et al., 2005), also as based emulsions for danazol (Serajuddin, 2012). Several studies that have been conducted in obtaining NE for cosmetic formulation are limonene oil (Li & Chiang, 2012), sunflower oil (Leong et al., 2009) and medium - chain (Y. Yang et al., 2012).

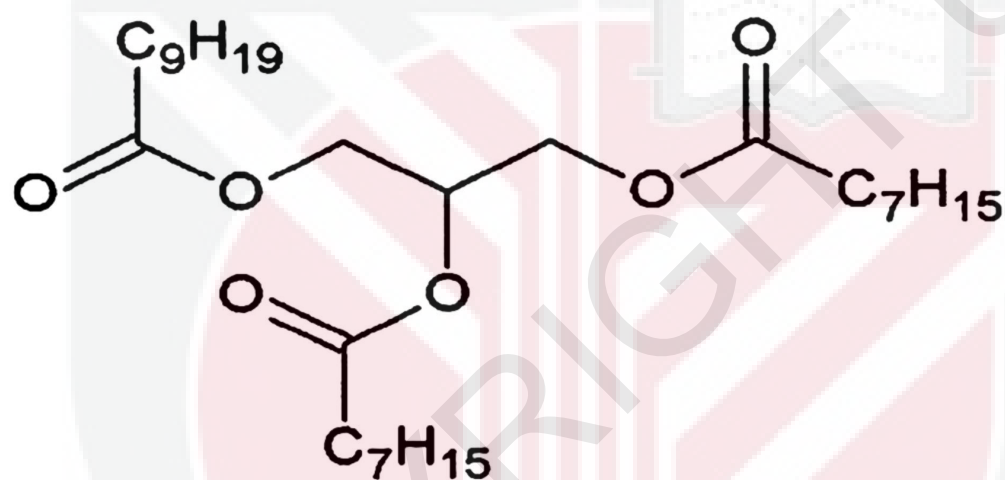


Figure 2. 15: Chemical structure of caprylic/capric triglyceride.
(Jaworska et al., 2014)

2.5 Selection of Surfactant

Surfactant is a group of substances that have the properties of good surface performance and have the availability to form the condensed interfacial film. According to Shahavi et al., (2015) surfactant is an amphiphilic molecule that has a polar region, which has a high affinity for water, and a nonpolar region, which has a high affinity to oil.

The use of the surfactant is vital in the formulation NE to produce a stable droplet size (Shahavi, Hosseini, Jahanshahi, Meyer, et al., 2015). Lane (2013) stated that surfactant has the function as skin penetration enhancer of the drugs. They were reported to improve the penetration of drugs by improving the skin permeability and solubility if they are present in the form of NE (Chen et al., 2012; Varshosaz et al., 2013). Selection of a suitable surfactant is an important aspect to be conducted in order to make sure that the active ingredients are chosen will effectively soluble for better permeation of the drugs (Fini et al., 2008; Javadzadeh & Hamishehkar, 2011).

Compare to ionic surfactant, non-ionic surfactants are considered safe to be used as cosmetic formulation as it is less irritating. A study by Nastiti et al., (2017) also found that non-ionic surfactants are able to ensure a minimum skin irritancy. According to Choi et al., (2014); Modi & Shelat, (2012) non-ionic surfactant are not sensitive to electrolyte concentration and pH as they have good biological acceptance properties. Besides, these surfactants are not harmful and known as biocompatible products because any alteration in the pH of the mixture will not affect them (Lv et al., 2014; Fini et al., 2008; Sakeena, et al., 2011). A Hydrophile-Lipophile Balance (HLB) value of more than 10 is needed in order to form NE when choosing the surfactant. The hydrophilic and lipophilic properties must be balanced in order to produce a stable NE (Shahavi, Hosseini, Jahanshahi, Meyer, et al., 2015). Hence, the right mixture of surfactant with a high and low value of HLB will help in the formation of a stable NE formulation (Azeem et al., 2009).

Apart from that, Vilasau et al. claimed that the stability of the emulsion system can be boosted up by using the mixture of surfactants. (Pey et al., 2006) also state that for a large number of applications, the use of surfactant mixture will lead to a better formulation of NE compare to when using an only pure surfactant. Various studied

that have been done reported that the emulsions with a mixture of surfactant will distribute and solubilize more readily into the continuous phase (Peng et al., 2010). Moreover, according to the study reported that the minimum size of NE can be achieved when we add another surfactant as it will provide better stability to the NE. Porras et al., (2004) state that mixtures of surfactants can provide better performance than pure surfactants from the result of various studies conducted previously.

A mixture of Tween 80 and Span 80 have the quality of high compatibility when it is being formulated with other materials and they are less toxic to be consumed compared to other surfactants. Span 80 has the properties of viscous, lipophilic and act as emulsifying liquid agent. However, Tween 80 is a subsidiary product of Span 80 which is hydrophilic in nature (Shahavi, Hosseini, Jahanshahi, Meyer, et al., 2015). Therefore, oil in water (O/W) interface layer and higher stability of NE can be achieved by using a mixture of Span 80 and Tween 80 as they possess an oleate-chain with 18 carbon atoms and one unsaturated bond. Which will provide more stability to the emulsion.

2.6 Ultrasound Method

Generally, there is two step in preparing NE which initially prepared the macroemulsion (ME) before NE can be produced which require high-energy or low-energy (Nastiti et al., 2017). The high energy method involves high-shear stirring using ultrasonication, a high-pressure homogenizer, microfluidization, and membrane emulsification. Besides, Musa et al., (2013) state that the high energy method uses the mechanical device to generate the force needed to break the oil and water phase which result in smaller oil droplet to be produced. The low-energy methods are phase inversion temperature (PIT), phase inversion composition (PIC) and spontaneous emulsification (Solans, Izquierdo, Nolla, Azemar, et al., 2005).

One of the method in NE preparation for pharmaceutical is ultrasound since the maintenance and production cost is minimized (Sivakumar et al., 2014). Recently, the high energy method which is ultrasound is widely being reported in the literature for cosmetic field involving oil in water (O/W) compare to water in oil (W/O). This method is based on the cavitation mechanism. The interfacial area of the droplet increase due to the collapse of cavitation bubble in the emulsion. From Yukuyama et al., (2016) studies found that once the optimum limit achieved, the sonication time no longer affects the droplet size. According to Nakabayashi et al., (2011) NE is produced from the agitation of vibration at the ultrasonic frequency which the particle size is broken down from the emulsion. Peshkovsky et al., (2013) further explained that ultrasound method possesses the advantages such as easier cleaning and servicing and lower cost compared to the homogenizer. A low frequency from the ultrasound results in droplet disruption due to the effect of cavitation (Mehmood et al., 2017).

The important parameter in ultrasound method is the time of sonication as it will affect the droplet size and surfactant rate of adsorption (Li & Chiang, 2012). As we increase the sonication time, the smaller size of NE tends to form. The optimum condition obtained by using ultrasound method is 3 min (Anarjan et al., 2010). A study by Hassan et al., (2015) on clove oil NE found that at 300 s sonication time, the size of 100 nm was obtained. A study by Silva et al., (2013) in producing sunscreen by using ultrasound method with avocado oil and non-ionic surfactant successfully obtained NE with a size of 6 to 10 nm. Studies by Kentish et al., (2008) obtained NE with droplet size lower than 135 nm by using ultrasound method at 20- 24 kHz. Then, NE with a size of about 200 nm was obtained by sonication at 30 min which consists of mixed surfactants which are Span 80 and Tween (Manchun et al., 2014). Sugumar et al., (2014) also reported that at 30 min of ultrasonication of eucalyptus oil, NE with

a size of 3.8 nm was obtained. However, Özgün, (2013) reported ultrasound method only can be used to prepare small batches of NE which is at laboratory scale. Figure 16 shows the representative nanoemulsions of water-insoluble drugs that are generated by ultrasonication while Figure 2.17 shows the nanoemulsions process mechanism, advantages, and, disadvantages.

Drug and route of administration	Dosage	Oil	Surfactant	Co-surfactant/ solvent	Indication	Mean droplet diameter (nm)	Stability
Saquinavir (SQV) (oral)	400 µg/ml	Flaxseed oil	Lipoid E80	Deoxycholic acid	HIV/AIDS	218.0 ± 13.9	2 months (stored at 4 °C in the dark place)
Bufadienolides (mBU) (l.v.)	0.28 mg/ml	Soybean oil	Lipoid E80, pluronic F68, Tween 80	Sodium oleate, glycerine	Antitumor analgesic	43.5 ± 13.8	3 months (stored in a dessicator with silica-gel at 25 °C)
Paclitaxel (oral)		Pine nut oil	Egg lecithin	Stearylamine, deoxycholic acid	Anticancer	90-120	
Granisetron (GRN)(oral)	0.4 mg/ml	Isopropyl myristate, lauroglycol 90	Lipoid E80	Pluronic F68, hydroxypropyl-β-cyclodextrin, polyvinylpyrrolidone K25	Antiemetic	50	3 months (stored at 4 °C in the dark place)
2-(Butylamino)-1-phenyl-1-ethanethiosulfuric acid (BphEA) (l.n.)	10 mg	Medium chain triglyceride	Tween 80, Span 80, stearylamine	Ethanol, glycerol	Schistosomiasis	225.8 ± 32.1	3 months (stored at 4 °C in the dark place)
Imipramine /doxepin (topical)	3% w/w	D-limonene	Labrasol, pluroleique	Propylene glycol, transcutole, oleic acid, isostearyl isostearate	Analgesic anti-allodynic	18-20	3 months (stored at room temperature)
Fisetin (Lp)	36.6 mg/kg	Miglyol 812	Tween 80, labrasol	Lipoid E80	Antitumor	153 ± 2	1 month (stored at 4 °C in the dark place)

Figure 2. 16: Representative nanoemulsions of water-insoluble drugs that are generated by ultrasonication. (Sivakumar et al., 2014).

Nanoemulsion process	Operation principles	Advantages	Disadvantages
High energy	Ultrasound generators	Cavitation mechanism	Limited to small batches
	High-pressure homogenization	Shear, collision and cavitation mechanism	More expensive than other high-energy equipment; more flexible on surfactant and internal structure selection than low-energy process
	Microfluidization	High-pressure injection pump	More flexible on surfactant and internal structure selection than low-energy process; low process time
Low energy	Phase inversion composition	Changing of the interfacial film curvature by progressive dilution of the dispersed phase	High cost; not recommended for thermo- or shear-sensitive compounds
	Phase inversion temperature	Changing of the interfacial film curvature by temperature variation	High cost; not recommended for large-scale production
	Spontaneous emulsification	Dispersion and condensation mechanism	Requires gradual addition of one phase into another; requires the presence of liquid crystal (LC) or mid-range microemulsion (ME) phases

Figure 2. 17: Nanoemulsions process mechanism, advantages and disadvantages. (Yukuyama et al., 2016).

2.7 Physical Properties of Nanoemulsions

The quality of the NE formulated by ultrasound method is measured in the term of its physical measurement. The measurement includes particle size, viscosity, pH, turbidity and the stability of NE.

2.7.1 Particle Size

Particle size is an important parameter to define application and stability (De Azevedo Ribeiro et al., 2015). (Gadhawe, 2014; Ghotbi, Khatibzadeh, & Kordbacheh, 2014; Solans, Izquierdo, Nolla, & Azemar, et al., 2005) state that NE is an emulsion that is kinetically stable with a mean droplet size of 20- 200 nm. NE possesses a smaller droplet size of ($< 1\mu\text{m}$) which provide a greater kinetic stability and transparent appearance (Gupta *et al.*, 2016). Shah et al., (2010) also reported that the suitable droplet sizes for NE is in the range of 20–200 nm. Based on my reading, according to Article, (2011); Gutiérrez et al., (2008) NE are emulsions with a very small droplet size 20 to 200 nm in range. Apart from that, a study done by (Gupta et al., 2016) found that NE is kinetically stable liquid-in-liquid dispersions with droplet sizes on the order of 100 nm. The smaller droplet size of emulsions prevents the coalescence to occur in the emulsion and support the penetration of the active agents.

2.7.2 Viscosity

An important parameter that will affect the stability and drug release of nanoemulsions and microemulsions formulations is the viscosity (Nastiti et al., 2017). NE has a very low viscosity (Devarajan & Ravichandran, 2011; Rao & McClements, 2011). Moreover, NE possesses the characteristics such as longer colloidal stability and low viscosity. Che Sulaiman et al., (2016) state that NE with a low viscosity is very suitable to be used in the pharmaceutical and cosmetic application. According to

Date et al., (2010) the low viscosity NE with translucent appearance give a good pleasure to the skin during its application.

However, when the viscosity increased, the droplet size in emulsion will be increased (Teo et al., 2010). The emulsion stability also can be achieved if the viscosity is increased as it will inhibit the movement of the droplet. Montenegro et al., (2015) reported that there is a correlation between the viscosity and the ability of the formulation to be spread during application. A cream with a lower viscosity will reduce the surface tension of the cream. Thus, the spread ability performance of the skin will improve. Although from the research found that viscosity is an important parameter in stability and efficiency of the bioactive penetration, the particle size of nanoemulsions also must be minimized (Tsai et al., 2011).

2.7.3 pH

pH is the measurement of H⁺ ion activity. The natural pH of the skin surface in the development of skin care products is vital in the field of cosmetics and dermatology. According to Bernardi et al., (2011) the quality of final product might be effect by the chemical reaction due to pH changes. Therefore, it is important to monitor the pH value of the Nano emulsion (NE). Besides, the acceptable pH for NE for skin formulation is around pH 5.4 to 6.9 which is considered to be non-skin irritating and suitable for topical administration (Borges et al., 2013). In the cosmetic formulation, the products that are too acidic and too alkaline should be avoided to prevent irritation to the skin. Mahdi, Noor, et al., (2011) state that skin irritation could occur and exposure to bacterial infection could occur to the skin if the skin care products are too alkaline. Therefore, it is best to have cosmetic products formulated with a pH about 7.0 which is close to the pH of the skin.

2.7.4 Turbidity

Turbidity is the measure of cloudiness in the water. McClements, (2011) states that the nanoemulsions (NE) has a very high stability and low turbidity. Thus, it is very suitable for lipophilic active ingredients to blend together. After a certain time, the transparency of nanoemulsions become turbid due to increase in the droplet size (Sharma et al., 2010). One of the methods to measure turbidity is by using a spectrophotometer to measure the absorbance of the sample (Ghosh et al., 2013). Apart from that, Radhika & Guruprasad, (2016) used turbidimeter in their study to measure the turbidity in the sample.

2.7.5 Stability Studies

One of the significant aspects of NE system is stability as they have small droplet size and large surface area. Stability against sedimentation and creaming can be achieved due to the small droplet size of NE (Article, 2011; Gadhave, 2014). According to Sonnevile-Aubrun et al., (2004b) the shelf life of the product can be prolonged and provide better stability due to its small particle size. The shelf life of the Nano emulsion can be improved with a stability ranging from months to years.

According to De Azevedo Ribeiro et al., (2015) the appearance of the formulation, physical properties, and stability are the important parameter to be considered in cosmetic products development. Hence, the stability studies include the analysis of the particle size, viscosity, pH and turbidity of the NE will be further conducted in this study.

CHAPTER 3

METHODOLOGY

3.1 Introduction

This chapter will describe in details on the method and materials used in conducting the experimental work. The materials used will be described in section 3.2 while the extraction of jackfruit leaves will be described in section 3.3 and section 3.4. Section 3.5 and section 3.6 will be on the preparation of emulsion and nanoemulsion respectively. In the meanwhile, Section 3.7 and Section 3.8 will describe the verification of the model and Analysis of physicochemical properties respectively. The summary of the process design is illustrated in the Figure 3.4.

3.2 Material of Experiment

Jackfruit leaves were obtained from (Maran, Pahang, Malaysia). Caprylic/Capric triglycerides (MCT) was supplied by KLK Oleo Klang (Selangor, Malaysia). Tween 80 (Polyoxyethylene Sorbitan Monoleate) and Span 80 (Sorbitan Monoleate) were purchased from R & M Chemical (Selangor, Malaysia). Ethanol Solute was purchased from John Kollin Corporation (Selangor, Malaysia). Distilled water was obtained from Bioreactor Lab Universiti Putra Malaysia.

3.3 Extraction by Maceration Method.

Fresh leaves of Jackfruit were oven dried and ground into fine powder. Then, soaked in the ethanol solvent and left overnight in the incubator shaker (Certomat IS) at 200 rpm at approximately 27 °C. A study done by Anwar & Przybylski, (2012) found that when 80% ethanol was used for extraction of flaxseed, the highest amount of flavonoids and antioxidant capacity was obtained compare to when using methanol as the solvent. Therefore, in this study ethanol was chosen as the solvent for the extraction process. The extracts were filtered, and concentrated by using a rotary evaporator (IKA HB 10, US) at approximately 50 °C and stored at -20 °C prior to further analysis (Che Sulaiman et al., 2016b)



Figure 3. 1: leaves powder and ethanol in incubator shaker



Figure 3. 2: Filtration of obtaining the extract



Figure 3. 3: Evaporation of leaves filtrate

3.4 Solubility of Extract.

Extract of Jackfruit leaves (JLE) at 0.025% w/w was added to the oil phase. The mixture was stirred in a water bath at 50°C until all the extracts were dissolved and a transparent mixture of oil and extract was observed. Another 0.025% (w/w) JLE was added to the previous MCT-extract mixture until a clear transparent oil mixture was formed. The step of extract addition was repeated until a non-transparent and saturated form MCT-extract mixture was observed. The total percentage of extracts that were successfully dissolved in the clear transparent MCT will be used in the formulation of Nanoemulsion (Musa et al., 2017).

3.5 Preparation of Emulsion

The experimental central composite design (CCD) was used to study the effect of three variable components of the JL extract nanoemulsions: water (A); oil to surfactant ratio (o/s ratio) (B); and sonication time (C), on the response variable: the particle size (Y). The constraints of variables proportions and the CCD obtained from

the software are presented in Table 3.1 and Table 3.2 respectively. The design was generated by using Design Expert Software 11 (Stat. Ease Inc., Minneapolis, USA).

Table 3. 1: Constraints of variables proportions

Types of Variables	Element	Level Value	Unit
Independent	Water	70 to 90	%
Independent	O/S ratio	0.71 to 1.15	-
Independent	Sonication time	5 to 25	min
Dependent	Particle size	20 to 200	nm

Table 3. 2: CCD design obtained from Design Expert software

Formulation	A:water (%)	B:O/S	C:Sonication time(min)
1	80	0.93	15
2	70	0.93	15
3	85	0.82	20
4	80	0.93	15
5	75	1.04	20
6	85	1.04	20
7	80	0.93	15
8	80	0.93	15
9	80	0.93	25
10	80	0.93	5
11	80	0.93	15
12	80	0.93	15
13	85	1.04	10
14	75	1.04	10
15	75	0.82	20
16	75	0.82	10
17	85	0.82	10
18	90	0.93	15
19	80	1.15	15
20	80	0.71	15

Based on the results of CCD design generated by the software, 20 formulations of oil in water (O/W) nanoemulsions will be prepared using Caprylic/Capric triglycerides (MCT) as the oil phase. Then, Tween 80/Span 80 with the ratio of (9:1),

and distilled water as the aqueous phase. MCT and non-ionic surfactants at various ratios were heated gently at 50°C and stirred with a magnetic stirrer at 350 rpm for 15 minutes. The oil phase was added dropwise from a burette at a rate of 1 mL/min into the aqueous phase. After complete addition of oil phase the emulsions was continuously mixed using a magnetic stirrer at 350 rpm for 15 minutes with the aid of spatula in order to overcome the liquid crystalline phase (Mahdi et al., 2011).

3.6 Preparation of Nanoemulsions: Ultrasonic Method

The previous emulsion will be subjected to high energy method in order to produce the emulsions which are in nanosized. The emulsions will be subjected to 20 kHz Sonicator Q500 (Qsonica, USA) with a maximum power output of 500 W. This simple bench-top ultrasonic device consisted of an electrical generator, a transducer and a titanium sonotrode (horn). The mechanical ultrasonic vibrations of sonotrode with 40% amplitude will be used. Then, sonication at the different time which are (5, 10, 15, 20, and 25 min) will be carried out. The cycle of the sonication consisted of 30 s pulses on and 30 s pulses off (Sugumar et al., 2014).

3.7 Verification of the Model

Analysis of variance (ANOVA) was performed to determine the significant differences between the independent variables. Then, three random formulations will be prepared to validate the prediction by the model (Che Sulaiman et al., 2016b). The percentage of the residual standard error (RSE) was calculated.

3.8 Analysis of Nanoemulsion Physicochemical Properties

This section will describe the analysis that will be conducted to the developed nanoemulsions. The physicochemical properties in term of the particle size, viscosity, pH and the turbidity of the developed nanoemulsions will be measured. Apart from that, the stability test for the nanoemulsion at the different temperature during storage period will be analysed.

3.8.1 Particle Size

The particle size of the nanoemulsions were measured by using the particle size analyzer (Nanoplus Zeta/ Nano particle Analyzer, Japan). The sample were diluted (1:50) with distilled water before being filled into a cuvette capillary cell to avoid multiple scattering effects. Measurements were performed with an angle of 165° at room temperature (28 ± 0.5 °C). The measurements were carried out in fully automatic mode, with each sample will be analyzed once and each analysis consisting of three replicates (Che Sulaiman et al., 2016b).

3.8.2 Viscosity

The viscosity of nanoemulsions will be determined by using rheometer (AR-G2 Rheometer). The sample will be carried out at room temperature with 1°/ 60 mm cone and plate geometry gap 0.030 mm. The reading of the viscometer will be note.

3.8.3 pH Determination

The pH measurement of the optimal nanoemulsions was performed at room temperature (28 ± 0.5 °C) by using a Sartorius Pb-10 Basic pH Meter. The pH Meter was first being standardized by using buffer solution of acidic and basic value Of 7.00

and 4.00. The electrode was dipped into the buffer solution to calibrate the pH Meter. Before being into the nanoemulsion solution, the electrode was rinsed with distilled water. The pH readings were obtained by dipping the electrode into the nanoemulsion and the measurements were taken when the reading is stable. The pH measurement is important to assess whether the sample is compatible with human skin (Che Sulaiman et al., 2016).

3.8.4 Turbidity

The turbidity of nanoemulsions will be measured by using a Hach 2100N Laboratory Turbidity Meter (Hach, USA). The sample were diluted (1:50) with distilled water before being measured. The experiment is carried out triplicate and the mean is calculated.

3.8.5 Stability Test

The stability study for optimal nanoemulsions involved the following: stability under storage at room temperature and 45 °C; to determine the change in size, pH, viscosity, turbidity (Che Sulaiman et al., 2016b). The measurement of the nanoemulsions will be conducted in every 2 weeks for two month storage period. Besides, the changes of the nanoemulsions appearance will be observed in order to determine the condition of the nanoemulsion during storage period.

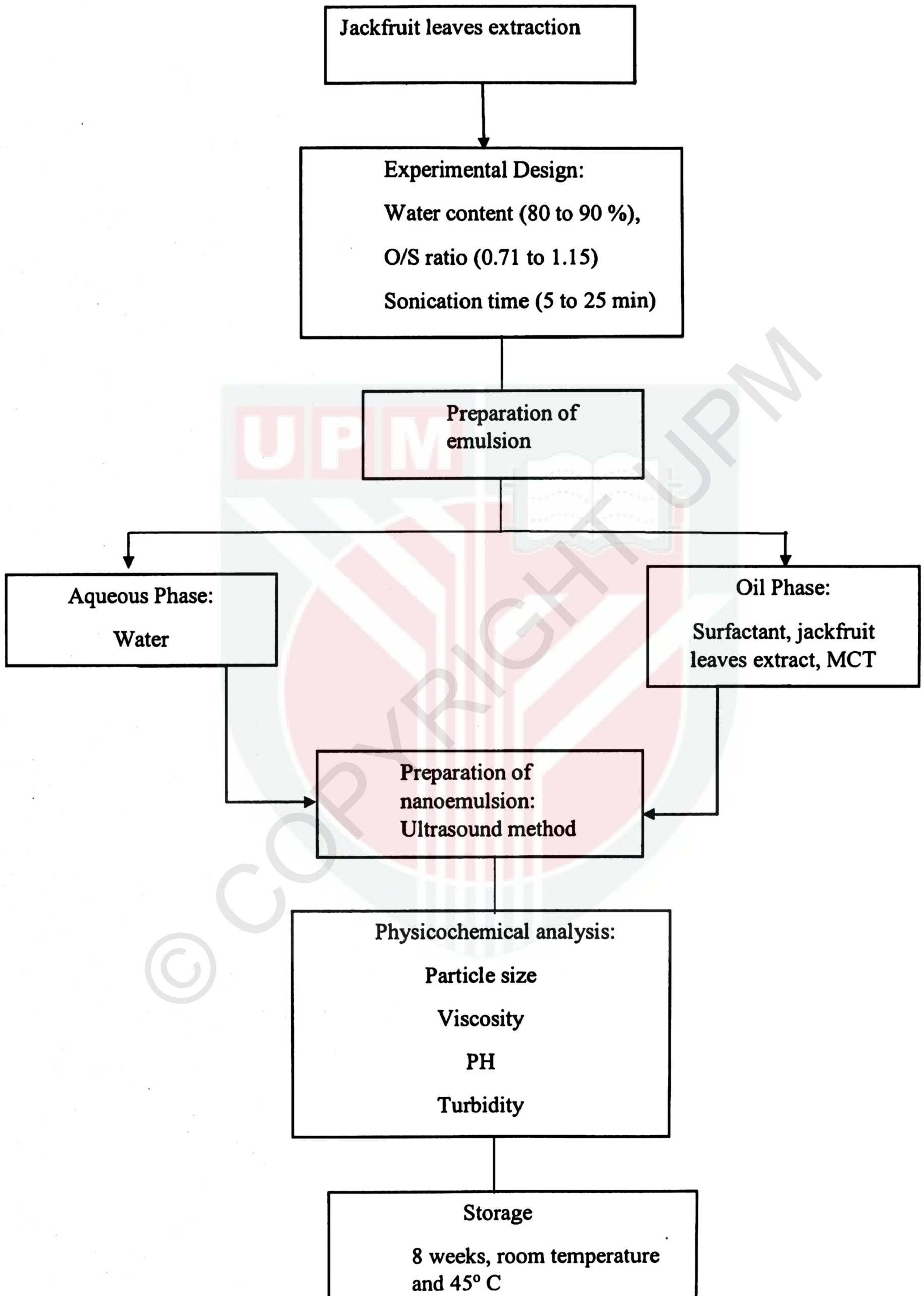


Figure 3. 4 : Summary of process design

CHAPTER 4

RESULT AND DISCUSSION

4.1 Introduction

This chapter discusses the finding of this research. The first section of this chapter (Section 4.2) is to analyse the solubility of jackfruit leave extract. In Section 4.3 is to investigate the experimental design used in order to find the optimum condition to formulate nanoemulsions that were generated by using Design Expert Software 11. In section 4.4 the change of physicochemical properties of the developed nanoemulsions during 8 weeks of storage time at room temperature and 45 °C is discussed. Section 4.5 analysed the stability of the nanoemulsions during 8 weeks of storage time. The data obtained from this study provide useful information on the best formulation can be used to design and establish processing method of nanoloaded jackfruit leave emulsion.

4.2 Solubility of Jackfruit Leaves Extract

Oil phase will act as the main medium in transferring the bioactive ingredient to the skin targeted area. Therefore, the extract carrying the bioactive ingredient must totally soluble in the oil phase to ensure that the efficiency of application to the skin

can be maximized. In this study, the jackfruit leaves extract (JLE) solubilise in the oil phase completely without any precipitation is observed in the mixture. Therefore, the jackfruit leaves extract can be incorporated in the nanoemulsion formulation with the oil phase.

4.3 Statistical Analysis

The collected data from the nanoemulsions developed were used for further statistical analysis of Response Surface Methodology and optimization of nanoemulsion formulation in the Design Expert 11 Software. Analysis of variance (ANOVA) was used for the statistical analysis.

4.3.1 Analysis of Variance (ANOVA)

Analysis of variance (ANOVA) was performed in order to check the compatibility and significance of the model generated from the software. This parameter used will form an empirical model that minimize the particle size and optimize each of the independent variables.

Central Composite Design (CCD) was used for Response Surface Methodology. From the software, 20 run of different compositions and condition was obtained. The experimental data A, B and C were inserted according to the number of run. Using the Design expert software, a quadratic model was the best fit compare to others model such as cubic, linear, and quartic model.

. The results of ANOVA for the effect of the three variables were shown in Table 4.1. Each of the linear components (A, B, and C) influenced the responses with respect to their evaluation of the coefficient and P-value of the linear mixture. The computed F-value and P-value of the model are 5.7 and 0.0045 respectively. The F-

value this large implies that there is only 0.45 % chance that it could be due to noise. In another hand, P-value less than 0.05 showing that the model design was significant. In this case, the variable B, AB, and C are significant model terms which any variations of this parameter will affect our desired response. For model term A and C, it cannot be eliminated from the model although its P-value is greater than 0.05, as it is required to support the hierarchy for the significant term.

In addition, the lack of fit value and P-value were 47.99 and 0.0003 is relatively due to pure error and noise while generating the result. This result shows that the predicted and experimental value revealed a good correspondence between them although the lack of fit value is quite high due to some noise and the model developed can be used to adequately describe the relationship between the responses and to predict the particle size of optimal jackfruit leaves extract nanoemulsions.

Table 4. 1: ANOVA of the fitted linear equation for particle size of nanoemulsions

Source	Sum of Squares	DF	Mean square	F-Value	p- Value
Model	464.50	5	92.90	5.70	0.0045*
A	1.44	1	1.44	0.08	0.7707***
B	125.44	1	125.44	7.69	0.0149*
C	11.56	1	11.56	0.71	0.4140***
C²	228.06	1	228.06	13.98	0.0022*
AB	98.00	1	98.00	6.01	0.0280*
Residual	228.06	14	16.31	-	-
Lack of Fit	225.74	9	25.08	47.99	0.0003*
Pure error	2.61	5	0.52	-	-
Cor Total	692.85	19			
R- squared	= 0.6704				
Adj. R- squared	= 0.5527				

*p-value <0.05 : Significant

0.05 < p-value ** <0.1 : marginally significant

P- Value *** > 0.1 : Not significant

Df : Degree of freedom

The model to predict the response (particle size) obtained is shown in the equation below. The predicted equation is generate to fit the data into the model for the desired optimization. The Design Expert 11 use the function to generate three-dimensional response surface plots by keep one response at its optimal level and plotting against three independent variables.

$$Y = 115.6 + 0.30A + 2.80B - 0.85C + 0.126B^2 - 2.90C^2 + 3.50AB \quad (4.1)$$

Where Y = Particle size (nm)

A = Water content (%)

B = O/S ratio

C = Sonication time (min)

The equation 4.1 was attained from the software in order to confirm the fitness of the model. In order to verify the model, ratio of R-squared (R^2) and Adj R- squares (Adj-R^2) are evaluated. From the study, the obtained R^2 and Adj-R^2 are 0.6704 and 0.5527 respectively. This high values explained the precision of the predicted data versus experimental data. Thus, the quadratic model can be considered acceptable in analysing the effect of independent variables on the minimization of the nanoemulsion particle size.

Overall, the mathematical model expressed in ANOVA is valid and acceptable to be applied in the fabrication of nano loaded jackfruit leave emulsion associated with the varying condition of water content (%), O/S ratio, and sonication time (min).

4.3.2 Optimization of Nanoemulsion Formulation

Response Surface Methodology (RSM) is used to find the optimal condition for the whole variable and find the relationship between them. Shahavi et al, (2015) in his study use modelling for prediction in order to generate response surface plot.

Therefore, in order to study the effect of each dependant and independent variable on the particle size of jackfruit leaves extract nanoemulsions, three dimensional (3D) plot were generated by using Design expert 11 Software. To achieve this purpose, two of the independent variables were varied within the experimental range while another variable is held constant at a central point.

As illustrated in figure 4.1, the 3D surface plots show the effect of water content and O/S ratio on the particles size of the nanoemulsions. With the increasing water content and increasing O/S ratio, it can be seen that the particle size is the biggest (123.5 nm). Li & Chiang, (2012) claimed this is due to the reason that the rigid film of the nanoemulsions is destroyed by the increasing water content and leads to coalescence to occur thus increases in particle size of the droplet.

Meanwhile, at O/S ratio of 0.93, the minimum particle size is achieved although the water content is high (more than 75 %). Musa et al., (2017) reported increasing surfactant will decrease the particle size. The smaller particle size is achieved as the higher amount of surfactant will prevent the oil droplet surface from sticking to each other. However, when the O/S is further increases the result we obtained are bigger particle size. The increasing O/S ratios cause the oil and water content receive an insufficient concentration of surfactant for the nanoemulsions to construct. Therefore, the low surfactant concentration unable to diffuse with the newly form droplet and it causes the oil droplet to merge together which contribute to the formation of larger particle size. Li & Chiang, (2012) also state that total droplet surface area, the rate of droplet dispersion, and coalescence can be controlled by the surfactant concentration. Besides, a larger particle size also could occur due to increase in the oil content of the system. According to Musa et al., (2013) larger oil droplets are

formed when the oil inhibit the flow resistance of the nanoemulsions which restricts the particle to breakdown during the formation of the system.

Thus, from figure 4.1, the area where minimum particle size of nanoemulsion obtained was at water content ~ 80 - 90 % and O/S ratio ~ 0.90 – 1.15.

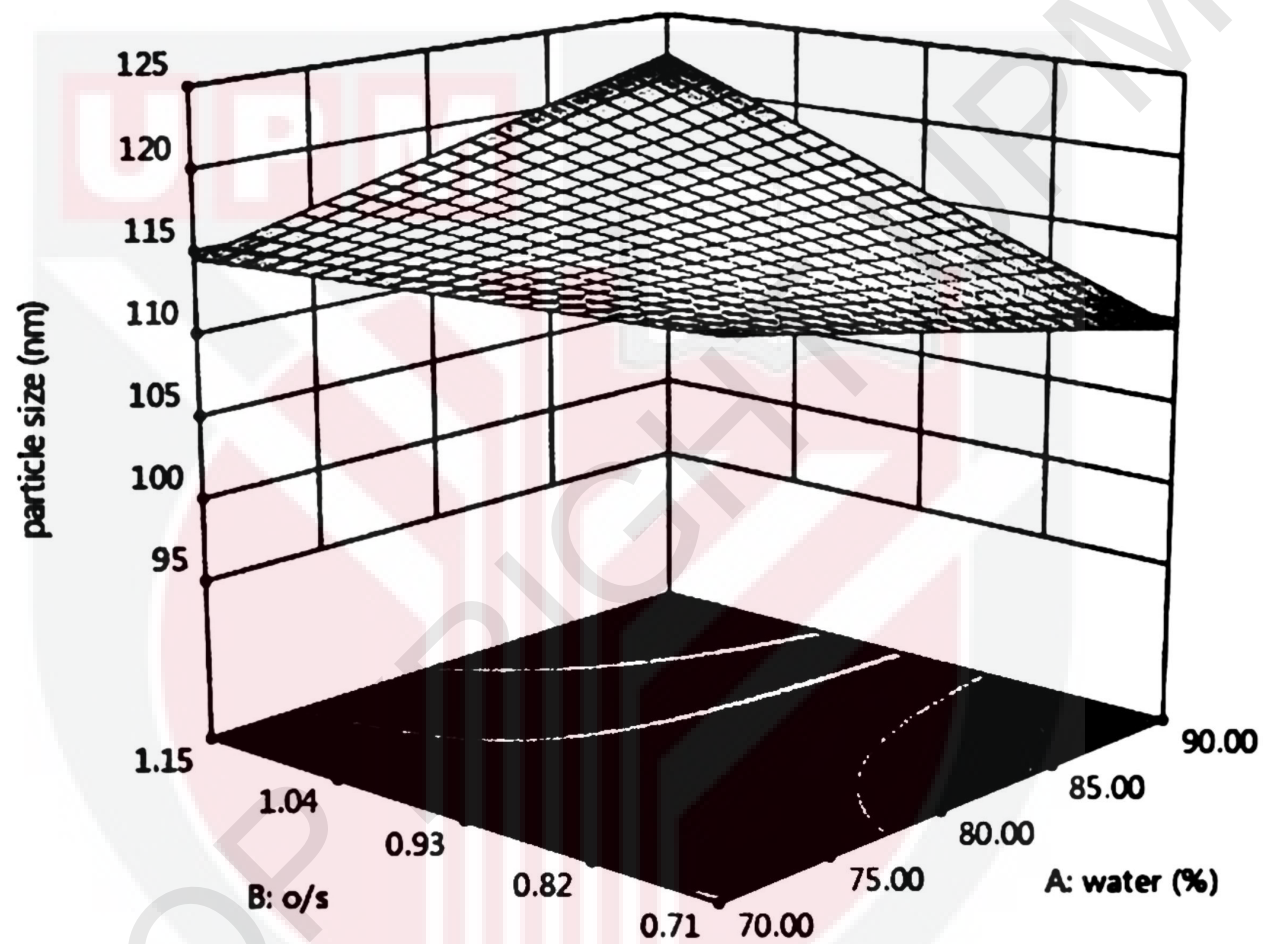


Figure 4. 1: Response surface plot showing the effect of O/S ratio and water content towards particle size

The plots in figure 4.2, shows the relationship of water content and sonication time towards the particles size of nano loaded jackfruit leave emulsion. Carpenter & Saharan, (2017) define sonication time as the time when the emulsion experience intense turbulence condition from the acoustic field. From the results, the largest particle size was obtained when the water content and sonication time is at 85 % and 10 minute respectively. A Similar result was found from the study done by (Rezaee et al., 2014) which stated that particle size increase with increases water content. This is

due to the reason that the rigid film of the nanoemulsions is damaged and leads to coalescence to occur thus increases in particle size of the developed nanoemulsion.

However, when the sonication time increases from 20 minutes to 25 minutes the particle size decreases gradually although the water content is about 80%. Ghosh et al., (2013) states particle size and stability of emulsion correspond with the sonication time. Thus, when the sonication time increases, smaller particle size achieved. Thus, the stability of the nanoemulsion can be improved. The result obtained match the result from their study as it was observed that by increasing the sonication time to 15 minutes, the particle size was decreasing compare to at 5 minutes. A similar trend was obtained in sunflower oil nanoemulsions formulation reported by Leong et al., (2009). According to Carpenter & Saharan, (2017), the stability of the emulsion also improved at higher sonication time. This is due to the reason that the probability of coalescence to take place was lessen as more energy are transferred to the system.

Thus, from figure 4.2, the area where minimum particle size of nanoemulsion obtained was at water content ~ 70 - 90 % and sonication time ~ 20 – 25 minute.

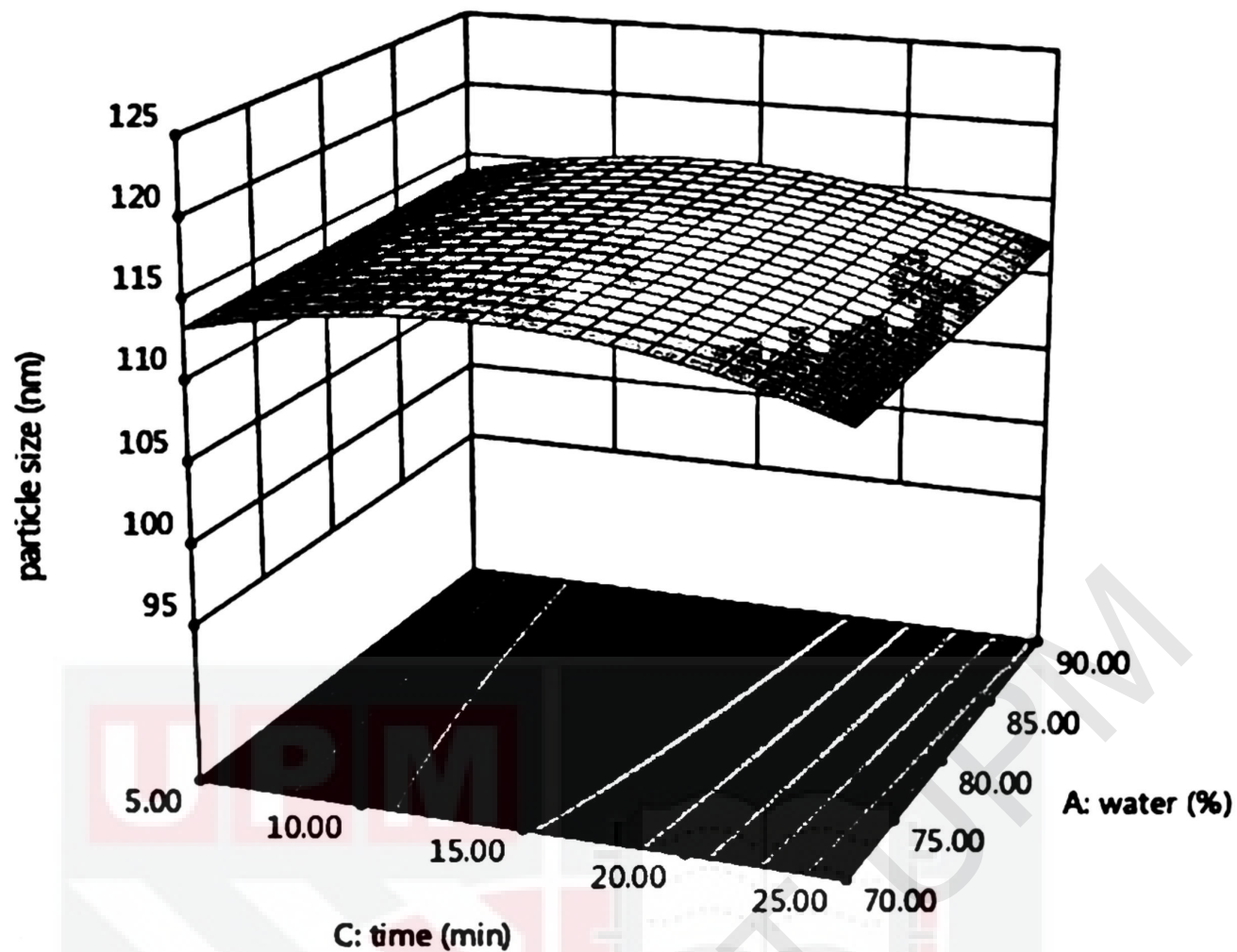


Figure 4. 2: Response surface plot showing the effect of water content and sonication time towards particle size

4.3.3 Validation of Responses

An optimum formulation of jackfruit leaves extracts nanoemulsions was determined to contain optimum levels of three independent variable (water, oil to surfactant ratio, and sonication time) with minimum particle size. Bioactive agent in the nanoemulsions can be more effective if the particle size is the smallest to obtain successful delivery of active ingredients through the skin barrier (Teo et al., 2010). Due to a large surface area, the nano-sizes system can offer rapid penetration and deposit uniformly on the substrate (Tadros et al., 2004). Since at higher surfactant level cause the particle size to increase, we want to minimize the amount of surfactant use. Thus, in the optimum formulation, we would have the surfactant at a minimum. This statement is supported by the study done by Musa et al., (2017) which found that high level of surfactant will expose the skin to irritation and increases cost of production.

Our goal is to find the optimum condition where we obtain a minimum particle size in the range of 99.6 – 123.5 nm. Table 4.2 shows predicted and actual response for random optimized formulation. The optimum condition obtained for jackfruit leaves extract nanoemulsions are 80.00 % water content, 0.93 O/S, and 25.0 min sonication time. Under the optimum composition, the predicted average particle size is 102.4 nm where the actual size obtain from experiment is much smaller (99.6 nm) with a residual standard error (RSE) of 2.73 %. The actual response showed that the model is in good agreement with the predicted value as the RSE is very small. Therefore, we can conclude that this model are reliable to be used in order to formulate the nanoemulsions.

Table 4. 2: Predicted and actual response for optimized formulation

Water (%)	O/S	Sonication time	Actual Particle size (nm)	Predicted Particle size (nm)	RSE (%)
80.00	0.93	15.00	118.6	116.3	1.98
85.00	0.82	20.00	105.0	106.0	0.94
80.00	0.93	25.00	99.6	102.4	2.73
85.00	0.82	10.00	108.0	107.6	0.37

4.4 Analysis of Physicochemical Properties during Storage

4.4.1 Particle Size

Figure 4.3 shows the changes in particle size of nano loaded jackfruit leave emulsion during storage at room temperature and 45°C. An increase of average particle size was observed during the storage period of 60 days. In the study, the variation of particle size ranged from 99.6-105.7 nm at room temperature and was from 99.6-127.8 nm at 45°C. From the result, at the end of week 8, the particle size showed a significant increment about 3.1 % and 28 % from its initial size at temperature room temperature and 45°C respectively. In addition, a similar trend was obtained from the studies done by Ševčíková et al., (2011) where the formulated nanoemulsions shows increase in droplet size during storage at room temperature from 140 nm to 250 nm.

According to Tadros et al., (2004) the change in particle size was possessed by two important processes which are droplet break-up and coalescence. The nanoemulsions formulation is considered stable since there no flocculation and coalescence is observed during the storage time. This is due to the reason that the non-ionic emulsifier used in this formulation (T80 and S80) stabilize the system. They create a thick steric barrier to encapsulate the particles and hold the droplet together from break up. Besides, emulsion stabilization is enhanced by the presence of the micellar form of surfactant which avoids coalescence to occur (Tang et al., 2012). Although there are increments to the particle size, the nanoemulsions is still in the nano range which is 20 – 200 nm which exhibit good stability against creaming for 2 months. This is further supported by Akhtar et al., (2008) which state that the change in size occurs due to the reason that the numerous droplet in the system is coalescing with the oil droplet which contributes to the formation of larger droplet size. It is important to ensure the formulated nanoemulsions are in nano range size as their small

size will improve the penetration of bioactive ingredients to the skin effectively (Bouchemal et al., 2004). Therefore, their large surface area will enhance the transfer, release and improve the bioavailability of topical application to the skin.

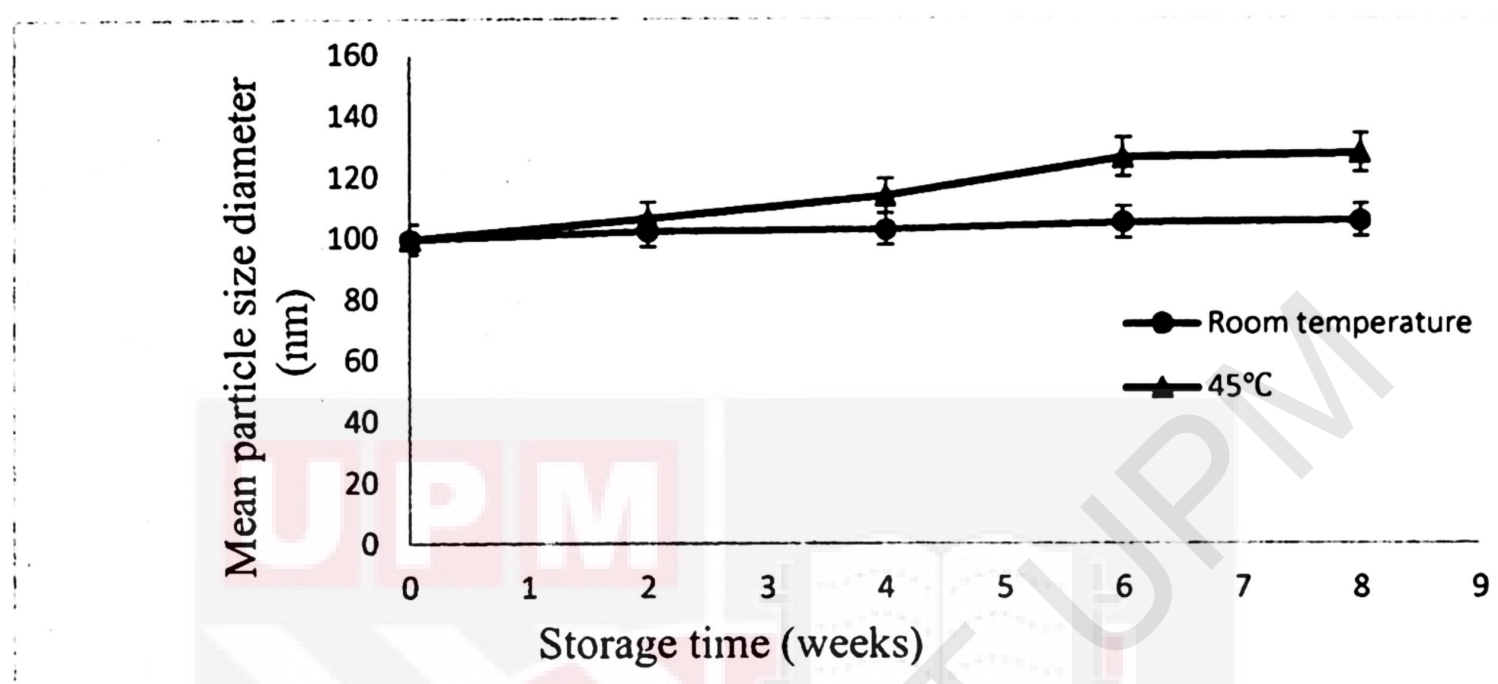


Figure 4. 3: Changes in particle size of nanoemulsions during storage for 8 weeks at room temperature and 45°C.

4.4.2 Viscosity

Figure 4.4 illustrated the changes in viscosity of nano loaded jackfruit leave emulsion during storage at room temperature and 45°C. In the beginning, the viscosity is quite high as the water molecule entangled in cross-linking chains of high concentration of surfactants (El Eini et al., 1976). However, as the storage time increases the viscosity starts to reduce continuously. Hakansson et al., (2009) states that the particle size is inversely proportional to the viscosity. In this work, the result obtained shows that the viscosity is reduce from week 0 to week 6 and from week 0 to week 8 at room temperature and 45°C respectively. The decrease in viscosity is due to hydrogen bonding and hydrophobic interaction which reduce the surface tension in the nanoemulsions.

Besides, the size enlargement causes the nanoemulsions become less cohesive. According to Mitri et al., (2011) due to destabilizing effect the viscosity will be lesser at higher storage temperature compare to lower temperature. A similar results are obtained from this work as the final viscosity at temperature 45°C is lower than the viscosity at room temperature. The oil-phase crystallization encourage half coalescence to rise and change the surfactant structure which directly affect the nanoemulsion viscosity (Cortés-Muñoz et al., 2009).

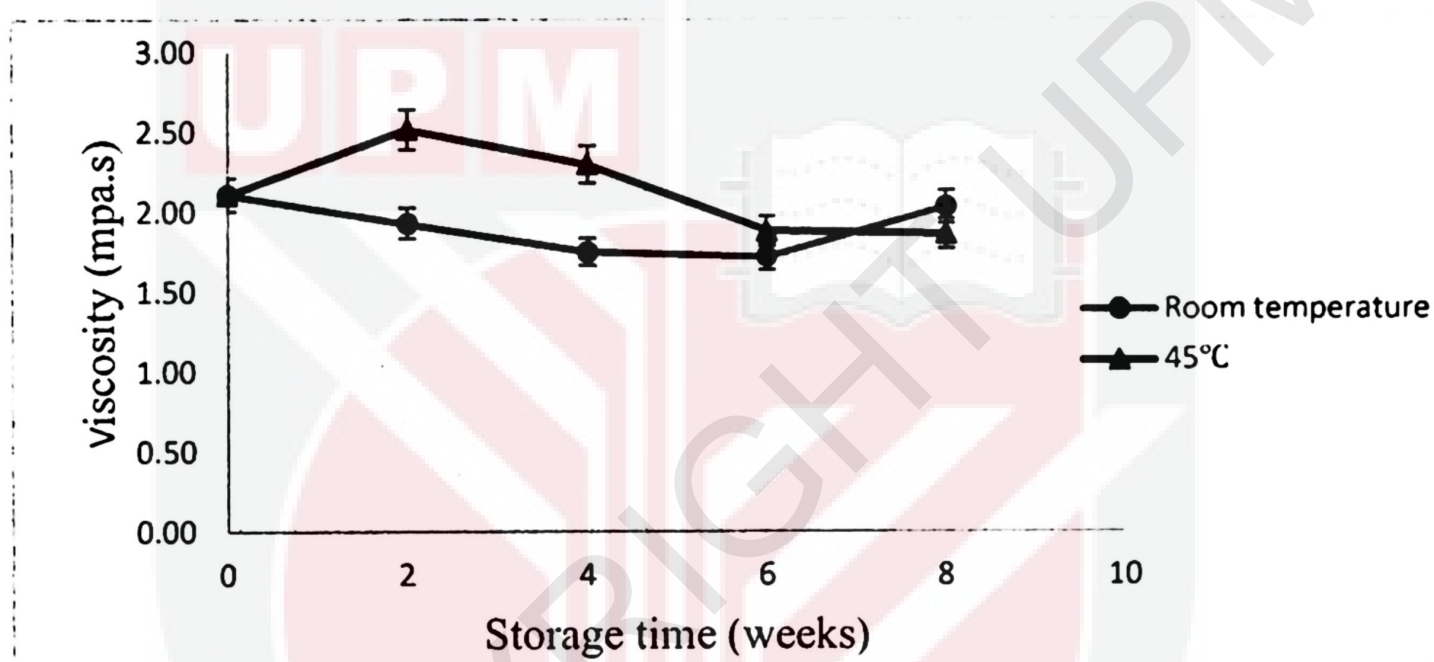


Figure 4. 4: Changes in viscosity of nanoemulsions during storage for 8 weeks at room temperature and 45°C.

4.4.3 pH

It is vital to monitor the pH of skin cosmetics products as we want to avoid any irritation to occur. The products which are too high in alkali will cause irritation which exposed the skin to the bacterial infection. According to Akhtar et al., (2008) the pH value for optimal nanoemulsion was 5.5 ± 0.01 . This pH value is suitable for the pH of human skin which range from 5 to 6. The low pH of the skin is due to reason that the buffer system in our skin only able to ingest a small amount of acid or alkali to reduce the level of skin irritation (Issachar et al., 1997). Therefore, a successful formulation for skin application should have pH within this range in order to avoid irritation to

happen. Initially, the result of pH obtained at week 0 was quite high which is at 6.11. However, Musa et al., (2017) claim at higher pH the nanoemulsions would provide better stability compare to at lower pH.

From this works, it is proven that the formulated nanoemulsions possessed good stability until week 8. In Figure 4.5, the pH for both storage temperature at room temperature and 45°C are decreasing significantly until week 8 which is from 6.11 to 5.92 and 5.82 at room temperature and 45°C respectively. Although the pH decreasing, they still remained at the ideal range of 5.0 – 6.0. The constituent in the Caprylic Capric Triglycerides such as Caprylic acid and Capric acid produce acidic by-product which contributes to the decrease in pH of the nanoemulsions. In addition according to Gallarate et al., (2013) the degeneration of some components in the oil phase which yield fatty acid also contribute to the low pH of our nanoemulsions. Similarly, Bernardi et al., (2011) also observed a decrease in pH values of O/W nanoemulsions containing rice bran oil after storage at 40 ± 2 °C for 90 days. The change that occur is acceptable as the pH values still remained in an acceptable range for the skin in that study.

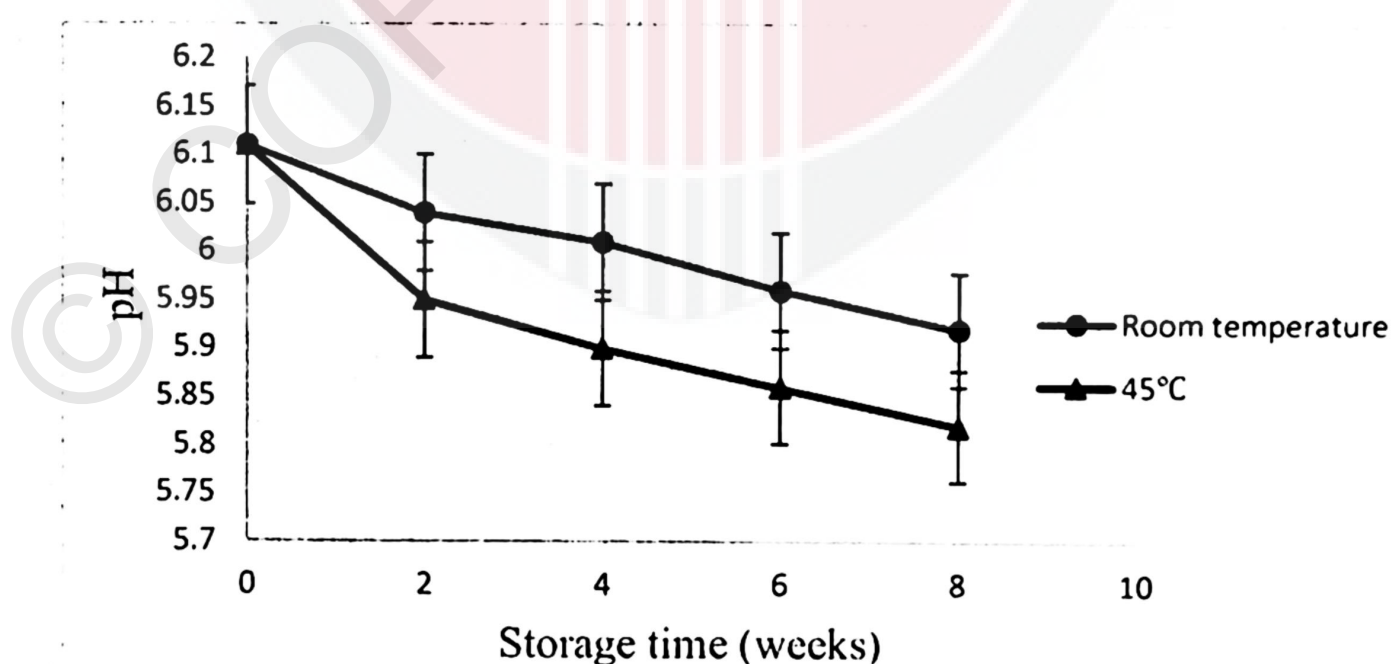


Figure 4. 5: Changes in pH of nanoemulsions during storage for 8 weeks at room temperature and 45°C

4.4.4 Turbidity

The measure of turbidity is correspondence to the relationship between particle size and concentration with the storage time. From figure 4.6 it is observed that the turbidity for both nano loaded jackfruit leave emulsion at temperature at room temperature and 45°C continuously increasing until week 8 of storage. Turbidity is directly proportional to the particle size. Therefore, the opaqueness and turbidity in nanoemulsion is correlate with the increment in particle size of nanoemulsion in storage period. Most of the previous studies observe a transparent to bluish appearance of the nanoemulsions generally. The optically transparent nanoemulsions is due to a higher concentration of surfactant use which causing a relatively weak scattering making the system less turbid (McClements, 2002). Apart from that, Zhang et al., (2015) clarify that the low turbidity is because the size of certain phase is lower than the wavelength of light ($r \ll \lambda$). A transmittance coefficient at 600 nm or by a turbidity ranging from 60 to 600 NTU are used to measure the turbidity.

According to Kong et al., (2011) the most desired range of turbidity is from 70 to 300 NTU which is quite small in range. In this work, the turbidity obtained for both storage condition exceed the range of the desired turbidity about 38 – 42 %. According to Mason et al., (2006), optical clarity may relate to the stability of the formulated the stability of nanoemulsions. However, the judgement will be more complex if the dispersed use are oil as they will produce a nanoemulsions with opaque, milky and white appearance although in nano range size (Bai et al : Xue et al. 2016). Therefore, Ghosh et al., (2013) claim the optical transparency cannot be used to precisely describe the particle size or stability of the formulations.

Typically nanoemulsions colour will resemble the compound in the bioactive extract although initially they contain oil which are milky white in colour. As for our formulated nanoemulsions, the milky green colour resembles the jackfruit leave extract. In addition, the surfactant used also will contribute to the colourless appearance of the nanoemulsions. Study done by Leong et al., (2011) on phytosterol Nano dispersion found that the emulsion are milky white in colour as they use sucrose fatty acid surfactant which is white in solution. A Similar result was observed from the studies done by Kabri et al., (2011) in cosmetic matrix enriched on omega-3 which obtained a turbidity value of 6248 NTU and they conclude that the increase in turbidity is due to colour and fluidity of the oil used (rapeseed oil). In addition, flocculation, coalescence, and aggregation contribute to the increase in turbidity during storage time (Mirhosseini et al., 2008).

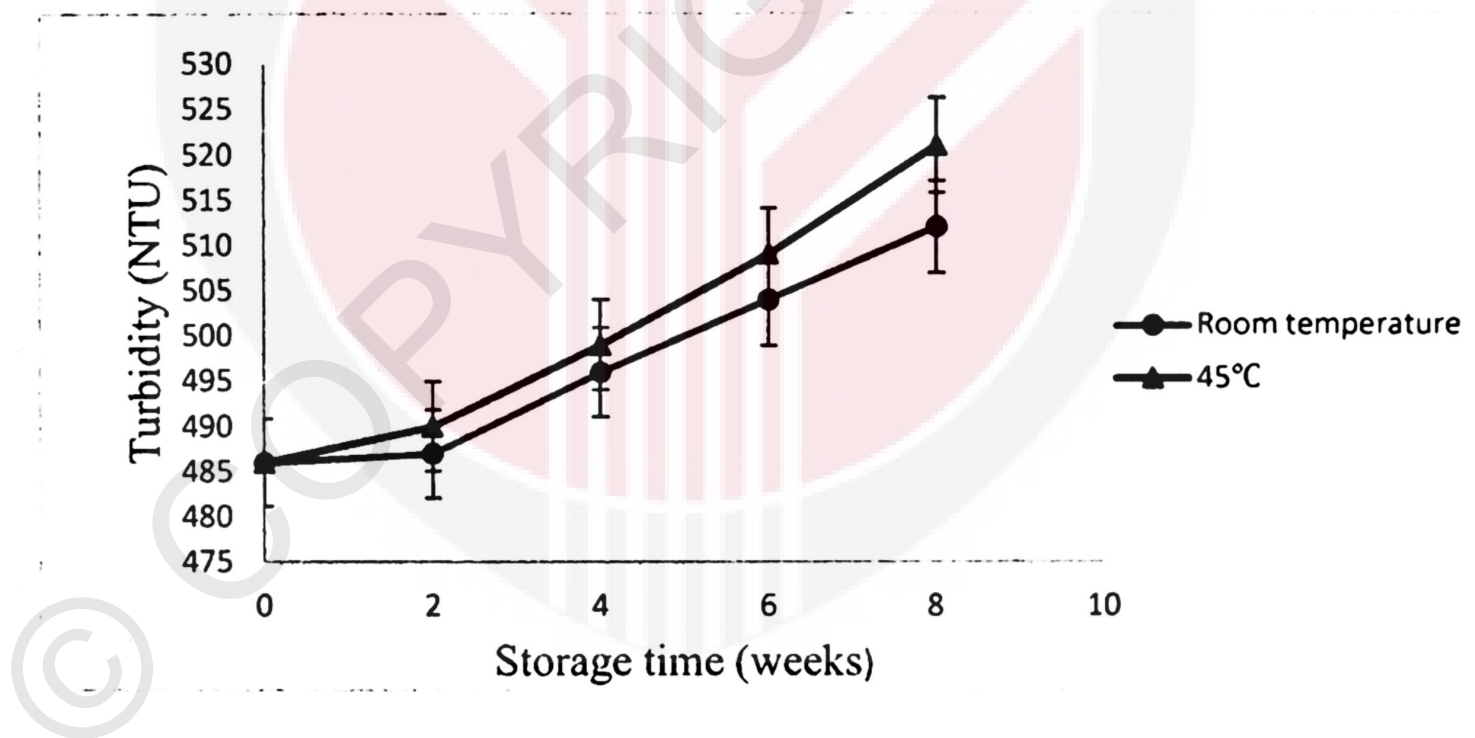


Figure 4. 6: Changes in turbidity of nanoemulsions during storage for 8 weeks at room temperature and 45

4.5 Stability Analysis

In the formulation study, it is very important to ensure the stability of the nanoemulsions. According to Ngan et al., (2014) physical properties and the process formulation are the factors that may contribute to the instability. Therefore, the action of nanoemulsions against separation, Coalescence, flocculation, and Ostwald ripening was observed. Flocculation is defined as the migration of the droplet as the element where they come closer together by attractive the force and Coalescence happened when the droplet combines and form larger particle (Rahn-Chique et al., 2012). Figure 4.7 shows the physical stability after storage of 2 months at room temperature and 45°C. From the figure illustrated, the formulations remained homogeneous and physically stable, with no creaming, phase separation, or flocculation. In addition, it is observed that the extreme change in temperature (45°C) applied on sample did not affect its stability. A similar trends were reported on the stability of nanoemulsions by ultrasound method. Ghosh et al., (2013) reported the basil oil nanoemulsions by ultrasound method formulated does not undergo any changes in the phase change during 1 month of storage and concluded that the emulsion was kinetically stable during that period. Besides, Jadhav et al., (2015) also report that the paraffin wax nanoemulsions produces by ultrasound method stable was stable during the 3 months storage. From the stability analysis, it can be concluded that products that possessed a longer shelf life without any physical changes will ensure the quality of the products.

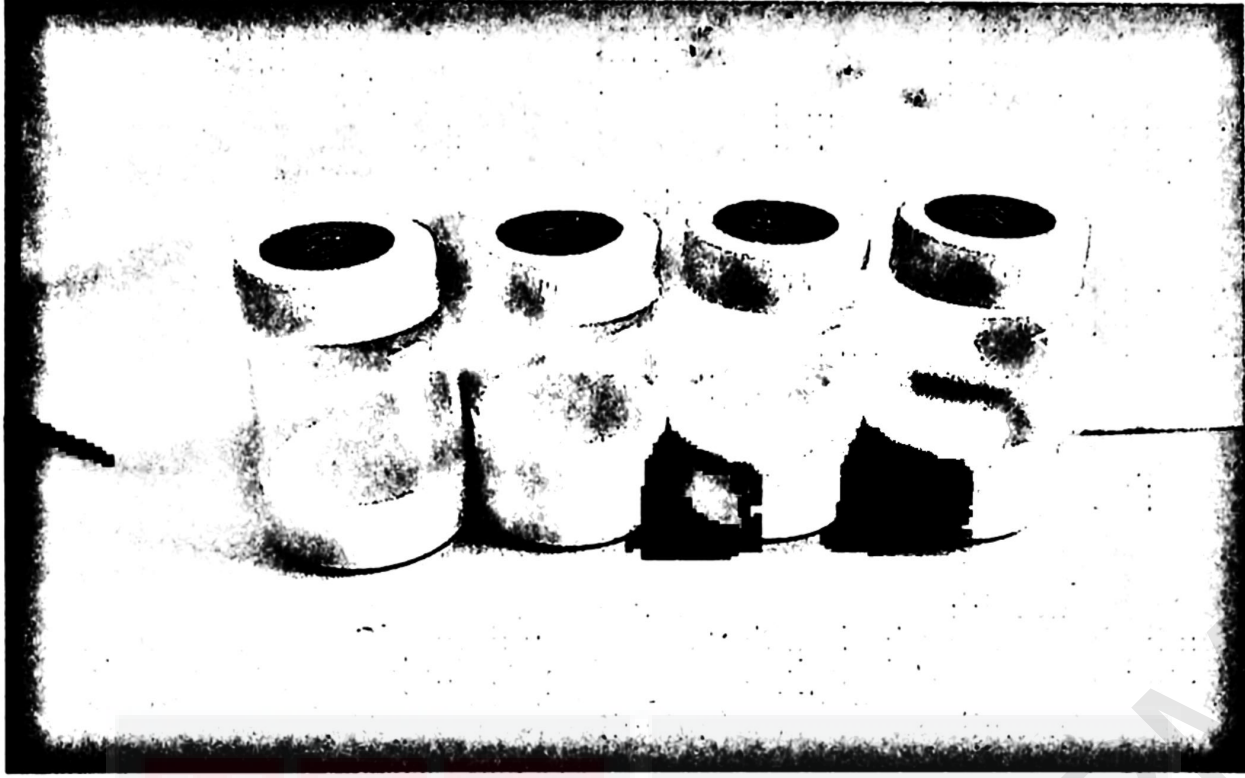


Figure 4. 7: Physical stability after storage of 2 month at room temperature, and 45°C

CHAPTER 5

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

The use of high energy method which is ultrasound is able to produce nanoemulsions formulations with a small particle size in the range of 20- 200 nm as we predicted earlier. In conducting the experiment design, 20 formulations of nanoemulsions were generated by the Response Surface Methodology which is the Central Composite Design through Design Expert 11 Software. The software is verified to be suitable to be used as process design in the formulation of nano loaded jackfruit leave emulsion as the residual standard error (RSE) that was obtained when the predicted and actual particle size were compared, it is only 2.73 %. Thus, the model used to obtain the nanoemulsions is considered to be reliable in this research work. From the experiment conducted, the smallest particle size of the nanoemulsions obtained was 99.6 nm at optimum formulation with 80 % water content, 0.93 O/S, and 25 minute ultrasonication time.

For the storage study, the stability of physicochemical properties of the optimum nanoemulsions changes during the storage time. The nano loaded jackfruit leave emulsion shows stability in the particle size, turbidity, pH and viscosity after storage of 8 weeks at storage of and 45°C. At the end of week 8, the particle size showed a significant increment about 3.1 % and 28 % from its initial size at temperature room temperature and 45°C respectively. For the viscosity, the viscosity is reduced during storage due to hydrogen bonding and hydrophobic interaction which reduce the surface tension in the nanoemulsions. However, during week 8, the viscosity of nanoemulsions at room temperature start shows instability as the quality of the nanoemulsions start to degrade with time. In addition, the pH of nanoemulsions still remained at the ideal range of 5.0 – 6.0 which is suitable for skin application although in the beginning the pH observed were 6.11. Besides, the turbidity of the nanoemulsions increases throughout the storage time as opaqueness and turbidity in the nanoemulsions were correlated with the increment in particle size of nanoemulsions in storage period. To conclude, this study resulted in useful information in order to formulate nanoemulsions with the ultrasound method.

5.2 Recommendations for Future Studies

In this study, it is found that by using ultrasound method the nano loaded jackfruit leave emulsion with a small particle size in the range of 20- 200 nm were successfully can be formulated. In other words, a successful formulation was obtained when the optimum condition such as the amount of water content, the ratio of oil to surfactant and the sonication time are properly being chosen during the formation of the nanoemulsions. The smallest particle size obtained from this study was 99.6 nm. However, the particle size can be reduced more if we increase the energy of sonicator as more energy will be dissipated to break the molecule of the nanoemulsions molecule. Thus, the penetration effect of the bioactive from the extract can be maximized.

Besides, there are several elements can be added in the formulation of the nanoemulsions for skin application such as tea tree oil as it can function as the natural anti-inflammatory, antibacterial, antimicrobial and anti-fungal to treat skin problems. Apart from that, honey will provide moisturizing effects, embrace antiseptic characteristic, support wound healing, fighting allergies or rashes, and helping to treat scars. Overall, various natural ingredients can be formulated in the skin care products as we want to reduce the number of harmful products used by the consumers as possible.

For future research, studies on the content of the phytochemical in leaves also can be conducted in order to determine the amount of the bioactive ingredients in the developed nanoemulsion. This is due to the reason that a maximize number of bioactive extract will increase the efficiency of the nanoemulsion. Then, the analysis

of the stability of the nanoemulsion can be improved by measuring the zeta potential and the interfacial tension (IFT) of the developed nanoemulsion.

Cosmetic products with a good quality required to have a longer shelf life for them to be marketed. Therefore, the formulation of the nanoemulsions must be properly choose in order to produce a high quality product with a good viscosity. The stability of the nanoemulsions can be enhanced by the addition of the thickener such as xanthan gum, carbomer, petroleum jelly and polyethylene glycol (PEG). The thickener will minimize the droplet movement of the particle, thus hold the particle together and reduce the effect of coalescence and phase separation which are not desired in the cosmetic formulations.

APPENDIX A

PHYSICOCHEMICAL PROPERTIES OF NANO LOADED JACKFRUIT LEAVES EMULSION DURING 8 WEEKS STORAGE

Table A. 1: Particle size of nanoemulsions during 8 weeks storage at room temperature and 45°C

	Week	Particle Size (nm)		Average
	Room temperature	0	99.3	99.8
2		101.3	102.8	102.1 ± 1.06
4		103.1	102.5	102.8 ± 0.42
6		105.5	104.5	105.0 ± 0.71
8		105.9	105.5	105.7 ± 0.28
Week		Particle Size (nm)		Average
45°C	0	99.3	99.8	99.6 ± 0.35
	2	107.0	105.8	106.4 ± 0.85
	4	115.4	111.9	113.7 ± 2.47
	6	129.5	122.8	126.2 ± 4.74
	8	133.4	122.1	127.8 ± 7.99

Table A. 2: pH of nanoemulsions during 8 weeks storage at Room temperature and 45 °C

	Week	pH		Average
	Room temperature	0	6.12	6.09
2		6.07	6.01	6.04 ± 0.04
4		6.02	5.99	6.01 ± 0.02
6		5.97	5.94	5.96 ± 0.02
8		5.93	5.91	5.92 ± 0.01
Week		pH		Average
45°C	0	6.12	6.09	6.11 ± 0.02
	2	5.90	6.00	5.95 ± 0.07
	4	5.85	5.94	5.90 ± 0.06
	6	5.82	5.90	5.86 ± 0.06
	8	5.77	5.86	5.82 ± 0.06

Table A. 3: Turbidity of nanoemulsion during 8 weeks storage at Room temperature and 45°C

	Week	Turbidity (NTU)		Average
	Room temperature	0	487	484
2		492	482	487 ± 7.07
4		494	497	496 ± 2.12
6		501	506	504 ± 3.54
8		510	513	512 ± 2.12
45°C		0	487	484
	2	494	486	490 ± 5.66
	4	503	495	499 ± 5.66
	6	511	507	509 ± 2.83
	8	526	516	521 ± 7.07

Table A. 4: Viscosity of nanoemulsion during 8 weeks storage at Room temperature and 45°C.

	Week	Viscosity (mPa.s)		Average
	Room temperature	0	2.23	1.98
2		2.14	1.71	1.93 ± 0.30
4		2.05	1.45	1.75 ± 0.42
6		1.66	1.45	1.72 ± 0.15
8		2.74	1.31	2.03 ± 1.01
45°C		0	2.23	1.98
	2	3.37	1.67	2.52 ± 1.20
	4	2.57	1.48	2.03 ± 0.77
	6	2.10	1.66	1.88 ± 0.31
	8	1.57	2.15	1.86 ± 0.41

APPENDIX B

STATISTICAL ANALYSIS RESULT

Table B. 1 Regression coefficients result of the final reduced models.

Regression coefficient	
Standard deviation	4.04
Ratio of R-Squared (R²)	0.6704
Adjusted R-Squared (Adj- R²)	0.5527
Predicted R-Squared	-0.0378
Adequate R-Squared	8.5361

Table B. 2 Comparison between experimental and predicted response

Formulation	A:water (%)	B:O/S	C:Sonication time(min)	Particle size (nm)	
				Experimental	Predicted
1	80	0.93	15	113.8 ± 2.97	116.3 ± 1.77
2	70	0.93	15	107.9 ± 0.28	113.0 ± 3.61
3	85	0.82	20	108.4 ± 0.42	106.0 ± 1.70
4	80	0.93	15	114.7 ± 0.35	116.3 ± 1.13
5	75	1.04	20	116.0 ± 0.49	111.2 ± 3.39
6	85	1.04	20	121.7 ± 0.28	118.4 ± 2.33
7	80	0.93	15	115.6 ± 0.49	116.3 ± 0.49
8	80	0.93	15	115.7 ± 0.42	116.3 ± 0.42
9	80	0.93	25	99.6 ± 0.35	102.4 ± 2.26
10	80	0.93	5	101.6 ± 0.71	106.0 ± 3.11
11	80	0.93	15	115.5 ± 0.35	116.3 ± 0.57
12	80	0.93	15	115.1 ± 0.07	116.3 ± 0.85
13	85	1.04	10	123.6 ± 0.64	120.0 ± 2.55
14	75	1.04	10	117.6 ± 0.78	112.8 ± 3.39
15	75	0.82	20	115.8 ± 0.21	112.2 ± 2.55
16	75	0.82	10	118.6 ± 0.85	113.8 ± 3.37
17	85	0.82	10	110.7 ± 1.27	107.6 ± 2.19
18	90	0.93	15	111.6 ± 1.27	114.0 ± 1.70
19	80	1.15	15	118.2 ± 0.49	122.6 ± 3.11
20	80	0.71	15	108.0 ± 3.39	111.2 ± 2.26

APPENDIX C

FIGURE OF EQUIPMENT USED



Figure C. 1: Front view of rotary evaporator



Figure C. 2 : Front view of ultrasound machine



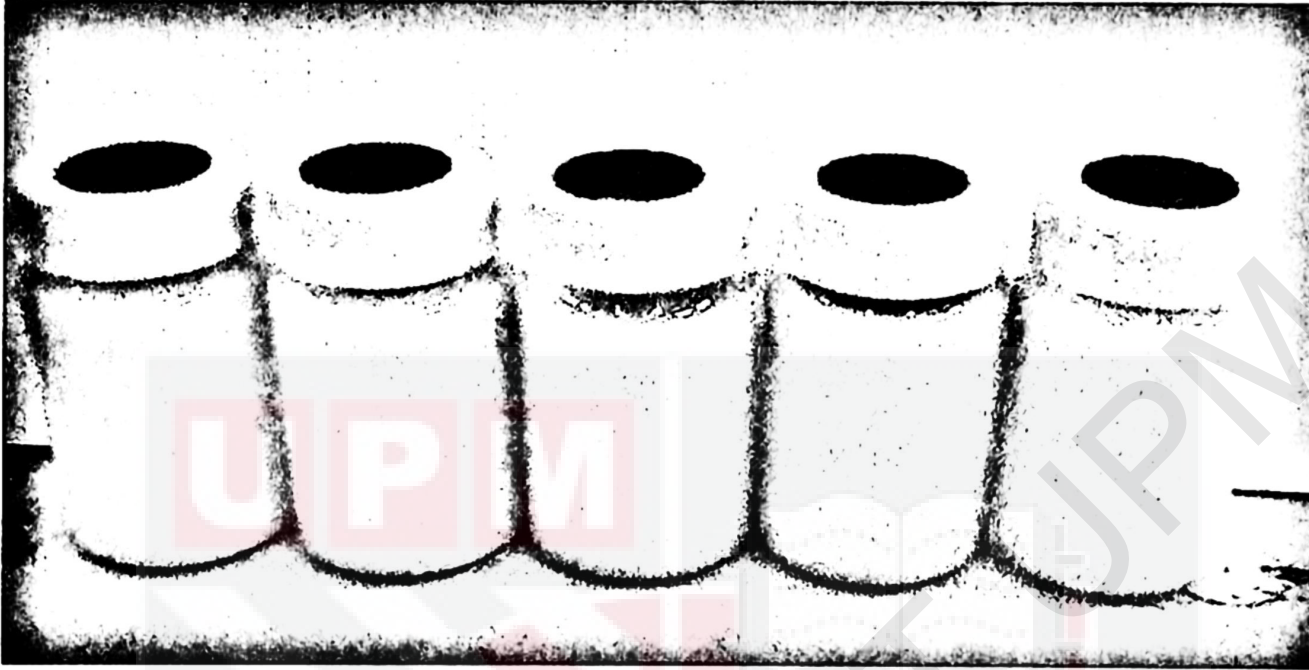
Figure C. 3 : Position of sample during ultrasonication



Figure C. 4: Front view of a rheometer

APPENDIX D

CAPTION OF FORMULATION OF NANO LOADED JACKFRUIT LEAVES EMULSION



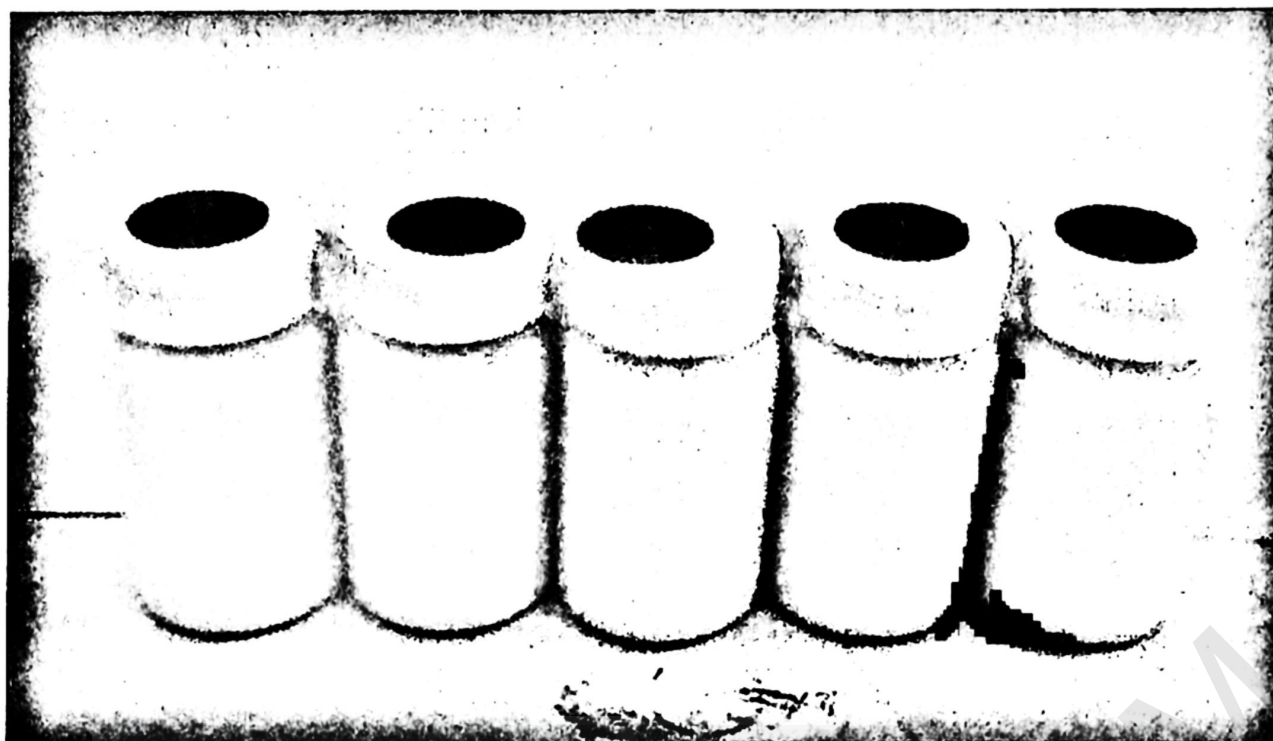


Figure D. 1: 20 Formulation of nanoemulsions by ultrasound method.

REFERENCES

- Akhtar, N., Ahmad, M., Gulfishan, Masood, M. I., & Aleem, M. (2008). Formulation and in Vitro evaluation of a cosmetic emulsion from almond oil. *Pakistan Journal of Pharmaceutical Sciences*, 21(4), 430–437. <https://doi.org/10.4103/0973-8398.49176>
- Anarjan, N., Mirhosseini, H., Baharin, B. S., & Tan, C. P. (2010). Effect of processing conditions on physicochemical properties of astaxanthin nanodispersions. *Food Chemistry*, 123(2), 477–483. <https://doi.org/10.1016/j.foodchem.2010.05.036>
- Anwar, F., & Przybylski, R. (2012). Effect of solvents extraction on total phenolics and antioxidant activity of extracts from flaxseed (*Linum usitatissimum* L.). *Acta Scientiarum Polonorum, Technologia Alimentaria*, 11(3), 293–302.
- Article, R. (2011). A DETAILED REVIEW ON NANOEMULSION DRUG DELIVERY SYSTEM, 2(10), 2482–2489.
- Artocarpus heterophyllus* Lam . (Moraceae): Phytochemistry , Pharmacology and Future Directions , a mini-review. (2017), (October).
- Arung, E. T., Shimizu, K., & Kondo, R. (2006). Inhibitory effect of artocarpone from *Artocarpus heterophyllus* on melanin biosynthesis. *Biological & Pharmaceutical Bulletin*, 29(9), 1966–1969.
- Arunkumar, N., Deecaraman, M., & Rani, C. (2011). Nanosuspension technology and its applications in drug delivery. *Asian J Pharm*, 3(3), 1–5. <https://doi.org/10.4103/0973-8398.56293>
- Azeem, A., Rizwan, M., Ahmad, F. J., Iqbal, Z., Khar, R. K., Aqil, M., & Talegaonkar, S. (2009). Nanoemulsion Components Screening and Selection: a Technical Note. *AAPS PharmSciTech*, 10(1), 69–76. <https://doi.org/10.1208/s12249-008-9178-x>
- Azizur Rahman, M., Nahar, N., Jabbar Mian, A., & Mosihuzzaman, M. (1999). Variation of carbohydrate composition of two forms of fruit from jack tree (*Artocarpus heterophyllus* L.) with maturity and climatic conditions. *Food Chemistry*, 65(1), 91–97. [https://doi.org/10.1016/S0308-8146\(98\)00175-7](https://doi.org/10.1016/S0308-8146(98)00175-7)
- Bernardi, D. S., Pereira, T. A., Maciel, N. R., Bortoloto, J., Viera, G. S., Oliveira, G. C., & Rocha-Filho, P. A. (2011). Formation and stability of oil-in-water nanoemulsions containing rice bran oil: in vitro and in vivo assessments. *Journal of Nanobiotechnology*, 9(1), 44. <https://doi.org/10.1186/1477-3155-9-44>
- Borges, V. R. de A., Simon, A., Sena, A. R. C., Cabral, L. M., & de Sousa, V. P. (2013). Nanoemulsion containing dapsona for topical administration: A study of in vitro release and epidermal permeation. *International Journal of Nanomedicine*, 8, 535–544.
- Bouchemal, K., Briançon, S., Perrier, E., & Fessi, H. (2004). Nano-emulsion formulation using spontaneous emulsification: Solvent, oil and surfactant optimisation. *International Journal of Pharmaceutics*, 280(1–2), 241–251.
- Carpenter, J., & Saharan, V. K. (2017). Ultrasonic assisted formation and stability of mustard oil in water nanoemulsion: Effect of process parameters and their optimization. *Ultrasonics Sonochemistry*, 35, 422–430.
- Che Sulaiman, I. S., Basri, M., Fard Masoumi, H. R., Ashari, S. E., & Ismail, M. (2016a). Design and development of a nanoemulsion system containing extract of *Clinacanthus nutans* (L.) leaves for transdermal delivery system by D-optimal mixture design and evaluation of its physicochemical properties. *RSC Adv.*, 6(71), 67378–67388.

- Che Sulaiman, I. S., Basri, M., Fard Masoumi, H. R., Ashari, S. E., & Ismail, M. (2016b). Design and development of a nanoemulsion system containing extract of *Clinacanthus nutans* (L.) leaves for transdermal delivery system by D-optimal mixture design and evaluation of its physicochemical properties. *RSC Adv.*, *6*(71), 67378–67388.
- Chen, H., Chang, X., Du, D., Li, J., Xu, H., & Yang, X. (2006). Microemulsion-based hydrogel formulation of ibuprofen for topical delivery. *International Journal of Pharmaceutics*, *315*(1–2), 52–58. <https://doi.org/10.1016/j.ica.2005.10.016>
- Chen, L., Tan, F., Wang, J., & Liu, F. (2012). Microemulsion: A novel transdermal delivery system to facilitate skin penetration of indomethacin. *Pharmazie*, *67*(4), 319–323.
- Cheng, N., Basri, M., Fui, L., Reza, H., Masoumi, F., Tripathy, M., ... Abdul-malek, E. (2014). Comparison of Box – Behnken and central composite designs in optimization of fullerene loaded palm-based nano-emulsions for cosmeceutical application. *Industrial Crops & Products*, *59*, 309–317. <https://doi.org/10.1016/j.indcrop.2014.05.042>
- Choi, K.-O., Aditya, N. P., & Ko, S. (2014). Effect of aqueous pH and electrolyte concentration on structure, stability and flow behavior of non-ionic surfactant based solid lipid nanoparticles. *Food Chemistry*, *147*, 239–44.
- Date, A. A., Desai, N., Dixit, R., & Nagarsenker, M. (2010). Self-nanoemulsifying drug delivery systems: formulation insights, applications and advances. *Nanomedicine (London, England)*, *5*(10), 1595–1616. <https://doi.org/10.2217/nnm.10.126>
- De Azevedo Ribeiro, R. C., Barreto, S. M. A. G., Ostrosky, E. A., Da Rocha-Filho, P. A., Verissimo, L. M., & Ferrari, M. (2015). Production and characterization of cosmetic nanoemulsions containing *Opuntia ficus-indica* (L.) Mill extract as moisturizing agent. *Molecules*, *20*(2), 2492–2509. <https://doi.org/10.3390/molecules20022492>
- Devarajan, V., & Ravichandran, V. (2011). Nanoemulsions: as modified drug delivery tool. *Pharmacie Globale*, *2*(4), 1–6. Retrieved from http://pharmacie-globale.info/index.php?option=com_docman&task=doc_download&gid=143&Itemid=41
- El Eini, D. I. D., Barry, B. W., & Rhodes, C. T. (1976). Micellar size, shape, and hydration of long-chain polyoxyethylene nonionic surfactants. *Journal of Colloid And Interface Science*, *54*(3), 348–351. [https://doi.org/10.1016/0021-9797\(76\)90314-3](https://doi.org/10.1016/0021-9797(76)90314-3)
- Elevitch, C. R., & Manner, H. I. (2006). *Artocarpus heterophyllus*. *Special Profiles for Pacific Island Agroforestry*, *1.1*(April).
- Fang, S. C., Hsu, C. L., & Yen, G. C. (2008). Anti-inflammatory effects of phenolic compounds isolated from the fruits of *Artocarpus heterophyllus*. *Journal of Agricultural and Food Chemistry*, *56*(12), 4463–4468.
- Fernando, M. R., Wickramasinghe, S. M. D. N., Thabrew, M. I., Ariyananda, P. L., & Karunanayake, E. H. (1991). Effect of *Artocarpus heterophyllus* and *Asteracanthus longifolia* on glucose tolerance in normal human subjects and in maturity-onset diabetic patients. *Journal of Ethnopharmacology*, *31*(3), 277–282.
- Gadhav, A. D. (2014). Nanoemulsions : Formation , Stability and Applications, (3), 38–43.
- Gallarate, M., Chirio, D., Bussano, R., Peira, E., Battaglia, L., Baratta, F., & Trotta, M. (2013). Development of O/W nanoemulsions for ophthalmic administration of timolol. *International Journal of Pharmaceutics*, *440*(2), 126–134.

- Gao, X. H., Zhang, L., Wei, H., & Chen, H. D. (2008). Efficacy and safety of innovative cosmeceuticals. *Clinics in Dermatology*, 26(4), 367–374.
- Ghosh, V., Mukherjee, A., & Chandrasekaran, N. (2013). Ultrasonic emulsification of food-grade nanoemulsion formulation and evaluation of its bactericidal activity. *Ultrasonics Sonochemistry*, 20(1), 338–344. <https://doi.org/10.1016/j.ultsonch.2012.08.010>
- Ghotbi, R. S., Khatibzadeh, M., & Kordbacheh, S. (2014). Preparation of Neem Seed Oil Nanoemulsion. *Proceedings of the 5th International Conference on Nanotechnology: Fundamentals and Applications*, (150), 1–5.
- Gupta, A., Eral, H. B., Hatton, T. A., & Doyle, P. S. (2016). Soft Matter and applications. *Soft Matter*. <https://doi.org/10.1039/C5SM02958A>
- Gutiérrez, J. M., González, C., Maestro, A., Solè, I., Pey, C. M., & Nolla, J. (2008). Nano-emulsions: New applications and optimization of their preparation. *Current Opinion in Colloid and Interface Science*. <https://doi.org/10.1016/j.cocis.2008.01.005>
- Hari, A., & Divya, D. (2014). Artocarpus, a Review of Its Phytochemistry and. *Journal of Pharma Search*, 9(1), 7–12. Retrieved from <http://nationalcollegeofpharmacy.yolasite.com/resources/2014-01-02.pdf>
- Hassan, M., Hosseini, M., Jahanshahi, M., Louise, R., & Najafpour, G. (2015). Evaluation of critical parameters for preparation of stable clove oil nanoemulsion. *ARABIAN JOURNAL OF CHEMISTRY*. <https://doi.org/10.1016/j.arabjc.2015.08.024>
- Haswani Maisarah Binti Mustafa (GS47065), Verbal Communication.
- Heuschkel, S., Goebel, A., & Neubert, R. H. H. (2008). Microemulsions - Modern colloidal carrier for dermal and transdermal drug delivery. *Journal of Pharmaceutical Sciences*.
- Hippalgaonkar, K., Majumdar, S., & Kansara, V. (2010). Injectable Lipid Emulsions—Advancements, Opportunities and Challenges. *AAPS PharmSciTech*, 11(4), 1526–1540.
- Ismail, N., & Kaur, B. (2013). Consumer Preference for Jackfruit Varieties in Malaysia. *Journal of Agribusiness Marketing*, 6, 37–51.
- Issachar, N., Gall, Y., Borell, M. T., & Poelman, M. C. (1997). pH measurements during lactic acid stinging test in normal and sensitive skin. *Contact Dermatitis*, 36(3), 152–155.
- Jadhav, A. J., Holkar, C. R., Karekar, S. E., Pinjari, D. V., & Pandit, A. B. (2015). Ultrasound assisted manufacturing of paraffin wax nanoemulsions: Process optimization. *Ultrasonics Sonochemistry*, 23, 201–207.
- Jagtap, U. B., Panaskar, S. N., & Bapat, V. A. (2010). Evaluation of antioxidant capacity and phenol content in jackfruit (*Artocarpus heterophyllus* Lam.) fruit pulp. *Plant Foods for Human Nutrition*, 65(2), 99–104.
- Jagtap, U. B., Waghmare, S. R., Lokhande, V. H., Suprasanna, P., & Bapat, V. A. (2011). Preparation and evaluation of antioxidant capacity of Jackfruit (*Artocarpus heterophyllus* Lam.) wine and its protective role against radiation induced DNA damage. *Industrial Crops and Products*, 34(3), 1595–1601.
- Javadzadeh, Y., & Hamishehkar, H. (2011). Enhancing percutaneous delivery of methotrexate using different types of surfactants. *Colloids and Surfaces B: Biointerfaces*, 82(2), 422–426.
- Jaworska, M., Sikora, E., & Ogonowski, J. (2014). The influence of glicerides oil phase on

O/W nanoemulsion formation by pic method. *Periodica Polytechnica Chemical Engineering*, 58(SUPPL), 43–48.

- Kabri, T. H., Arab-Tehrany, E., Belhaj, N., & Linder, M. (2011). Physico-chemical characterization of nano-emulsions in cosmetic matrix enriched on omega-3. *Journal of Nanobiotechnology*, 9.
- Kentish, S., Wooster, T. J., Ashokkumar, M., Balachandran, S., Mawson, R., & Simons, L. (2008). The use of ultrasonics for nanoemulsion preparation. *Innovative Food Science and Emerging Technologies*, 9(2), 170–175.
- Khan, M. R., Omoloso, A. D., & Kihara, M. (2003). Antibacterial activity of *Artocarpus heterophyllus*. *Fitoterapia*, 74(5), 501–505. Kong, S., Application, F., & Data, P. (2011). (12) United States Patent, 2(12), 12–15.
- Lane, M. E. (2013). Skin penetration enhancers. *International Journal of Pharmaceutics*.
- Leong, T. S. H., Wooster, T. J., Kentish, S. E., & Ashokkumar, M. (2009a). Minimising oil droplet size using ultrasonic emulsification. *Ultrasonics Sonochemistry*, 16(6), 721–
- Leong, T. S. H., Wooster, T. J., Kentish, S. E., & Ashokkumar, M. (2009b). Minimising oil droplet size using ultrasonic emulsification. *Ultrasonics Sonochemistry*, 16(6), 721–727.
- Li, P. H., & Chiang, B. H. (2012). Process optimization and stability of d-limonene-in-water nanoemulsions prepared by ultrasonic emulsification using response surface methodology. *Ultrasonics Sonochemistry*, 19(1), 192–197.
- Lv, G., Wang, F., Cai, W., & Zhang, X. (2014). Characterization of the addition of lipophilic Span 80 to the hydrophilic Tween 80-stabilized emulsions. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 447, 8–13. 6
- Mahdi, E. S., Noor, A. M., Sakeena, M. H., Abdullah, G. Z., Abdulkarim, M. F., & Sattar, M. A. (2011). Formulation and in vitro release evaluation of newly synthesized palm kernel oil esters-based nanoemulsion delivery system for 30% ethanolic dried extract derived from local *Phyllanthus urinaria* for skin antiaging. *International Journal of Nanomedicine*, 6, 2499–2512.
- Manchun, S., Dass, C. R., & Sriamornsak, P. (2014). Designing nanoemulsion templates for fabrication of dextrin nanoparticles via emulsion cross-linking technique. *Carbohydrate Polymers*, 101(1), 650–655.
- McClements, D. J. (2002). Theoretical prediction of emulsion color. *Advances in Colloid and Interface Science*, 97(1–3), 63–89. [https://doi.org/10.1016/S0001-8686\(01\)00047-1](https://doi.org/10.1016/S0001-8686(01)00047-1)
- McClements, D. J. (2011). Edible nanoemulsions: fabrication, properties, and functional performance. *Soft Matter*, 7(6), 2297–2316. <https://doi.org/10.1039/C0SM00549E>
- Medical, T., & Applications, N. (1981). Technical News Features _____, (January), 23–25.
- Mehmood, T., Ahmad, A., Ahmed, A., & Ahmed, Z. (2017). Optimization of olive oil based O/W nanoemulsions prepared through ultrasonic homogenization: A response surface methodology approach. *Food Chemistry*, 229, 790–796.
- Modi, K., & Shelat, P. (2012). Applications of novel vesicular drug delivery system as ocular drug vehicles: a review. ... *Journal of Pharmaceutical Sciences and Research*, 3(12), 4554–4561. Retrieved from <http://scholar.google.co.in/scholar?start=70&q=K.B.+Institute+of+Pharmaceutical+Ed>

- Moke, L. E., Ngbolua, K., Bongo, G. N., Messi, L. M., Noté, O. P., Mbing, J. N., & Mpiana, P. T. (2017). *Artocarpus heterophyllus* Lam. (Moraceae): Phytochemistry , Pharmacology and Future Directions , a mini-review, 5(3), 1–8.
- Montenegro, L., Rapisarda, L., Ministeri, C., & Puglisi, G. (2015). Effects of Lipids and Emulsifiers on the Physicochemical and Sensory Properties of Cosmetic Emulsions Containing Vitamin E. *Cosmetics*, 2(1), 35–47.
- Musa, S. H., Basri, M., Masoumi, H. R. F., Karjiban, R. A., Malek, E. A., Basri, H., & Shamsuddin, A. F. (2013). Formulation optimization of palm kernel oil esters nanoemulsion-loaded with chloramphenicol suitable for meningitis treatment. *Colloids and Surfaces B: Biointerfaces*, 112, 113–119.
- Musa, S. H., Basri, M., Masoumi, H. R. F., Shamsudin, N., & Salim, N. (2017). Enhancement of physicochemical properties of nanocolloidal carrier loaded with cyclosporine for topical treatment of psoriasis: In vitro diffusion and in vivo hydrating action. *International Journal of Nanomedicine*, 12, 2427–2441.
- Nandi, S., Gangopadhyay, S., & Ghosh, S. (2005). Production of medium chain glycerides from coconut and palm kernel fatty acid distillates by lipase-catalyzed reactions. *Enzyme and Microbial Technology*, 36(5–6), 725–728.
- Nastiti, C. M. R. R., Ponto, T., Abd, E., Grice, J. E., Benson, H. A. E., & Roberts, M. S. (2017). Topical nano and microemulsions for skin delivery. *Pharmaceutics*, 9(4), 1–25.
- Ngan, C. L. oong, Basri, M., Lye, F. F. ang, Fard Masoumi, H. R. eza, Tripathy, M., Karjiban, R. A. bedi, & Abdul-Malek, E. (2014). Comparison of process parameter optimization using different designs in nanoemulsion-based formulation for transdermal delivery of fullerene. *International Journal of Nanomedicine*, 9, 4375–4386.
- Omar, H. S., El-Beshbishy, H. A., Moussa, Z., Taha, K. F., & Singab, A. N. B. (2011). Antioxidant Activity of *Artocarpus heterophyllus* Lam. (Jack Fruit) Leaf Extracts: Remarkable Attenuations of Hyperglycemia and Hyperlipidemia in Streptozotocin-Diabetic Rats. *The Scientific World JOURNAL*, 11, 788–800.
- Onivogui, G., Letsididi, R., Diaby, M., Wang, L., & Song, Y. (2016). Influence of extraction solvents on antioxidant and antimicrobial activities of the pulp and seed of *Anisophyllea laurina* R. Br. ex Sabine fruits. *Asian Pacific Journal of Tropical Biomedicine*, 6(1), 20–25.
- Özgün, S. (2013). Nanoemulsions in Cosmetics. *Anadolu Univ*, (January 2013). Retrieved from http://www.researchgate.net/profile/Sinan_Oezguen/publication/235890223_Nanoemulsions_in_Cosmetics/links/0c96051d2acc0a9b9e000000.pdf
- Paolino, D., Ventura, C. A., Nisticò, S., Puglisi, G., & Fresta, M. (2002). Lecithin microemulsions for the topical administration of ketoprofen: Percutaneous adsorption through human skin and in vivo human skin tolerability. *International Journal of Pharmaceutics*, 244(1–2), 21–31. [https://doi.org/10.1016/S0378-5173\(02\)00295-8](https://doi.org/10.1016/S0378-5173(02)00295-8)
- Peng, L. C., Liu, C. H., Kwan, C. C., & Huang, K. F. (2010). Optimization of water-in-oil nanoemulsions by mixed surfactants. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 370(1–3), 136–142.
- Peshkovsky, A. S., Peshkovsky, S. L., & Bystryak, S. (2013). Scalable high-power ultrasonic

- technology for the production of translucent nanoemulsions. *Chemical Engineering and Processing: Process Intensification*, 69, 77–82.
- Pey, C. M., Maestro, A., Solé, I., González, C., Solans, C., & Gutiérrez, J. M. (2006). Optimization of nano-emulsions prepared by low-energy emulsification methods at constant temperature using a factorial design study. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 288(1–3), 144–150.
- Porras, M., Solans, C., González, C., Martínez, A., Guinart, A., & Gutiérrez, J. M. (2004). Studies of formation of W/O nano-emulsions. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 249(1–3), 115–118.
- Prakash, O., Kumar, R., Chandra, D., Kumar, A., & Kumar, P. (2015). Effect of *Artocarpus heterophyllus* Lam. (Jackfruit) on Indomethacin-Induced ulcer model in albino rats. *Der Pharmacia Lettre*, 7(1), 81–85.
- Radhika, P. R., & Guruprasad, S. (2016). Nanoemulsion based emulgel formulation of lipophilic drug for topical delivery. *International Journal of PharmTech Research*, 9(6), 210–223.
- Rahn-Chique, K., Puertas, A. M., Romero-Cano, M. S., Rojas, C., & Urbina-Villalba, G. (2012). Nanoemulsion stability: Experimental evaluation of the flocculation rate from turbidity measurements. *Advances in Colloid and Interface Science*.
- Raj, S., Sumod, U., Jose, S., & Sabitha, M. (2012). Nanotechnology in cosmetics: Opportunities and challenges. *Journal of Pharmacy and Bioallied Sciences*, 4(3), 186.
- Rao, J., & McClements, D. J. (2011). Formation of flavor oil microemulsions, nanoemulsions and emulsions: Influence of composition and preparation method. *Journal of Agricultural and Food Chemistry*, 59(9), 5026–5035.
- Rezaee, M., Basri, M., Raja, R. N. Z., Rahman, A., Salleh, A. B., Chaibakhsh, N., & Karjiban, R. A. (2014). Formulation development and optimization of palm kernel oil esters-based nanoemulsions containing sodium diclofenac. *International Journal of Nanomedicine*, 9(1), 539–548.
- Roberts, M. S., Mohammed, Y., Pastore, M. N., Namjoshi, S., Yousef, S., Alinaghi, A., ... Grice, J. E. (2017). Topical and cutaneous delivery using nanosystems. *Journal of Controlled Release*.
- Rocha-Filho, P., Ferrari, M., Maruno, M., Souza, O., & Gumiero, V. (2017). In Vitro and In Vivo Evaluation of Nanoemulsion Containing Vegetable Extracts. *Cosmetics*, 4(3), 32. 2
- Sadurní, N., Solans, C., Azemar, N., & García-Celma, M. J. (2005). Studies on the formation of O/W nano-emulsions, by low-energy emulsification methods, suitable for pharmaceutical applications. *European Journal of Pharmaceutical Sciences*, 26(5), 438–445.
- Salim, N., Basri, M., Rahman, M. B. A., Abdullah, D. K., & Basri, H. (2012). Modification of palm kernel oil esters nanoemulsions with hydrocolloid gum for enhanced topical delivery of ibuprofen. *International Journal of Nanomedicine*, 7, 4739–4747.
- Serajuddin, A. T. M. (2012). Enhanced microemulsion formation in lipid-based drug delivery systems by combining mono-esters of medium-chain fatty acids with di- or tri-esters. *Journal of Excipients and Food Chemicals*, 3(2), 29–44. Retrieved from <https://ojs.abo.fi/index.php/jefc/article/view/138>

- Shah, P., Bhalodia, D., & Shelat, P. (2010). Nanoemulsion: A pharmaceutical review. *Systematic Reviews in Pharmacy*, 1(1), 24.
- Shahavi, M. H., Hosseini, M., Jahanshahi, M., & Darzi, G. N. (2015). Optimization of encapsulated clove oil particle size with biodegradable shell using design expert methodology. *Pakistan Journal of Biotechnology*, 12(2), 149–160.
- Shahavi, M. H., Hosseini, M., Jahanshahi, M., Meyer, R. L., & Darzi, G. N. (2015). Evaluation of critical parameters for preparation of stable clove oil nanoemulsion. *Arabian Journal of Chemistry*.
- Sharma, N., Bansal, M., Visht, S., Sharma, P. K., & Kulkarni, G. T. (2010). Nanoemulsion : A new concept of delivery system anoemulsion : A new concept of delivery system, (May 2014).
- Shen, Q., Wang, Y., & Zhang, Y. (2011). Improvement of colchicine oral bioavailability by incorporating eugenol in the nanoemulsion as an oil excipient and enhancer. *International Journal of Nanomedicine*, 6, 1237–1243.
- Silva, F. F. F., Ricci-Júnior, E., & Mansur, C. R. E. (2013). Nanoemulsions containing octyl methoxycinnamate and solid particles of TiO₂: preparation, characterization and in vitro evaluation of the solar protection factor. *Drug Development and Industrial Pharmacy*, 39(9), 1378–1388.
- Sivakumar, M., Tang, S. Y., & Tan, K. W. (2014). Cavitation technology - A greener processing technique for the generation of pharmaceutical nanoemulsions. *Ultrasonics Sonochemistry*, 21(6), 2069–2083.
- Solans, C., Izquierdo, P., Nolla, J., & Azemar, N. (2005). Nano-emulsions, 10, 102–110.
- Solans, C., Izquierdo, P., Nolla, J., Azemar, N., & Garcia-Celma, M. J. (2005). Nano-emulsions. *Current Opinion in Colloid and Interface Science*.
- Sonneville-Aubrun, O., Simonnet, J. T., & L'Alloret, F. (2004). Nanoemulsions: A new vehicle for skincare products. *Advances in Colloid and Interface Science*, 108–109, 145–149.
- Sugumar, S., Ghosh, V., Nirmala, M. J., Mukherjee, A., & Chandrasekaran, N. (2014). Ultrasonic emulsification of eucalyptus oil nanoemulsion: Antibacterial activity against *Staphylococcus aureus* and wound healing activity in Wistar rats. *Ultrasonics Sonochemistry*, 21(3), 1044–1049. <https://doi.org/10.1016/j.ultsonch.2013.10.021>
- Sultana, B., Anwar, F., & Ashraf, M. (2009). Effect of extraction solvent/technique on the antioxidant activity of selected medicinal plant extracts. *Molecules*, 14(6), 2167–2180.
- Swami, S. B., Thakor, N. J., Haldankar, P. M., & Kalse, S. B. (2012). Jackfruit and Its Many Functional Components as Related to Human Health: A Review. *Comprehensive Reviews in Food Science and Food Safety*, 11(6), 565–576.
- Tadros, T., Izquierdo, P., Esquena, J., & Solans, C. (2004). Formation and stability of nano-emulsions. *Advances in Colloid and Interface Science*.
- Tang, S. Y., Manickam, S., Wei, T. K., & Nashiru, B. (2012). Formulation development and optimization of a novel Cremophore EL-based nanoemulsion using ultrasound cavitation. *Ultrasonics Sonochemistry*, 19(2), 330–345.
- Teo, B., Basri, M., Zakaria, M., Salleh, A., Rahman, R., & Rahman, M. (2010). A potential tocopherol acetate loaded palm oil esters-in-water nanoemulsions for nanocosmeceuticals. *Journal of Nanobiotechnology*, 8(1), 4.

- Tsai, Y. H., Chang, J. T., Chang, J. S., Huang, C. Te, Huang, Y. Bin, & Wu, P. C. (2011). The effect of component of microemulsions on transdermal delivery of buspirone hydrochloride. *Journal of Pharmaceutical Sciences*, 100(6), 2358–2365.
- Varshosaz, J., Andalib, S., Tabbakhian, M., & Ebrahimzadeh, N. (2013). Development of lecithin nanoemulsion based organogels for permeation enhancement of metoprolol through rat skin. *Journal of Nanomaterials*, 2013.
- Wei, B. L., Weng, J. R., Chiu, P. H., Hung, C. F., Wang, J. P., & Lin, C. N. (2005). Antiinflammatory flavonoids from *Artocarpus heterophyllus* and *Artocarpus communis*. *Journal of Agricultural and Food Chemistry*, 53(10), 3867–3871.
- Wetprasit, N., Threesangsri, W., Klamklai, N., & Chulavatnatol, M. (2000). Jackfruit lectin: Properties of mitogenicity and the inhibition of herpesvirus infection. *Japanese Journal of Infectious Diseases*, 53(4), 156–161.
- Yang, L. (2017). Formulation , development , and optimization of a novel octyldodecanol-based nanoemulsion for transdermal delivery of ceramide III B, 5203–5221.
- Yang, Y., Marshall-Breton, C., Leser, M. E., Sher, A. A., & McClements, D. J. (2012). Fabrication of ultrafine edible emulsions: Comparison of high-energy and low-energy homogenization methods. *Food Hydrocolloids*, 29(2), 398–406.
- Yukuyama, M. N., Ghisleni, D. D. M., Pinto, T. J. A., & Bou-Chacra, N. A. (2016). Nanoemulsion: Process selection and application in cosmetics - A review. *International Journal of Cosmetic Science*, 38(1), 13–24.
- Zhang, J., Peppard, T. L., & Reineccius, G. A. (2015). Preparation and characterization of nanoemulsions stabilized by food biopolymers using microfluidization. *Flavour and Fragrance Journal*, 30(4), 288–294.