Review Article

General Formulation of Nanoemulsions Using High-Energy and Low-Energy Principal Methods: A Brief Review

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Abstract

Nanoemulsions represent the droplet size as 100 nm and defined as biphasic dispersion of two immiscible liquids. Nanoemulsions are thermodynamically unstable while kinetically stable and they are used in many industrial fields; such as food, medicinal, pharmaceutical, cosmetic, agricultural and chemical. Formation of nanoemulsions using mechanical devices with high energy input is referred as "High energy method" while the method that utilize internal chemical energy of components known as "Low energy method". The High energy methods can be described in five sub-categories: the high-pressure homogenization method, microfluidizer method, ultrasonication method, jet disperser method and high shear stirring method. The low energy methods are self-nano emulsification, Phase inversion, D phase emulsification, Membrane emulsification, Microemulsion dilution, and Solvent displacement. Since low-energy approaches can develop nanoemulsions without high energy and without any expensive instruments, it can be considered a cost effective nanoemulsions formation method. But use of synthetic surfactants in low-energy method might limit their applications for food systems due to high requirement of surfactants for the stability of formed nanoemulsions with small sizes. In that case, high energy method is most effective even it consumes much energy. This review aims to highlight the high-energy and low energy nanoemulsions formation methods, major differences of those two methods and potential applications of nanoemulsions.

Keywords: *Applications, high energy methods, low energy methods, Nanoemulsions*

1. Introduction

In the last few decades, Nanoemulsions have gained much interest for their use in many different applications due to their unique properties such as low viscosity, high kinetic stability against creaming or sedimentation and a large interfacial area. Nanoemulsions contain two immiscible liquids as a colloidal dispersion, which has stable kinetic properties with unstable thermodynamic properties. Among these two immiscible liquids, one forms the dispersed phase and the other forms the continuous phase also known as the dispersing medium. Nanoemulsions comprise droplets with diameters ranging from 10~200 nm and each droplet has a protective coating of emulsifier molecules for protection as well as stability (Acosta, 2009). The emulsifier used for this purpose is generally a surfactant, which is classified based on hydrophilic-lipophilic balance (HLB) (Safaya and Rotliwala, 2020). The most important and effective surfactants are the non-ionic surfactants which are used to design oil in water emulsions (o/w) or water in oil emulsions (w/o) (Figure 1) (Fryd and Mason, 2012b; Gupta *et al.*, 2016).

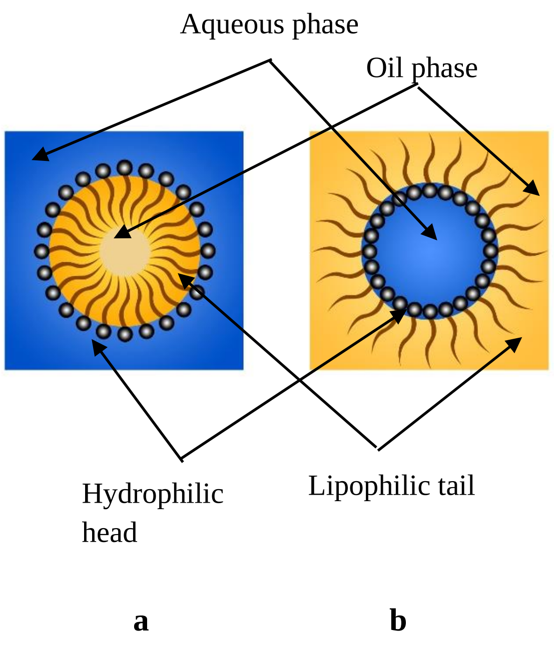

Figure 01: Schematic diagram of a) Oil in water O/W emulsion: b) Water in oil W/O emulsions

Numerous research papers have discussed the advantages and disadvantages of nanoemulsions formation methods, including high-energy and low-energy methods. Many argued that, Low energy methods is the more preferable method than high energy methods thus, it requires less energy, and do not require any sophisticated instruments to perfume more efficient processes while some researches show high energy methods are more favorable specially in food grade emulsion thus it requires lower quantities of surfactant throughout the process than low energy methods.

The focus of the current review is on creating nanoemulsions using high and low energy techniques. To increase the stability of nanoemulsions, the preparation process is a crucial factor to take into consideration. Both high and low emulsification energy techniques can be used to prepare these systems. High-energy procedures require for a significant amount of mechanical energy input from tools like microfluidizers, high pressure homogenizers, or ultrasonic techniques that produce powerful forces capable of producing very small oil droplets with a high kinetic energy. (Anton *et al.*, 2008). Low-energy techniques use the chemical energy contained in components, which uses less energy than high-energy processes. (Sole *et al.*, 2006). The low-energy method includes spontaneous emulsification, phase inversion methods, and solvent displacement method which will describe extensively in this paper.

2. High energy nanoemulsions formation methods

Nanoemulsion formation method that require high energy input from a mechanical device is known as a ‘High-energy method/ Dispersion method. These mechanical devices should have ability to generate intense disruptive forces to form smallest particle sizes. The high mechanical energy produced in ultrasonicators, microfluidizer, and high-pressure homogenizers used to provide strong disruptive forces, which leads to break up large droplets to nano-sized droplets and produce nanoemulsions with high kinetic energy (Gonçalves *et al.*, 2018). Depending on the type of mechanical device used, high energy nanoemulsions formation methods can be classified as follows.

2.1. High pressure homogenization

High-pressure homogenizers are mechanical devices which require high energy input to provide the smallest droplet size by created homogeneous flow (Figure 2). High-pressure homogenizers create intensely disruptive forces that gives driving force to form nanoemulsions of extremely small particle sizes (up to 1 nm) (Rai *et al.*, 2018; Kumar *et al.*, 2019). The method's particle size formation is dependent on a number of important variables, including sample composition, homogenizer type, and homogenizer working settings (energy intensity, time, and temperature) (Qian and McClements, 2011) . The size of the droplets in the resulting nanoemulsions is reduced when the energy intensity and temperature of homogenization are increased. The majority of beneficial nanoemulsions produced by high-pressure homogenization are used in food, medicinal, and biotechnological applications. (Hsieh *et al.*, 2012).

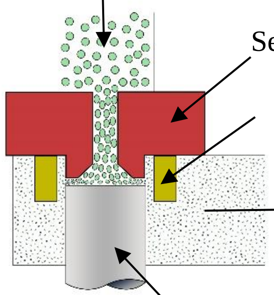

Figure 02: High pressure homogenization method

2.2. Microfludization

Microfludization is a mixing technique that use microfluidizer with high pressure displacement pump in order to produce small size nanoemulsions. In this method, two immiscible liquids (oil and water) from two opposite microchannels as shown in Figure 3 allowed to collide with each other at a common impingement area which leads to develop high pressure. The resulting Crude emulsion then passes repeatedly through the interaction chamber microfluidizer until the desired size of droplets is obtained (Çinar, 2017). By increasing the homogenization pressure, the passages through microchannel devices, the surfactant concentration, and the ratio of the dispersed to continuous phase viscosities, it is possible to reduce the droplet size of the dispersion phase of nanoemulsions. (Kentish *et al.*, 2008). Since the Microfludization method is very expensive, it is not suitable for the preparation of large amounts of nanoemulsions (Kentish *et al.*, 2008). But, Microfludization methods have been used to produce food ingredient nanoemulsions (Goh *et al.*, 2015)

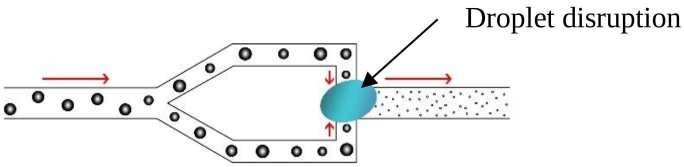

Figure 3: Microfluidizer

2.3. Ultrasonication

This technique makes use of ultrasonicators, which are just a probe that emits ultrasonic waves (Figure 4). The cavitation forces created by these ultrasonic waves cause the macroemulsion to shatter into a nanoemulsion. In terms of use and cleaning, this technique outperforms the high energy methods outlined earlier. (Leong *et al.*, 2009). Desired particle size and the stability of nanoemulsion can be achieved via varying the ultrasonic energy input and time. The process of acoustic cavitation is primarily responsible for providing physical shear in ultrasonic emulsification. (Jayasooriya *et al.*, 2004). Cavitation is a phenomenon in which microbubbles emerge, grow, and collide with one another as a result of changes in the acoustic wave's pressure. The collision of the microbubbles produces nano-sized droplets. (Leong *et al.*, 2009).

As per the previous studies through ultrasonication, nanoemulsions can be produced without surfactants (Gaikwad and Pandit, 2008). The production of medicinal and food ingredient nanoemulsions has made considerable use of ultrasonication. Compared to previous high energy technologies, food grade ultrasonication nanoemulsion has superior stability and smaller droplet size. (Salvia-Trujillo *et al.*, 2014). The main disadvantage of ultrasonication method is that it is not suitable for preparation of large volumes of nanoemulsions (Majid *et al.*, 2015).

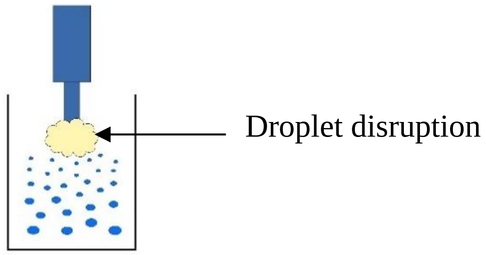

Figure 4: Ultrasonication

2.4. Jet disperser method

In this method, two or more jets of crude emulsion from bores which are opposing to each colloid with each other (Figure 5). Even though this is somewhat similar to microfluidizer method, crude emulsion colloid in a different way than in microfluidizer. The diameter of the bores in jet dispersers is around 0.3 – 0.5 mm (Buza and Dizdar, 2017). There is an “orifice plate” in the homogenizing nozzle which is coordinate energy dispersion of emulsion jet. Due to laminar elongation in front of the bores, droplets are overly disrupted. In the jet dispersers and orifice plate, there are no any moving parts. Therefore, they can be utilized even at high pressures up to 300-400 MPa (Buza and Dizdar, 2017).

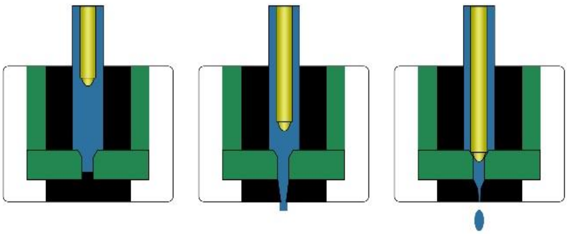

Figure 5: Jet disperser method

2.5. High shear stirring

Earlier, high-energy mixers were used for the preparation of nanoemulsions. By increasing the mixing intensity in these devices, droplet size of the internal phase can significantly decrease. But the preparation of less than 200-300 nm sizes nanoemulsions is very difficult with those techniques (Koroleva and Yurtov, 2012). Therefore, High shear mixers (Figure 6) were used in energy intensive processes such as homogenization, dispersion, emulsification, grinding, dissolving, and cell disruption in different fields like agricultural and food manufacturing and chemical reaction processes. These mixers also named as rotor stator mixers, high shear reactors, and high shear homogenizers (Azrini *et al.*, 2019). This system widely used in formation nanoemulsions from medium and low viscos liquids (Solans and Solé, 2012). This method connected with two different operations such as continuous and discontinuous. Continuous operation done with a smooth or toothed colloid mill consist of coaxially corn shaped rotors and stators. Further discontinuous operations followed up replacing an agitator or gear rim dispersion machine (Komaiko and Mcclements, 2016). The rotation of the rotor generates a lower pressure to flow liquid in and out between the blades of the stator, to result the emulsification (Solans, Morales and Homs, 2016). In the process large volume of bulk disperse phase slowly comminuted into tiny single droplets due to Top-Down approach (Bouchemal *et al.*, 2004). The inject of disperse phase directly into the shear field zone can enhance the mixing rate (Solans *etal.,* 2016).

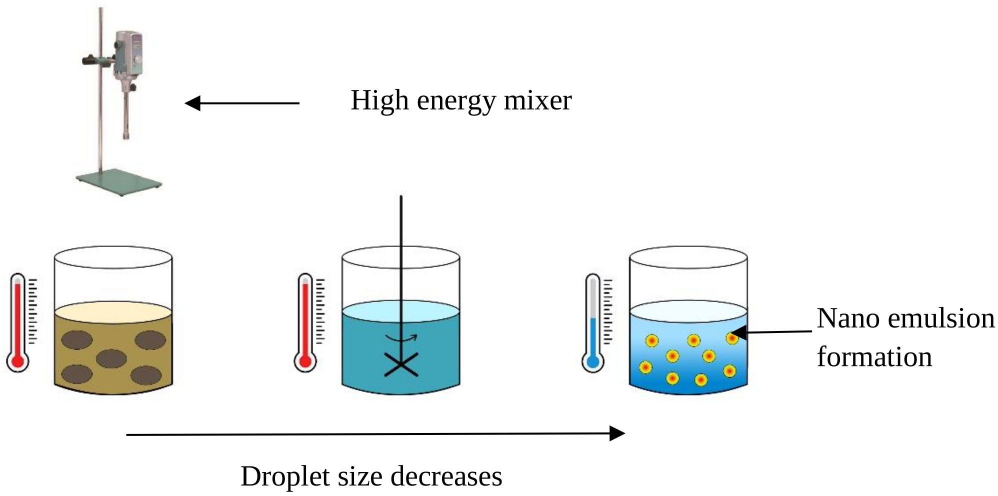

Figure 6: High shear stirring method

3. Low energy nanoemulsion formation methods

The methods that are using chemical energy stored in the components is referred to as low energy method or condensation method. Since this method require low energy for production of nanoemulsion systems, more energy efficient compared to high energy methods. Low energy nanoemulsion generation techniques use the systems intrinsic chemical energy and only gentle stirring to produce the nanoemulsions. (Solans and Solé, 2012). Since low-energy approaches call for large concentrations of surfactant, which have a negative impact on food formulation flavor and safety, they are typically not taken into consideration for the formulation of food grade nanoemulsions (Komaiko and Mcclements, 2016). This method further classified depending on whether changes in the surfactant spontaneous curvature are produced during the process or not.

3.1. The self-nano emulsification method

In self-nanoemulsification or so-called spontaneous emulsification method (Figure 7), Nano-emulsion formation activated by the rapid diffusion of surfactant and/or solvent molecules from the dispersed phase to the continuous phase without involving a change in the spontaneous curvature of the surfactant (Solans, Morales and Homs, 2016).

This approach does not require the use of any specialized machinery and can create nanoemulsions at ambient temperature. To create O/W nanoemulsions, water is gradually added to the solution of oil and surfactant while stirring gently at a steady temperature. The spontaneity of the emulsification process also depends on interfacial tension, interfacial and bulk viscosity, phase transition region, surfactant structure and its concentration (Bouchemal *et al.*, 2004). The main disadvantage of this method is the limited amount of the oil phase and the presence of the solvent (Maali and Mosavian, 2013). This method also used in delivery of bioactive food components (Meena *et al.*, 2012; Kheawfu *et al.*, 2018). Due to World Health Organization standards, financial constraints, and sensory considerations, high levels of surfactants and cosurfactants are not permitted in the food and pharmaceutical industries. Using the spontaneous emulsification technique, certain experiments have been done to lower the amount of cosurfactants as well as the ratio of surfactant to dispersed phase. (Ahmed *et al.*, 2012)

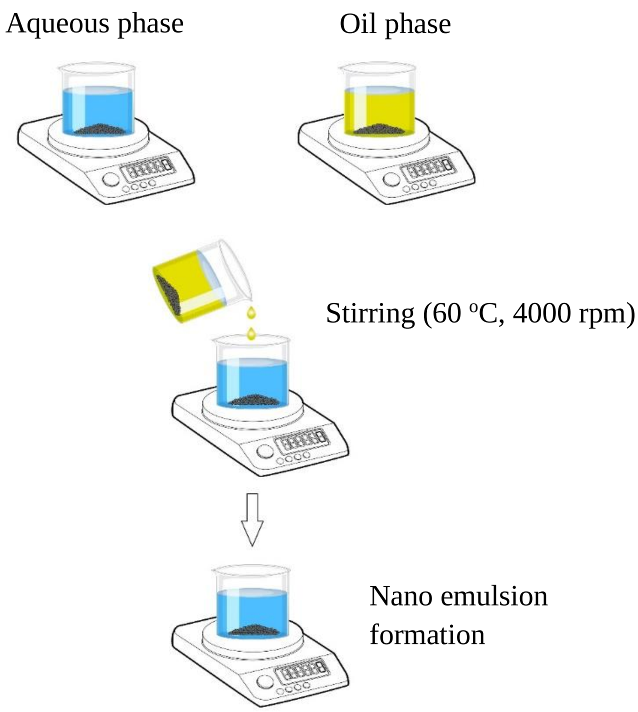

Figure 7: Self nano-emulsification method

3.2. Phase inversion emulsification method

These "phase inversion" approaches for emulsion production, which alter the surfactant spontaneous curvature from negative to positive (to create O/W emulsions) or from positive to negative (to create W/O emulsions) during the emulsification process, are another low energy technique. Surfactant's spontaneous curvature changes as a result of temperature and composition changes. (Solè *et al.*, 2010). The two primary categories of phase inversion emulsification techniques that are classified according to the change in spontaneous curvature caused by temperature or composition are phase inversion temperature (PIT) and phase inversion composition (PIC), which will be covered in great detail in this review.

3.2.1. Phase inversion temperature (PIT)

In the PIT emulsification process, variations in surfactant curvature brought on by temperature or variations in the lipophilic-hydrophilic balance of various nonionic surfactants with temperature (temperature emulsification method). Therefore, this technique can only be used with surfactants that are sensitive to temperature fluctuations, such as nonionic polyoxymethylene-type (POE) surfactants (Solans and Solé, 2012). For the reason that, when the temperature rises, a certain type of nonionic surfactant becomes less soluble in water.

The PIT method is used to combine the non-ionic surfactants, oil, and water at room temperature and gently stir them together to generate a nano-emulsion. The mixture is then progressively heated after that. Consequently, the surfactant's HLB gradually decreases from larger than 7 to less than 7. The phase of the emulsion may invert close to PIT (Gohtani and Prasert, 2014).

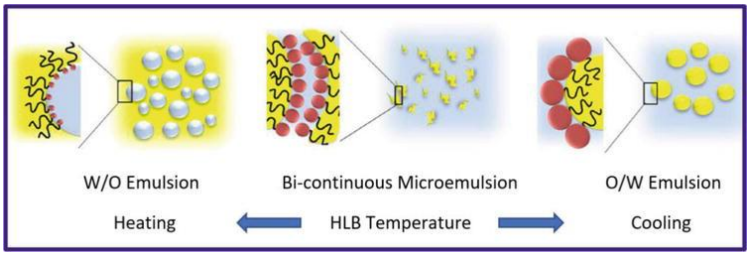

Figure 8: Schematic representation of PIT nanoemulsion formation method(Gohtani and Prasert, 2014)

As shown in Figure 8, at temperatures below the HLB, the ternary system of water, nonionic surfactant, and oil exhibits a macro-emulsion, with the nonionic surfactant's mostly hydrophilic heads being hydrated and gaining a disproportionately large volume in comparison to its lipophilic tail. The surfactant an O/W emulsion is created when the surfactant monolayer has a significant positive curvature toward the water phase. A W/O emulsion develops when the temperature rises higher, leading to the dehydration of the nonionic surfactant's heads and a negative curvature toward the water phase. It means the changes in temperature induce a change in the hydration of the head group of the surfactant, and a consequent change in its curvature (Gohtani and Prasert, 2014). A thermodynamically stable planar structure with zero curvature appears at the PIT/HLB or intermediate temperature, where amphiphile affinities for each phase are comparable and interfacial curvature is very low (almost close to zero). Lamellar liquid crystals or bicontinuous microemulsions are then formed. The main disadvantages of this method are; it is limited to nonionic surfactants, necessity of thermal energy, requiring phases such as liquid crystal or midrange microemulsion (Koroleva and Yurtov, 2012).

3.2.2. Phase inversion composition (PIC)

This process creates nanoemulsions at room temperature without the need of solvent or energy-intensive machinery required by high energy methods or spontaneous emulsification methods. Similar to the PIT method, the phase inversion composition (PIC) method achieves phase inversion by altering the system composition rather than the system temperature. (Sokolov, 2014).

Phase inversion in PIC can be accomplished by adding new substances or altering the concentration of compounds that are already present, which can change the hydrophilic lipophilic equilibrium of the system at constant temperature. This can happen when adding electrolytes, additional surfactants, alcohols, or changing the ratio of the oil-water phase while maintaining the same surfactant qualities to the system. (Salager *et al.*, 2004). Although other forms of nonionic surfactants may also be employed, POE type nonionic surfactants are typically used in the PIC method to create nanoemulsions. Surfactant POE chain hydration takes place as water is gradually added to the oil phase and as the volume of the water fraction rises. Similar to the HLB temperature in the PIT approach, the water phase's surfactant hydrophilic and lipophilic characteristics will balance out, and the surfactant's spontaneous curvature will alter to zero. A bi-continuous or lamellar structure is created during this transition. The structures of the surfactant layer with zero curvature change to having large positive curvature as more water is added, exceeding the transition composition (Kumar *et al.*, 2019). Phase inversion and the creation of nano-sized droplets are caused by this change in curvature. Phase inversion arises as a result of the system's composition altering (Solans and Solé, 2012). Similarly, other composition parameters, such as the addition of salt and pH changes, also cause nano-size emulsion droplets by transitional phase inversion (Sokolov, 2014). In literature this method also named as Emulsion inversion point (EIP) method. EIP method can be used to form O/W emulsion and to measure the amount of aqueous phase required for the formation of nanoemulsion with high stability.

3.3. D phase emulsification method

Very few investigations and studies were done on D- Phase emulsification (DPE) methods for last few decades and this method was first reported by Sagitani,Hattori, Nabeta and Nagai in 1983 (Yukuyama *et al.*, 2018). This method also consisting of surfactants, water and oil similar to the other methods. But the difference is, this method uses alkyl polyol as an extra component to form O/W nanoemulsions. These alkyl polyols are mor important in preparation of micro emulsions such as the act as co-surfactants and fully water dilutable(Wang *et al.*, 2015).Compared to other low energy methods, DPE method require low concentration of surfactants and there is no need of a solvent which is reportedly consume less energy than above mentioned PIC method.

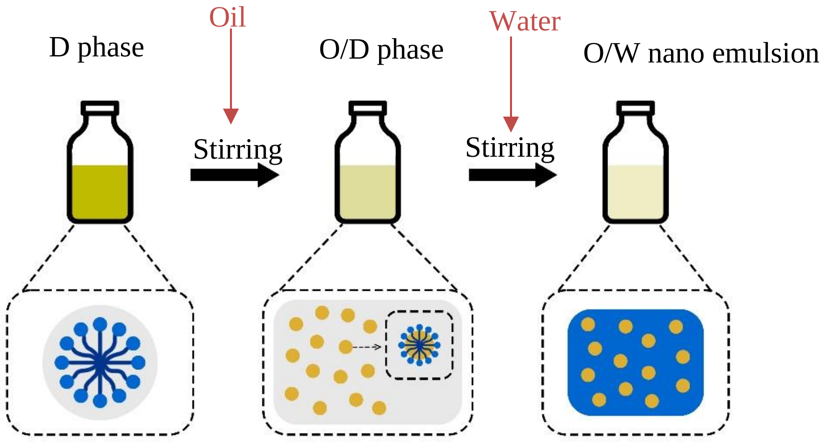

Figure 9: D space emulsification

3.4. Membrane emulsification method

This method involves the formation of a dispersed phase (droplets) through a pore of microporous membrane into a continuous phase under a force (Figure 10). It requires less surfactant/emulsifier concentration, less energy and produces emulsions with a narrow size distribution than the high-energy techniques. Due to low shear stress given in this method, some shear sensitive components such as starch and proteins are used for passing dispersed phase through the porous membrane into the continuous phase (Salem and Ezzat, 2019). The low flux rate of dispersed phase through membrane is the major limitation for maximizing the production of this method (Borthakur *et al.*, 2016).

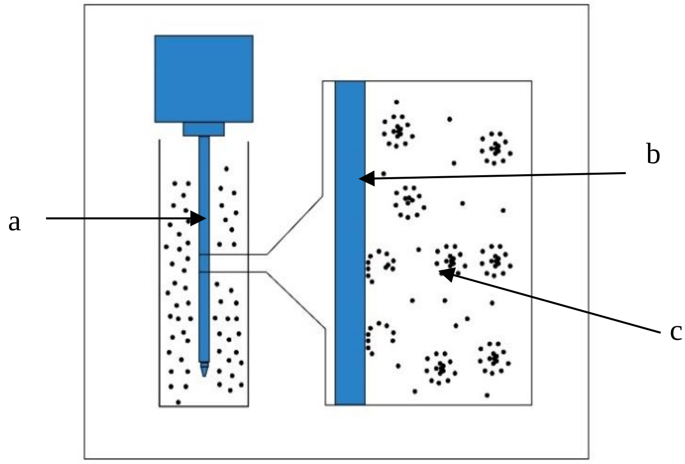

Figure 10: Membrane emulsification method (a: rotating microporous membrane; b: dispersed phase; c: stabilized droplets of colloidal particles)

3.5. Microemulsion dilution

Microemulsion dilution (MD) technique, A dilution technique carried out at a steady temperature is used to create nanoemulsion. The surfactant concentration necessary to ensure thermodynamic stability is decreased by rapidly diluting the oil-in-water microemulsion with a high volume of water. Production of nanoemulsion can be maximized easily through this technique. Due to the constant temperature and lack of the need for vigorous stirring, it uses less energy (Feng *et al.*, 2018).

3.6. Solvent displacement method

Solvent displacement method (SDM) method (Figure 11) also known as nanoprecipitation method which employed at room temperature with simple stirring and form nanoemulsions spontaneously. This process diffuses oil phase into continuous phase by combining it with low molecular weight organic solvents like alcohols and ketones, such as ethanol and acetone. Due to their low molecular weight, these organic solvents are miscible with aqueous phase. Nanoemulsions produced naturally as a result of organic solvent diffusion. The production of small-sized nanoemulsions even in the absence of mechanical stirring is mainly caused by the rapid diffusion of the solvent into the aqueous phase where interfacial tension between the two phases is reduced and the surface area is increased. After the formation of nanoemulsions, the organic solvent used is removed by vacuum evaporation technique.

The choice of emulsifying device is influenced mainly on some factors such as, volume of the emulsion, viscosities and phases of the emulsion, the type and concentration of the surfactant, the temperature, the size and size distribution of the droplets of the dispersion phase. (Jaiswal *et al*., 2015). According to the previous study conducted to achieve desired nanoemulsion formulation some parameters of emulsification like flow rate, density of interface, temperature and time of emulsification should be optimized (Jaiswal *et al.*, 2015).

Since this method needs high ratio of organic solvent to oil phase to obtain small sized nanoemulsion droplets (Çinar, 2017), solvent removal process causes several difficulties. Further, this method is limited to water-miscible solvents (Borthakur *et al.*, 2016). However, this technique is totally convenient for the preparation of monodisperse, small-sized polymeric nanoparticles and it is a reproducible, fast, and economical one-step manufacturing process (Buza and Dizdar, 2017).

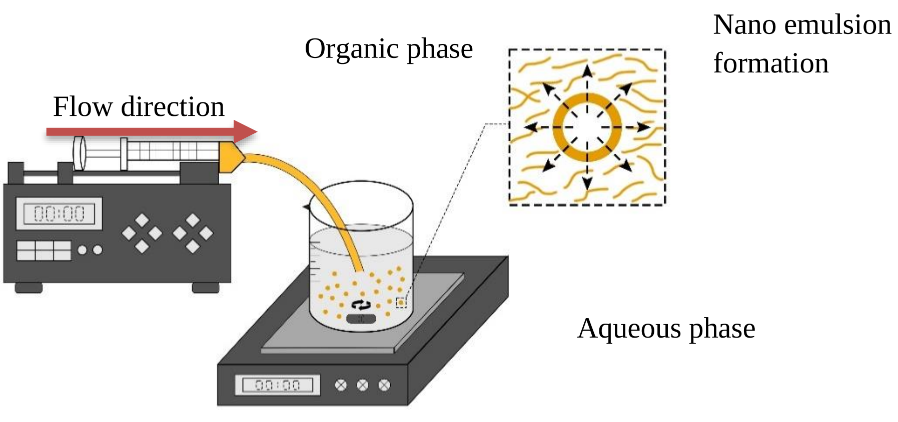

Figure 11: Solvent displacement method

4. Low energy Vs High energy nanoemulsion formation methods

There are pros and cons of using either technique in preparation of nanoemulsions and in their applications. In certain situations, the use of low-energy techniques may be favored to high-energy ones. For example, considering the initial capital cost of both methods, high-energy equipment is too large to overcome, then low-energy methods become the best solution since it does not require any expensive equipment's for the preparation of nanoemulsions. However, for the formation of nanoemulsions using Low energy method require higher levels of surfactant. In applications of food grade nanoemulsions usage of high concentrations of synthetic surfactants considered as the major drawback of low energy method. For example, in beverages where the final oil and surfactant concentrations are very low (<0.1%) (Komaiko and Mcclements, 2016). Nanoemulsions formed through Low energy techniques shows high stability at 4 ^{o}C and at ambient temperature and also there is no any alterations of size and the polydispersity over more than two months according to Singh, A.K et al (Singh *et al.*, 2018). Therefore, low energy methods especially spontaneous emulsification method has high possibility to use in nanomulsion based delivery systems (Singh *et al.*, 2018).

In addition, the temperature rise caused by homogenization inhibits the high-energy procedures for encapsulating numerous bioactive compounds. Other solutions exist, such using ice to keep the area around a homogenizer cool, which ultimately be more expensive than using high-energy methods. In these circumstances, isothermal techniques may be useful to encapsulate heat-sensitive compounds as high temperatures are not required (Komaiko and Mcclements, 2016). This means the low energy methods like self-emulsification or emulsion phase inversion methods may be useful for these types of encapsulates. Moreover, nanoemulsion through low energy formation method is more efficient for the formation of heat sensitive flavor ingredients (Saffarionpour, 2019).

Although low-energy processes offer some benefits over high-energy ones. At the meantime, only synthetic surfactants like Tweens and Spans have been demonstrated to function with low-energy methods (Komaiko and Mcclements, 2016). Natural emulsifiers like phospholipids, proteins, or polysaccharides would be ideal for many applications but may not be feasible due to the physicochemical factors involved in nanoemulsion production again by low-energy methods (Piorkowski and

McClements, 2014). Additionally, creating nanoemulsions requires rather large quantities of synthetic surfactants, which may be prohibitive for some applications for reasons of taste, safety, and cost (Guttoff *et al.*, 2015). Without a doubt, using natural emulsifiers with low-energy techniques might make the processes considerably more suitable in terms of cost and safety, particularly in applications involving food grade materials.

5. Applications of nanoemulsions

Due to the small size of droplets of the internal phase, relatively high kinetic stability and optical transparency compared to the conventional macroemulsions, nanoemulsions attract much attention of many researchers. The authors who extensively studied on the preparation and investigation of nanoemulsions emphasize great prospects for practical applications of these systems. Major studies on applications of nanoemulsions are related to the possibility of their use in encapsulation of medicines and their controlled delivery into the human body. Numerous studies have demonstrated that hydrophobic bioactives, such as vitamins, minerals, and nutraceuticals, can be integrated into nanoemulsions made using low-energy techniques. For instance, oil-in-water nanoemulsions can contain carotenoids, vitamin D, and vitamin E.(Saberi *et al.*, 2013; Zhang *et al.*, 2013; Guttoff *et al.*, 2015). Nanoemulsions formed by low-energy methods utilized in the production of antimicrobial delivery systems also (Esteban *et al.*, 2014).

In cosmetic industry, nanoemulsion used for dispersion of active ingredients in particular skin layer and for control delivery of lipophilic compounds due to having lipophilic part in nanoemulsion. Nanoemulsion can be easily appliedto skin since they are nontoxic and nonirritant. Even though there is little possibility of irritation by surfactants used for formation of nanoemulsion, that also can be avoided by using high energy equipment’s (Nor Bainun *at al.,* 2015). Using oil in water emulsion lipophilic drug delivery to the eye also improved. Recently, nasal administration of drugs is advantageous over oral administration through nanoemulsion technology due to increase on contact time between emulsion droplet and nasal mucosa. Not only that, the absorption also can be increased by solubilizing the drug in the inner phase of an emulsion.

Many studies were done in order to show the possibility of nanoemulsion to recover residue oil from reservoir rock due to having good penetration ability with smaller size of nanoemulsion than the pores in reservoir matrix rock (Nor Bainun *et al.*, 2015). There are several publications on possibility of usage of nanoemulsions in food industry specially using high energy formation methods, agrochemical and petroleum industries, in production of printer's ink and there will be many more applications that will ease our lives in near future.

6. Microemulsion and nano- emulsion

There is a little confusion on the two terms “microemulsion” and ‘nanoemulsion”. It is not technically accurate to predict the size of the emulsion from the phrases, but it does offer an indication. Thermodynamic stability of microemulsions is higher than that of nanoemulsions. Micrometer-sized emulsions are not what is meant by the word "microemulsion." The terms micelles, reverse micelles, rod-like micelles, lamella structure, bicontinuous structure, and swelling micelle are all used to describe the structure of a microemulsion. According to McClements (2012), nano-emulsions and microemulsions can be distinguished easily based on free energy. He also noticed that the free energy of nanoemulsions or nanoscale droplets distributed in a continuous phase, or the disperse state was larger than that of discrete states of oil and water. A nano-emulsion might be kinetically stable because there is an energy barrier separating the two states. The dispersed state's free energy is smaller for microemulsions than the separate states.

7. Advantages and disadvantage of high energy method and low energy method

Table 01: Advantages and disadvantages of using each method in high energy emulsification

High energy method	Advantages	Disadvantages
High-pressure homogenizer	High stability nano emulsion production simple production method (Hidajat *et al.*, 2020)	Utilizes a lot of energy and frequently elevates the temperature during processing (Bhosale *et al.*, 2014; Buza and Dizdar, 2017)
	Either high- or low-temperature processing is possible and effectively applied to thermolabile materials (Modarres-Gheisari *et al.*, 2019)	Due to the high pressure, powerful breaking forces including cavitation and shearing cause severe turbulent flows (Modarres-Gheisari *et al.*, 2019).
	High mixing efficiency (Yuh-Fun and Chung, 1999)	Proteins, enzymes, and nucleic acids are examples of thermolabile substances that might suffer damage (Changediya *et al.*, 2019).
	Useful in simple scaling up, avoiding organic solvents, and quick processing (Maali and Mosavian, 2013).	
	More effective at emulsifying viscous fluids, and it generated less thermal stresses on the system (Yuh-Fun and Chung, 1999).	
Microfludization	Submicron can produce even at lower emulsifier concentration (Pinnamaneni *et al.*, 2003)	In appropriate for the preparation of large-scale Nano emulsion concentrations
	The submicron has tiny diameter and It exhibit less droplet diameter over compared time than other processes (Mahdi Jafari *et al.*, 2006).	It is not suitable for use in high pressure and extended emulsification times due to the re-coalescence of emulsion droplets (Maali and Mosavian, 2013).
	Multiple nanoemulsions could be produced with regulated dispersed phase droplet sizes (Buza and Dizdar, 2017).	Relatively low efficiency on encapsulation (Vuillemard, 1991)

		It's more difficult to operate, clean and scale-up (Yuh-Fun and Chung, 1999)
Ultrasonication	High energy efficacy and good emulsion stability (Taha *et al.*, 2020)	Not suitable for emulsion systems containing heat-sensitive components since it produces high heat (Modarres-Gheisari *et al.*, 2019).
	Produce kinetically stable nano emulsions (Buza and Dizdar, 2017)	Low mixing efficiency (Yuh-Fun and Chung, 1999)
	High potential for making nanometer-sized droplets to prepare Pickering emulsions (Albert *et al.*, 2019)	As the power levels and concentration of the surfactant increase, the particle size of the dispersed phase in nanoemulsions decreases (Buza and Dizdar, 2017).
		not appropriate for making significant amounts of nanoemulsions (Buza and Dizdar, 2017)
Jet disperser method	Due to the high pressure and tremendous shearing action, it provides extremely fine nanoemulsions (Kumar, 2014)	
Rotor-Stator emulsification method	It is feasible to construct an emulsion at a greater disperse phase ratio in order to improve the overall viscosity of the emulsion (van der Schaaf and Karbstein, 2018).	It is challenging to produce emulsions with average droplet sizes less than 200/300 nm (Changediya *et al.*, 2019).
	Rotor-stator equipment performs significantly better than high-pressure homogenizers when it comes to emulsifiers with delayed adsorption kinetics (van der Schaaf and Karbstein, 2018).	Difficult to obtain homogenized nanoemulsions specially when operating close to the probe's maximum volume (Albert *et al.*, 2019).
	Slow-adsorbing emulsifiers can even stabilize very small droplets in the operations due to the extended residence durations (Donsì *et al.*, 2013)	During the emulsification process, the high shear rate that occurs between the rotor and the stator may destabilize or deform delicate particles or aggregates (Destribats *et al.*, 2013).

Table 02: Advantages and disadvantages of using each method in low energy emulsification

Low energy method	**Advantages**	**Disadvantages**
The self-nano emulsification method	This technique doesn't need any extra equipment in order to create nanoemulsions at ambient temperature (Changediya *et al.*, 2019). There are no mechanical forces or temperature changes necessary to create nanoemulsions (Saffarionpour, 2019). The high efficacy method to form nano particles with potential concentrations in a large scale of sizes due to kinetic stability (Anton and Vandamme, 2011)	Spontaneous emulsification only haens under specific conditions (Yukuyama *et al.*, 2016).
Phase inversion (PI) nano emulsification method	Avoid forming hexagonal and phases during process which will be more efficient in forming nano emulsion without intermediate phases (Zhang *et al.*, 2021). The PI method use to emulsify oils which has low polarity with poor water solubility and low surface tension (Singh *et al.*, 2014)	These techniques' drawbacks include complexity, the need for accuracy, and the usage of artificial surfactants (Forgiarini *et al.*, 2001; Izquierdo *et al.*, 2005) Very high temperatures are required (Saffarionpour, 2019). Phase inversion temperature (PIT) method only can use for the temperature sensitive surfactants (Solans and Solé, 2012).
D phase emulsification method	Even in the law concentration of the surfactant's emulsions will be formed (Yukuyama *et al.*, 2018; Safaya and Rotliwala, 2020) This method not required of well-balanced surfactant systems with compared to PI method (Yukuyama *et al.*, 2018; Pirvaram *et al.*, 2019) Do not required any specific solvents for the nano emulsion preparation with compared to self-nano emulsion method (Tadros, 2004)	

Membrane emulsification method	The resulting nano emulsions has low droplet size distribution and longer shelf life due to emulsifier use for avoid the coalescence (Lee *et al.*, 2013). Minimal energy use and low running costs (Thompson *et al.*, 2011) Eliminating the generation of heat during the emulsification procedure reduces the possibility of thermosensitive particles and emulsions becoming unstable (Albert *et al.*, 2019).	Low viscosity liquids can only flow via the microchannel, posing a restriction on their use (Nie *et al.*, 2008).
Microemulsion dilution	The process involve with low energy consumption and no temperature changes during the process (Safaya and Rotliwala, 2020). In the situation of narrow size distribution use as the membrane it forms monodisperse droplets (Solanki and Murthy, 2011)	Require specific controlled porous glass membranes for the emulsification process (Badnjevic, 2017).
Solvent displacement method	The emulsification process can be conduct in the room temperature without giving any specific conditions (Badnjevic, 2017). The lipophilic drug encapsulation efficiency is higher greater than 70 % (Quintanar-Guerrero *et al.*, 1998)	When compared to the droplet size formation the requirement of solvent to oil ratio is high (Fryd and Mason, 2012a) During emulsification, large amounts of water are removed from the suspension, and this lead to decreasing the effectiveness of encapsulation (Pinto Reis *et al.*, 2006).

8. Conclusions

Nanoemulsions are taking the increase attraction of many scientists and expected to gradually become the area of research and development. Up to date, it has been demonstrated that nanoemulsions can preserve labile drugs, regulate drug release, boost drug solubility, increase bioavailability, and minimize patient variability. The preparation of nanoemulsions basically divided into two methods. High energy and Low energy method. There are pros and cons in both high energy and low energy nanoemulsion formation methods. However, many challenges still need to be overcome in terms of extensive energy use and considering the safety at the same time. To overcome the economic issues, nanoemulsion formulation by low energy methods become the suitable option. But the major limitation in low energy method is the usage of high concentrations of artificial surfactants which directly limits the food industrial applications. The instability of nanoemulsion limits its applicability in varies fields. The stability can improve by Controlling several aspects such as concentration of surfactant and cosurfactant, the type of oil phase, process variables, and the addition of additives used across the inter phases of nanoemulsion formulation.

9. Further directions

Hence this is the high time to do extensive studies on usage of natural emulsifiers, such as lecithin, pea protein instead of artificial emulsifiers in low-energy methods. Also, natural-based emulsifiers, in conjunction with new emulsion technology breakthroughs, have the potential to lay the groundwork for a new generation of healthful and nutritious food products. Further, few researches were done to increase the efficacy of antimicrobial, antioxidant activity of herbal plants by encapsulating to nanoemulsion. But studies on seagrass based herbal plants on this purpose is very poor even though they are showing good antimicrobial activity. Therefore, sea grass essential oil based nanoemulsion formation using any of above-mentioned techniques and evaluate the efficacy of each method will be really helpful in the field of medicine in near future. Also, the encapsulation of natural colorants from different plant species should study on nanoemulsification for the application of foods. This would be help in the future studies to cover-up the most critical capability gaps connected to green and food-grade nanoemulsions.

10. References

Acosta, E., 2009. 'Bioavailability of nanoparticles in nutrient and nutraceutical delivery', *Current Opinion in Colloid and Interface Science*, 14(1), 3–15. Available at: https://doi.org/10.1016/j.cocis.2008.01.002.

Ahmed, K., Li, Y., McClements, D.J. and Xiao, H., 2012. Nanoemulsion-and emulsion-based delivery systems for curcumin: Encapsulation and release properties. *Food chemistry*, 132(2), 799-807.

Albert, C., Beladjine, M., Tsapis, N., Fattal, E., Agnely, F. and Huang, N., 2019. Pickering emulsions: Preparation processes, key parameters governing their properties and potential for pharmaceutical applications. *Journal of Controlled Release*, 309, 302-332.

Anton, N., Benoit, J.P. and Saulnier, P., 2008. Design and production of nanoparticles formulated from nano-emulsion templates—a review. *Journal of Controlled Release*, 128(3), 185-199.

Anton, N. and Vandamme, T.F., 2011. Nano-emulsions and micro-emulsions: clarifications of the critical differences. *Pharmaceutical Research*, 28, 978-985.

Azmi, N.A.N., Elgharbawy, A.A., Motlagh, S.R., Samsudin, N. and Salleh, H.M., 2019. Nanoemulsions: Factory for food, pharmaceutical and cosmetics. *Processes*, 7(9), 617.

Badnjevic, A. ed., 2017. *CMBEBIH 2017: Proceedings of the International Conference on Medical and Biological Engineering 2017*, 62, Springer.

Bhosale, R.R., Osmani, R.A., Ghodake, P.P., Shaikh, S.M. and Chavan, S.R., 2014. Nanoemulsion: A review on novel profusion in advanced drug delivery. *Indian Journal of Pharmaceutical and Biological Research*, 2(1), 122.

Borthakur, P., Boruah, P.K., Sharma, B. and Das, M.R., 2016. Nanoemulsion: Preparation and its application in food industry. In *Emulsions*, 153-191. Academic Press.

Bouchemal, K., Briançon, S., Perrier, E. and Fessi, H., 2004. Nano-emulsion formulation using spontaneous emulsification: solvent, oil and surfactant optimisation. *International Journal of Pharmaceutics*, 280(1-2), 241-251.

Changediya, V.V., Jani, R. and Kakde, P., 2019. A review on nanoemulsions: A recent drug delivery tool. *Journal of Drug Delivery and Therapeutics*, 9(5), 185-191.

Çınar, K., 2017. A review on nanoemulsions: preparation methods and stability. *Trakya Üniversitesi Mühendislik Bilimleri Dergisi*.

Destribats, M., Wolfs, M., Pinaud, F., Lapeyre, V., Sellier, E., Schmitt, V. and Ravaine, V., 2013. Pickering emulsions stabilized by soft microgels: influence of the emulsification process on particle interfacial organization and emulsion properties. *Langmuir*, 29(40), 12367-12374.

Esteban, P.P., Alves, D.R., Enright, M.C., Bean, J.E., Gaudion, A., Jenkins, A.T.A., Young, A.E. and Arnot, T.C., 2014. Enhancement of the antimicrobial properties of bacteriophage-K via stabilization using oil-in-water nano-emulsions. *Biotechnology Progress*, 30(4), 932-944.

Feng, J., Zhang, Q.I., Liu, Q.I., Zhu, Z., McClements, D.J. and Jafari, S.M., 2018. Application of nanoemulsions in formulation of pesticides. In *Nanoemulsions*, 379-413. Academic Press.

Forgiarini, A., Esquena, J., Gonzalez, C. and Solans, C., 2001. Formation of nano-emulsions by low-energy emulsification methods at constant temperature. *Langmuir*, 17(7), 2076-2083.

Fryd, M.M. and Mason, T.G., 2012. Nanoinclusions in cryogenically quenched nanoemulsions. *Langmuir*, 28(33), 12015-12021.

Gaikwad, S.G. and Pandit, A.B., 2008. Ultrasound emulsification: effect of ultrasonic and physicochemical properties on dispersed phase volume and droplet size. *Ultrasonics Sonochemistry*, 15(4), 554-563.

Goh, P.S., Ng, M.H., Choo, Y.M., Nasrulhaq Boyce, A. and Chuah, C.H., 2015. Production of nanoemulsions from palm-based tocotrienol rich fraction by microfluidization. *Molecules*, 20(11), 19936-19946.

Gohtani, S. and Prasert, W., 2014. Nano-emulsions; Emulsification using low energy methods. *Japan Journal of Food Engineering*, 15(3), 119-130.

Gonçalves, A., Nikmaram, N., Roohinejad, S., Estevinho, B.N., Rocha, F., Greiner, R. and McClements, D.J., 2018. Production, properties, and applications of solid self-emulsifying delivery systems (S-SEDS) in the food and pharmaceutical industries. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 538, 108-126.

Gupta, A., Eral, H.B., Hatton, T.A. and Doyle, P.S., 2016. Nanoemulsions: formation, properties and applications. *Soft Matter*, 12(11), 2826-2841.

Guttoff, M., Saberi, A.H. and McClements, D.J., 2015. Formation of vitamin D nanoemulsion-based delivery systems by spontaneous emulsification: factors affecting particle size and stability. *Food Chemistry*, 171, 117-122.

Hidajat, M.J., Jo, W., Kim, H. and Noh, J., 2020. Effective droplet size reduction and excellent stability of limonene nanoemulsion formed by high-pressure homogenizer. *Colloids and Interfaces*, 4(1), 5.

Hsieh, C.W., Li, P.H., Lu, I.C. and Wang, T.H., 2012. Preparing glabridin-in-water nanoemulsions by high pressure homogenization with response surface methodology. *Journal of Oleo Science*, *61*(9), 483-489.

Izquierdo, P., Feng, J., Esquena, J., Tadros, T.F., Dederen, J.C., Garcia, M.J., Azemar, N. and Solans, C., 2005. The influence of surfactant mixing ratio on nano-emulsion formation by the pit method. *Journal of colloid and interface science*, 285(1), 388-394.

Jaiswal, M., Dudhe, R. and Sharma, P.K., 2015. Nanoemulsion: an advanced mode of drug delivery system. *3 Biotech*, 5, 123-127.

Jayasooriya, S.D., Bhandari, B.R., Torley, P. and D'arcy, B.R., 2004. Effect of high power ultrasound waves on properties of meat: a review. *International Journal of Food Properties*, 7(2), 301-319.

Kentish, S., Wooster, T.J., Ashokkumar, M., Balachandran, S., Mawson, R. and Simons, L., 2008. The use of ultrasonics for nanoemulsion preparation. *Innovative Food Science & Emerging Technologies*, 9(2), 170-175.

Kheawfu, K., Pikulkaew, S., Rades, T., Müllertz, A. and Okonogi, S., 2018. Development and characterization of clove oil nanoemulsions and self-microemulsifying drug delivery systems. *Journal of Drug Delivery Science and Technology*, 46, 330-338.

Komaiko, J.S. and McClements, D.J., 2016. Formation of food-grade nanoemulsions using low-energy preparation methods: A review of available methods. *Comprehensive Reviews in Food Science and Food Safety*, 15(2), 331-352.

Koroleva, M.Y. and Yurtov, E.V., 2012. Nanoemulsions: the properties, methods of preparation and promising applications. *Russian Chemical Reviews*, 81(1), 21.

Kumar, M., Bishnoi, R.S., Shukla, A.K. and Jain, C.P., 2019. Techniques for formulation of nanoemulsion drug delivery system: A review. *Preventive Nutrition and Food Science*, 24(3), 225.

Kumar, S., 2014. Role of nano-emulsion in pharmaceutical sciences: A review. *AJRPSB*, *2*(1), 1-5.

Lee, L.L., Niknafs, N., Hancocks, R.D. and Norton, I.T., 2013. Emulsification: mechanistic understanding. *Trends in Food Science & Technology*, 31(1), 72-78.

Leong, T.S.H., Wooster, T.J., Kentish, S.E. and Ashokkumar, M., 2009. Minimising oil droplet size using ultrasonic emulsification. *Ultrasonics Sonochemistry*, 16(6), 721-727.

Maali, A. and Mosavian, M.H., 2013. Preparation and application of nanoemulsions in the last decade (2000–2010). *Journal of Dispersion Science and Technology*, 34(1), 92-105.

Maa, Y.F. and Hsu, C.C., 1999. Performance of sonication and microfluidization for liquid–liquid emulsification. *Pharmaceutical Development and Technology*, 4(2), 233-240.

Mahdi Jafari, S., He, Y. and Bhandari, B., 2006. Nano-emulsion production by sonication and microfluidization—a comparison. *International Journal of Food*

Properties, 9(3), 475-485.

Majid, I., Nayik, G.A. and Nanda, V., 2015. Ultrasonication and food technology: A review. *Cogent Food & Agriculture*, 1(1), 1071022.

McClements, D.J., 2012. Nanoemulsions versus microemulsions: Terminology, differences, and similarities. *Soft Matter*, 8(6), 1719-1729.

Meena, A.K., Sharma, K., Kandaswamy, M.U.R.U.G.E.S.H., Rajagopal, S. and Mullangi, R., 2012. Formulation development of an albendazole self-emulsifying drug delivery system (SEDDS) with enhanced systemic exposure. *Acta Pharmaceutica*, 62(4), 563-580.

Modarres-Gheisari, S.M.M., Gavagsaz-Ghoachani, R., Malaki, M., Safarpour, P. and Zandi, M., 2019. Ultrasonic nano-emulsification–A review. *Ultrasonics Sonochemistry*, 52, 88-105.

Nie, Z., Park, J.I., Li, W., Bon, S.A. and Kumacheva, E., 2008. An "inside-out" microfluidic approach to monodisperse emulsions stabilized by solid particles. *Journal of the American Chemical Society*, 130(49), 16508-16509.

Nor Bainun, I., Alias, N.H. and Syed-Hassan, S.S.A., 2015. Nanoemulsion: formation, characterization, properties and applications-a review. *Advanced Materials Research*, 1113, 147-152.

Pinnamaneni, S., Das, N.G. and Das, S.K., 2003. Comparison of oil-in-water emulsions manufactured by microfluidization and homogenization. *Die Pharmazie-An International Journal of Pharmaceutical Sciences*, 58(8), 554-558.

Piorkowski, D.T. and McClements, D.J., 2014. Beverage emulsions: Recent developments in formulation, production, and applications. *Food Hydrocolloids*, 42, 5-41.

Pirvaram, A., Sadrameli, S.M. and Abdolmaleki, L., 2019. Energy management of a household refrigerator using eutectic environmental friendly PCMs in a cascaded condition. *Energy*, 181, 321-330.

Qian, C. and McClements, D.J., 2011. Formation of nanoemulsions stabilized by model food-grade emulsifiers using high-pressure homogenization: Factors affecting particle size. *Food Hydrocolloids*, 25(5), 1000-1008.

Quintanar-Guerrero, D., Allémann, E., Fessi, H. and Doelker, E., 1998. Preparation techniques and mechanisms of formation of biodegradable nanoparticles from preformed polymers. *Drug Development and Industrial Pharmacy*, 24(12), 1113-1128.

Rai, V.K., Mishra, N., Yadav, K.S. and Yadav, N.P., 2018. Nanoemulsion as pharmaceutical carrier for dermal and transdermal drug delivery: Formulation development, stability issues, basic considerations and applications. *Journal of Controlled Release*, 270, 203-225.

Reis, C.P., Neufeld, R.J., Ribeiro, A.J. and Veiga, F., 2006. Nanoencapsulation I. Methods for preparation of drug-loaded polymeric nanoparticles. *Nanomedicine: Nanotechnology, Biology and Medicine*, 2(1), 8-21.

Saberi, A.H., Fang, Y. and McClements, D.J., 2013. Fabrication of vitamin E-enriched nanoemulsions: Factors affecting particle size using spontaneous emulsification. *Journal of Colloid and Interface Science*, 391, 95-102.

Safaya, M. and Rotliwala, Y.C., 2020. Nanoemulsions: A review on low energy formulation methods, characterization, applications and optimization technique. *Materials Today: Proceedings*, 27, 454-459.

Saffarionpour, S., 2019. Preparation of food flavor nanoemulsions by high-and low-energy emulsification approaches. *Food Engineering Reviews*, 11(4), 259-289.

Salager, J.L., Forgiarini, A., Marquez, L., Pena, A., Pizzino, A., Rodriguez, M.P. and Rondon-Gonzalez, M., 2004. Using emulsion inversion in industrial processes. *Advances in Colloid and Interface Science*, 108, 259-272.

Salem, M.A. and Ezzat, S.M., 2019. Nanoemulsions in food industry. *Some New Aspects of Colloidal Systems in Foods*, 2, 238-267.

Salvia-Trujillo, L., Rojas-Graü, M.A., Soliva-Fortuny, R. and Martín-Belloso, O., 2014. Impact of microfluidization or ultrasound processing on the antimicrobial activity against Escherichia coli of lemongrass oil-loaded nanoemulsions. *Food*

Control, 37, 292-297.

Singh, A.K., Yadav, T.P., Pandey, B., Gupta, V. and Singh, S.P., 2019. Engineering nanomaterials for smart drug release: recent advances and challenges. *Applications of Targeted Nano Drugs and Delivery Systems*, 411-449.

Singh, P.K., Iqubal, M.K., Shukla, V.K. and Shuaib, M., 2014. Microemulsions: current trends in novel drug delivery systems. *Journal of Pharmaceutical, Chemical and Biological Sciences*, 1(1), 39-51.

Sokolov, Y.V., 2014. Nanoemulsion formation by low-energy methods: a review. *News of Pharmacy*, 3(79), 16-19.

Solanki, J.N. and Murthy, Z.V.P., 2011. Controlled size silver nanoparticles synthesis with water-in-oil microemulsion method: a topical review. *Industrial & Engineering Chemistry Research*, 50(22), 12311-12323.

Solans, C., Morales, D. and Homs, M., 2016. Spontaneous emulsification. *Current Opinion in Colloid & Interface Science*, 22, 88-93.

Solans, C. and Solé, I., 2012. Nano-emulsions: formation by low-energy methods. *Current Opinion in Colloid & Interface Science*, 17(5), 246-254.

Solè, I., Pey, C.M., Maestro, A., González, C., Porras, M., Solans, C. and Gutiérrez, J.M., 2010. Nano-emulsions prepared by the phase inversion composition method: Preparation variables and scale up. *Journal of Colloid and Interface Science*, 344(2), 417-423.

Tadros, T., 2004. Application of rheology for assessment and prediction of the long-term physical stability of emulsions. *Advances in Colloid and Interface Science*, 108, 227-258.

Taha, A., Ahmed, E., Ismaiel, A., Ashokkumar, M., Xu, X., Pan, S. and Hu, H., 2020. Ultrasonic emulsification: An overview on the preparation of different emulsifiers-stabilized emulsions. *Trends in Food Science & Technology*, 105, 363-377.

Tarhan, O. and Spotti, M.J., 2021. Nutraceutical delivery through nano-emulsions: General aspects, recent applications and patented inventions. *Colloids and Surfaces B: Biointerfaces*, 200, 111526.

Thompson, K.L., Armes, S.P. and York, D.W., 2011. Preparation of pickering emulsions and colloidosomes with relatively narrow size distributions by stirred cell membrane emulsification. *Langmuir*, 27(6), 2357-2363.

van der Schaaf, U.S. and Karbstein, H.P., 2018. Fabrication of nanoemulsions by rotor-stator emulsification. In *Nanoemulsions*, 141-174. Academic Press.

Vuillemard, J.C., 1991. Recent advances in the large-scale production of lipid vesicles for use in food products: microfluidization. *Journal of Microencapsulation*, 8(4), 547-562.

Wang, W., Wei, H., Du, Z., Tai, X. and Wang, G., 2015. Formation and characterization of fully dilutable microemulsion with fatty acid methyl esters as oil phase. *ACS Sustainable Chemistry & Engineering*, 3(3), 443-450.

Yukuyama, M.N., Ghisleni, D.D.M., Pinto, T.D.J.A. and Bou-Chacra, N.A., 2016. Nanoemulsion: process selection and application in cosmetics–a review. *International Journal of Cosmetic Science*, 38(1), 13-24.

Yukuyama, M.N., Oseliero, P.L.F., Kato, E.T.M., Lobënberg, R., de Oliveira, C.L.P., de Araujo, G.L.B. and Bou-Chacra, N.A., 2018. High internal vegetable oil nanoemulsion: D-phase emulsification as a unique low energy process. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 554, 296-305.

Zhang, L., Hayes, D.G., Chen, G. and Zhong, Q., 2013. Transparent dispersions of milk-fat-based nanostructured lipid carriers for delivery of β-carotene. *Journal of Agricultural and Food Chemistry*, 61(39), 9435-9443.

Zhang, W., Qin, Y., Chang, S., Zhu, H. and Zhang, Q., 2021. Influence of oil types on the formation and stability of nano-emulsions by D phase emulsification. *Journal of Dispersion Science and Technology*, 42(8), 1225-1232.