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Treball Final de Grau

Study of the formation of edible coatings from nanoemulsions with vegan components.

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What we know is a drop, what we don't know is an ocean.

Isaac Newton

CONTENTS

SUMMARY

In recent years, nanotechnology has generated several advances in various fields, one of them in the field of food industry, –with the development of nanoemulsions in the food products. Nanoemulsions have led to an improvement in the preservation and increased shelf-life of foods, such as fruits and vegetables. There are many applications where they show an improvement –That is why this work is about the study of edible coatings used on food made from nanoemulsions with vegan components. The main characteristics of nanoemulsions are explained: they are made up of two immiscible liquids which in most cases are water and oil, one of which forms droplets (dispersed phase) in the other (continuous phase); depending on the size of these droplets, the nanoemulsion may be transparent or not; they are thermodynamically unstable although they can achieve kinetic stability that can keep them stable for an extended period of time. The methods by which nanoemulsions can be formed are divided into high and low energy methods. In high-energy methods, the most widely used at the industrial level is high-pressure valve homogenization. In low-energy methods, phase inversion temperature is the most important method. Edible nanoemulsion coatings have a wide variety of applications, it should be remarkable its applications as a food preservative. These coatings reduce the growth of bacteria and fungi, so the shelf-life of food is improved.

Keywords: Nanoemulsions, edible coatings, vegan.

RESUM

En els darrers anys, la nanotecnologia ha generat un gran número d'avenços en diversos camps, un d'ells en el camp de la indústria alimentària, −amb el desenvolupament de nanoemulsions en els productes alimentaris. Les nanoemulsions han suposat una millora a l'hora de la conservació i augment de la vida útil d'aliments com fruites i verdures, entre d'altres. No són poques les aplicacions on demostren una millora −Per això aquest treball tracta de l'estudi de recobriments comestibles fets a partir de nanoemulsions amb components vegans que s'utilitzen sobre aliments. Primerament, s'exposen les principals característiques de les nanoemulsions: estan formades per dos líquids immiscibles que en la majoria dels casos son l'aigua i l'oli, un d'aquests dos forma gotes (fase dispersa) en l'altre (fase continua); depenent de la mida d'aquestes gotes, la nanoemulsió podrà ser més transparent o menys; són termodinàmicament inestables tot i que poden aconseguir una estabilitat cinètica que les pot mantenir estables en un període prolongat de temps. Els mètodes pels quals es poden formar les nanoemulsions es divideixen en els mètodes d'alta i de baixa energia. En els mètodes d'alta energia destaca el més utilitzat a nivell industrial que es la homogeneïtzació per vàlvula a alta pressió. En els mètodes de baixa energia cal destacar la inversió de fases per temperatura com el mètode més important. Finalment, es comenten les aplicacions que tenen els recobriments comestibles fets amb nanoemulsions dins de la indústria alimentaria, d'entre elles la més remarcable com a conservant d'aliments. Aquests recobriments redueixen el creixement de bacteris i fongs, de manera que es millora la vida útil dels aliments.

Paraules clau: Nanoemulsions, recobriments comestibles, vegà.

1. INTRODUCTION

Food industry is facing the challenge of increasing the shelf-life of its products. Chemical treatment is one of the techniques that are normally used to prolong shelf-life and reduce food spoilage, such as the use of pesticides in fruits and vegetables which can directly affect human health (Flores-López et al., 2015). Nowadays, consumer demands are increasing in terms of safety, healthy, and ecological food products (Montero et al., 2016). The application of edible coatings onto the food surface is a treatment that allows to enlarge shelf-life of food products, maintaining their safety and quality (Flores-López et al., 2015). Edibility is an advantage that plastics cannot provide. Nanoemulsions have knocked at the door of food industry proposing the great number of advantages that they can provide. Furthermore, active components can be added to the coating, thus giving specific functional properties (Acevedo et al., 2017). This fact has grown the number of applications that nanoemulsions can provide when combined with active ingredients, giving lots of benefits.

In nanotechnology applied to food, nanoemulsions are one of the most important applications, because they can carry and liberate lipophilic components such as nutraceutical ingredients, flavourings, antimicrobial and antioxidant agents. A big advantage that nanoemulsions present are their capacity to enhance the bioavailability of components encapsulated (Cardoso-Ugarte and Jiménez-Mungía, 2015). More reasons to encapsulate lipophilic components are to improve the facility in the manipulation, utilization, and application within a product; to control the speed and location at the time of the release; or to cover it from chemical degradation (McClements, 2011).

The droplets of nanoemulsions are much smaller than visible light wavelengths, so the droplets scatter weakly the light waves, what gives the solution an appearance optically transparent or translucid. That is why nanoemulsions have an important application on beverages or other transparent products (Mason et al., 2006; McClements, 2011).

Nanoemulsions could be formed by two different types of methods: high-energy methods and low-energy methods. High-energy methods (e.g., high-pressure valve homogenization and ultrasonication) use mechanical energy, while low-energy methods (e.g., spontaneous emulsification and phase inversion) use energy stored in the components (Velikov and Pelan, 2008). Mechanical methods predominate in industry because they can produce on a large-scale and continuously (Hakansson and Rayner, 2018).

As those coatings may be applied to fruits and vegetables, which are vegan products, it is important to conserve this condition when the coating is applied, that is why those coatings must be made from vegan components, in other words, non-containing animal derived ingredients.

2. OBJECTIVES

The main objective of this study is the research of current solutions for the preservation of food using edible coatings formed from nanoemulsions. In order to achieve this general objective, the following specific objectives have been set:

- Study of the literature on nanotechnology appliqued to food industry as the use of nanoemulsions on food.
- Find out applications of active compounds encapsulated or integrated in the nanoemulsions with antimicrobial and antioxidant properties.
- Study of the components forming the coating, check that all of them meet the vegan requirements.

3. CHARACTERISTICS AND PROPERTIES OF NANOEMULSIONS

Emulsions are described as a dispersion of two immiscible liquids, in which one of them forms spherical droplets into the other. These droplets usually have a diameter between 100 nm and 100 μm when it is a food emulsion but there's an increasing interest in smaller diameters because of the physicochemical properties (McClements, 1999) and potential advantages over conventional emulsions they can provide (McClements, 2011). When the diameter of the droplet is between 10 and 200 nm (Rehman et al., 2021), it consists in a nanoemulsion. The substance that forms the droplets is referred to as the dispersed phase while the substance around the droplets is referred to as the continuous phase. Generally, these liquids are water phase and oil or lipid phase.

Due to the small diameter of particles, nanoemulsions are transparent or translucent and presents a high solubility and stability, so they offer excellent oral bioavailability and biological efficacy to dispersed bioactives (Abbas et al., 2013).

3.1. STRUCTURE

The most common liquids that form nanoemulsions are water and oil. When the oil is dispersed in the water forming droplets is an oil-in-water nanoemulsion and is known as O/W, it is the most common type. When is the water that forms droplets dispersed in the oil it is called water-in-oil nanoemulsion or W/O –All of these are simple nanoemulsions, but multiple emulsions can also be formed, such as water-in-oil-in-water (W/O/W) or oil-in-water-in-oil (O/W/O) emulsions (Fig. 1). The structure of a W/O/W emulsion is produced by initially forming a W/O nanoemulsion and then dispersing it in a water phase. Whereas in an O/W/O emulsion, O/W nanoemulsion is first formed and then dispersed in an oily phase (McClements and Jafari, 2018).

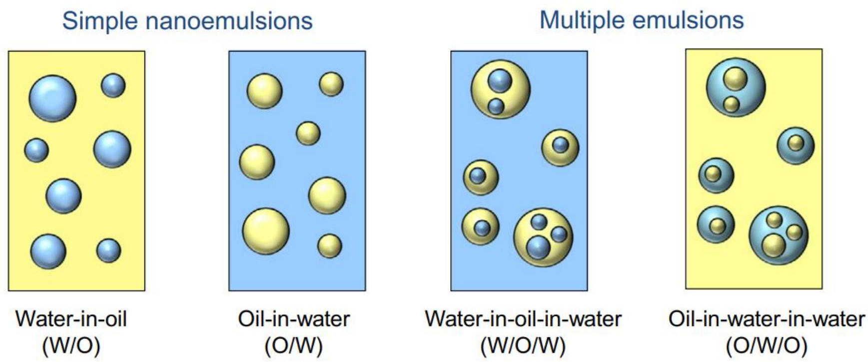

Fig. 1. Schematic illustration of nanoemulsions structures. *(Image extracted from McClements and Jafari, 2018).*

3.1.1. Droplet characteristics

The characteristics of the droplets determine the physicochemical characteristics of nanoemulsion-based products (Huang et al., 2010).

In O/W nanoemulsions, the droplets are composed of a centre made from hydrophobic material, which usually is oil, and it is surrounded by a shell formed by emulsifiers. In emulsions, the hydrophobic centre radius is much bigger than the shell thickness, so the properties mainly depend on the type of hydrophobic material. However, in nanoemulsions, both centre and shell have similar thickness, or have the same order of magnitude, so both contribute equitably on the nanoemulsion properties (Fig. 2). The composition of the droplet impacts in formation and properties of the nanoemulsion. Physicochemical properties, such as appearance and creaming stability depend on the density and the refractive index, respectively. These properties can be controlled by changing the density contrast and refractive index contrast, respectively, between the aqueous and oil phase (Zhang and McClements, 2018).

The droplet concentration is expressed as the volume of droplets per unit volume of nanoemulsion, which is known as volume fraction (ϕ). The droplet concentration can be varied changing the initial quantity of dispersed phase before the emulsion formation. It can be decreased by dilution with continuous phase or increased by concentration with methods such as filtration, centrifugation, or evaporation after the fabrication (Zhang and McClements, 2018).

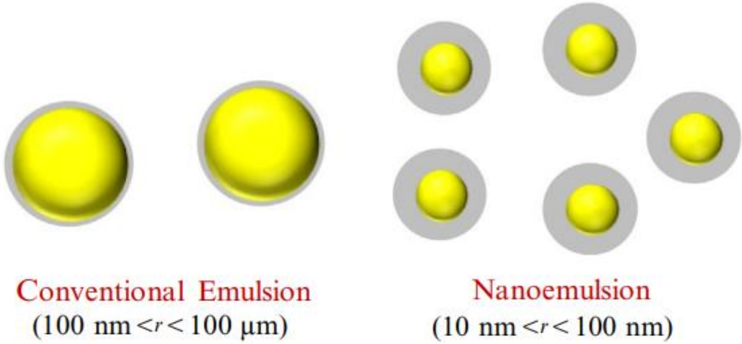

Fig. 2. Schematic illustration of droplet structure (core-shell) in emulsions and nanoemulsions. *(Image extracted from Zhang and McClements, 2018).*

The droplets are often electrically charged when they absorb charged materials. The zeta potential of the droplets can be positive, neutral, or negative depending on whether they absorb a cationic, non-ionic, or anionic emulsifier, respectively. The sign and magnitude of the surface is important in the determination of the properties and function of the nanoemulsion. The zeta potential can be varied by changing the pH or the ionic strength (Zhang and McClements, 2018).

3.2. APPEARANCE

Optical properties of nanoemulsions depend on the structure and composition of them, this can change the way they scatter and absorb the light. So, the transparency or opacity of a nanoemulsion depends on the light scattering profile and this, at the same time, depends on the size and concentration of the droplets and refractive index contrast of the continuous phase and the droplets. On the other side, the colour of a nanoemulsion depends on how the light wave is absorbed by chromophoric substances present in the droplets or in the continuous phase (McClements, 2002; McClements and Jafari, 2018).

When light enters to nanoemulsion and a wave encounters a droplet, part of this wave is transmitted, and part is scattered. The fraction that represents the part scattered and the direction it takes depends on the refractive index of both phases and on the relative size of the droplet respect the wavelength of light. If there is a low concentration of droplets, a dilute nanoemulsion, the light wave may encounter a reduced number of droplets in his way through de nanoemulsion. If the concentration of droplets is higher, the probability of a light wave to encounter more droplets increases, and the fraction that returns to the surface due to scattering increases too, showing a diffusely reflected light (McClements, 2002). When the size of the droplets is near the wavelength of light ($r \approx \lambda$), what usually happens in emulsions, the light is strongly scattered, and the appearance of the emulsion is opaque. As the droplet diameter is smaller respect the wavelength of light ($r \ll \lambda$), the appearance becomes clearer, and the nanoemulsion tend to be optically transparent or slightly turbid (Fig. 3) (McClements, 2011).

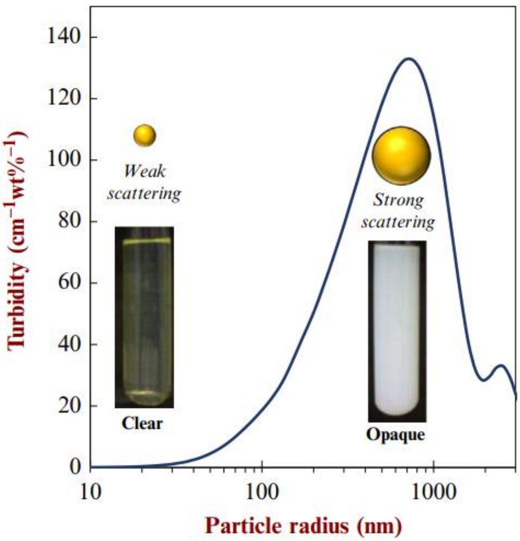

Fig. 3. The droplets much smaller than the wavelength of light, scatter it weakly and give clear nanoemulsions (left). The droplets similar in size to the wavelength of light, scatter it strongly and give opaque emulsions (right). *(Image extracted from Zhang and McClements, 2018).*

The other phenomenon that can occur when light enters to nanoemulsion is absorption. Both, continuous and dispersed, phases can absorb light waves. Some wavelengths are absorbed more strongly than others, so the emerging light is no longer white but of another colour, depending on the wavelength absorbed. Chromophoric substances are responsible for absorb the light waves. The colour of the emerged light depends on the concentration and absorptivity of the chromophores and the wavelength of the light that enters the nanoemulsion (McClements, 2002).

3.3. STABILITY

Emulsions are thermodynamically unstable. Phases will separate if the required time is given, because the separated oil and water phases have lower free energy than the emulsified ones (McClements, 2011). Furthermore, the formation of nanoemulsions has two steps. The first one consists in the formation of a coarse emulsion and the second one consists in the reduction of large droplets into the nanoemulsion droplet size. This step has an increase on Gibbs energy, so the process is not spontaneous, what implies that the reverted process, which is the destabilization into large droplets or into two separated phases, is spontaneous. The time for the process to reverse depends on his design, if it is well designed, it can reach kinetic stability and least for several years (Hakansson and Rayner, 2018). In most designs it is sought that it has at least kinetic stability for commercial applications (McClements, 2011).

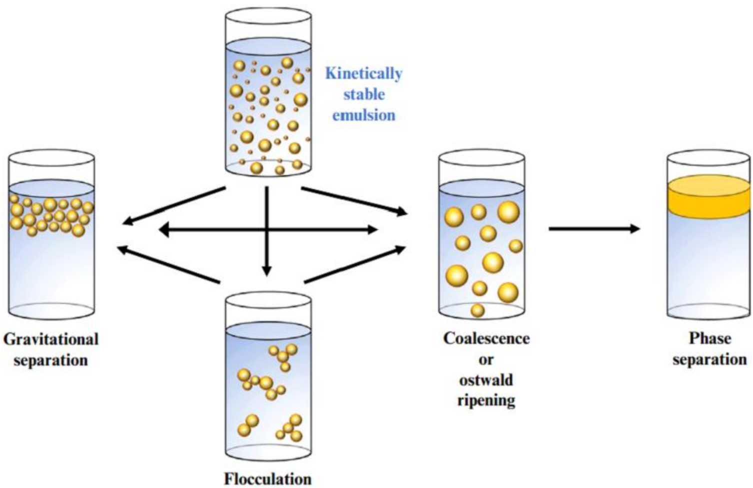

Fig. 4. Schematic illustration of instability mechanisms that occur in nanoemulsions: gravitational separation, flocculation, coalescence, Ostwald ripening, and phase separation. *(Image extracted from Zhang and McClements, 2018).*

Once the nanoemulsion is formed, it can be broken down by flocculation, coalescence, gravitational separation, Ostwald ripening, and phase separation mechanisms (Fig. 4). The rates at which these processes occur depend mostly on particle size. Some of these mechanisms do not work resoundingly for nanoemulsions, as they have very small droplets (McClements, 2011).

When very small droplets are formed, the surface area increases, so more emulsifier is needed to stabilize these small droplets. If the amount of emulsifier is the required, recoalescence is evited (Hakansson and Rayner, 2018).

3.3.1. Gravitational stability

Gravitational force act upon droplets in nanoemulsions since them normally have different density from the continuous phase. Creaming occurs when droplets density is much smaller than continuous phase one, so the droplets move upward. However, when droplets density is much bigger than continuous phase one, droplets move downward, which is called sedimentation (Fig. 5). Generally, oil have lower density than water, so creaming often happen in O/W nanoemulsions and sedimentation in W/O. Stokes' law (Eq. 1) describes the creaming velocity, v_{Stokes}, of an ideal particle (isolated rigid homogeneous spherical droplet) in an ideal liquid (Newtonian fluid):

$$v_{Stokes} = -\frac{2gr^2(\rho_2 - \rho_1)}{9\eta_1} \quad (1)$$

With g as the gravitational acceleration, r as the particle radius, ρ as the density, η as the shear viscosity, sub-index 1 referred as the continuous phase and sub-index 2 as the dispersed one. When creaming velocity is positive, particles go upward, while when it is negative, they go downward. Unfortunately, ideal particles and liquids are rarely found in nanoemulsions, and Brownian motion plays an important role, so Stokes' law does not work at all. Brownian motion favours a random distribution of the droplets throughout the system, while gravitational force favours the movement towards the top or bottom of the system. For small droplets, with diameters under 100 nm, Brownian motion dominates the movement, while for diameters above 100 nm, gravitational force dominates it (Zhang and McClements, 2018).

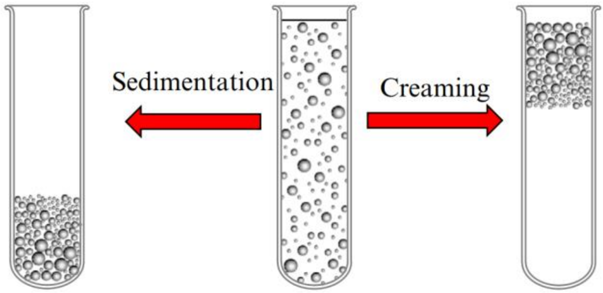

Fig. 5. Schematic illustration of sedimentation and creaming phenomena. *(Image extracted from Zhang and McClements, 2018).*

In the nanoemulsion droplets, the volume of the shell is appreciable in comparison to the entire droplet volume. The total density of a particle is the sum of the parts of density that each one, shell (s) and core (c), contributes in relation to the volume (V) they occupy (Eq. 2):

$$\rho_{particle} = \frac{V_c}{V_{particle}}\rho_c + \frac{V_s}{V_{particle}}\rho_s \quad (2)$$

The shell is normally denser than the core, so it contributes to gran part of the particle density, and it is decisive on the vertical movement of the particle. It plays an important role when it is possible to reduce the creaming velocity by reducing the density difference between dispersed and continuous phases (Zhang and McClements, 2018). It is seen in Lee and McClements (2010) that O/W nanoemulsions tend to sediment and O/W emulsions tend to cream due to the contrast between densities of droplets and water.

3.3.2. Droplet aggregation

In a nanoemulsion, the droplets are in motion, so they are more likely to collide. In these collisions, some droplets may stick together, depending on the forces acting on them and interactions, such as van der Waals, hydrophobic, electrostatic, and steric interactions. The first two interactions are attractive, while steric interaction is repulsive, and electrostatic interaction can be both attractive and repulsive depending on the zeta potential of the droplets and ionic composition of aqueous phase (McClements, 2011). Generally, as the droplet size increases, the strength of interactions is greater, and droplet aggregation is more likely to occur. When droplets stick together, they can preserve their individual integrity, what is called flocculation, or merge to form a larger droplet, what is called coalescence (Fig. 6) (Zhang and McClements, 2018).

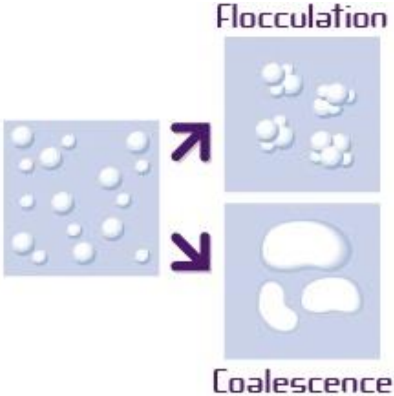

Fig. 6. Schematic illustration of flocculation and coalescence. (Hdihang,*13/1/22 via Wikimedia Commons, Creative Commons Attribution).*

3.3.3. Ostwald ripening

Ostwald ripening is the phenomenon by which a droplet grows at the cost of a smaller droplet due to the difference in chemical potential of the material within the droplets. The mean droplet size of the nanoemulsion increases at the same time as material of the small droplets goes to the larger ones (Taylor, 1998). In O/W nanoemulsions, this phenomenon deals with the diffusion of the oil from small to large droplets. The oil molecules move from the surface of small droplets to larger droplets and make them grow, while small droplets reduce in size (Fig. 7). As the droplet size decreases, the water solubility of oil molecules within the droplet increases. The water solubility of the oil phase is what most determines the stability of a nanoemulsion to Ostwald ripening. That is why nanoemulsions made from low water solubility oils are not usually prone to Ostwald ripening. On the other hand, nanoemulsions made from considerably water solubility oils tend to be very unstable to Ostwald ripening. To improve the stability to this phenomenon, the oil phase of nanoemulsions that are made from substantial water solubility oils is mixed with water soluble oil before the nanoemulsion formation (Zhang and McClements, 2018).

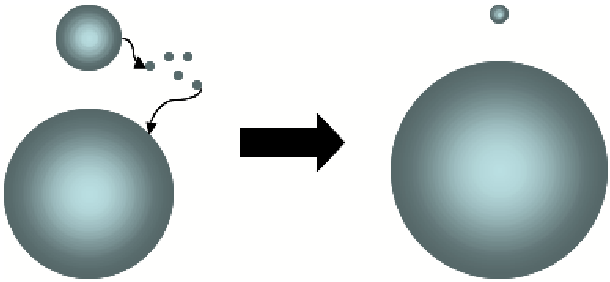

Fig. 7. Schematic illustration of the material of the small droplet going towards the large one and modifying their sizes. (Bfigura,*13/1/22 via Wikimedia Commons, Creative Commons Attribution).*

3.4. RHEOLOGY

Rheology is the scientific discipline that studies the deformation and flow of matter. Rheological responses are viewed at macroscopic level, nevertheless, they are originated at microscopic level (Rao, 2013). So, the measurement of rheological properties may provide important information about microscopic organization (Quemada and Berli, 2002). The rheology of a nanoemulsion may determine the shelf-life and sensory attributes of products based on nanoemulsions, the efficiency of droplet disruption in homogenization, and the design and operation of processes of nanoemulsions manufacturing. Rheological properties of nanoemulsions are ranged from low viscosity liquid to semisolid, they depend on the composition, structure, and interactions (Zhang and McClements, 2018).

In dilute systems, droplets are widely separated from each other, so they practically do not interact. As concentration of droplets increases, there are more interactions between particles. Linear viscosity increase as the concentration of droplets increases when continuous phase is considered as ideal liquid and dispersed phase is considered as spherical isolated particles (Zhang and McClements, 2018).

In concentrated systems, droplets interact hydrodynamically or colloidally so shear viscosity can be affected (Tadros, 2010). The fluid flow around the droplets may determine the shear viscosity. At low shear stresses, the droplets are distributed randomly due to Brownian motion, which leads to high shear viscosity nanoemulsion. At high shear stresses, the droplets are arranged according to fluid flow lines, the resistance is lowered, and it reduces the viscosity (Zhang and McClements, 2018).

Nanoemulsions often contain charged droplets, and they can alter viscosity if they interact with each other. The repulsive interactions keep the droplets from getting too close when they meet each other (Tadros, 2010). The repulsion between particles rarely happens when droplet radius is much bigger than shell thickness. But, when droplet radius is “comparable with shell thickness, there are repulsive interactions and overlap occurs, which makes the viscosity higher (Zhang and McClements, 2018). The attractive interactions can cause flocculation when they beat repulsive interactions (Tadros, 2010) Generally, flocculated nanoemulsions have higher viscosity than non-flocculated, this is due to the continuous phase retained between flocculated particles (Zhang and McClements, 2018).

4. FORMATION OF NANOEMULSIONS

As the process of formation of nanoemulsions is not spontaneous, the application of energy is required. Energy can be applied either to break-up droplets or to create new droplets. In top-bottom approaches large droplets are broken to make small ones, such as in emulsification and milling (Fig. 8) (Velikov and Pelan, 2008). Nevertheless, the Laplace pressure, which is the pressure difference between outside and inside of the droplet (Abbas et al., 2013), must be overcome to deform and then break the droplets, that is why a huge amount of energy is required, especially when droplets to break are already small because the Laplace pressure is bigger (Kanasaki et al., 2021). In bottom-up approaches, small droplets are made by the self-organization, and they are an assembly of molecules, monomers, ions, or atoms. These processes can be physical or chemical, such as precipitation and polymerisation (Fig. 8) (Velikov and Pelan, 2008). These approaches give a greater control over particle size and structure, and consume less energy (Rehman et al., 2021).

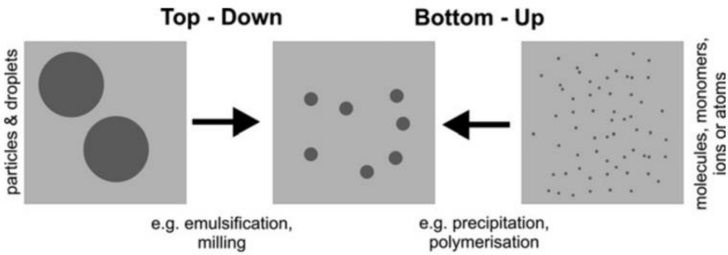

Fig. 8. Schematic illustration of top-down and bottom-up approaches. *(Image extracted from Velikov and Pelan, 2008).*

Nanoemulsion fabrication methods can be divided, according to the energy applied, into high-energy and low-energy methods. High-energy methods require the application of mechanical energy. Low-energy methods uses the energy released when the conditions of the system are changed (McClements and Jafari, 2018).

4.1. HIGH-ENERGY METHODS

High-energy methods are the most extensively used in industry for the fabrication of nanoemulsions. Devices used in these methods generate shear, turbulent, and cavitational flow profiles to blend the continuous and dispersed phases (McClements and Jafari, 2018). The most frequent used methods are high-pressure valve homogenization, microfluidization, and ultrasonication. Although there are more, such as rotor-stator and membrane emulsification.

The resulting droplet size of a prepared emulsion by homogenization is determined by the balance between droplet break-up and droplet re-coalescence. When the applied shear is higher than the Laplace pressure of the emulsion, droplet break-up occurs. Surfactants have an important role in both terms of the balance. They contribute to droplet break-up by making the interfacial tension lower, what decreases the resistance to droplet deformation (Leong et al., 2009; Walstra, 1993).

4.1.1. High-pressure valve homogenization

One of the most common methods used in industrial nanoemulsion formation is high-pressure valve homogenization (HPVH) (Schubert et al., 2003). In this method the large droplets of coarse emulsion are reduced by the application of pressure. It consists of a pump that forces the emulsion through a valve that forms a narrow gap, where the droplets are reduced (Hakansson and Rayner, 2018).

HPVH presents two advantages that make the difference over other methods; it can generate very high local stresses, what allows the creation of very small droplets, and its technology permits continuous production, what is important in large-scale processing, generating a production up to 50 000 L/h. The capacity to form nanoemulsions depends on the ability to give high fragmentation rate and low coalescence rate (Hakansson, 2018). Furthermore, it is considered the standard technology for emulsification for low to intermediate viscosity fluids (1 to 200 mPa s) (Jafari et al., 2008; Schultz et al., 2004). That's why a part of its extended use in dairy production, it is used in food, cosmetics, and pharmaceutical production (Hakansson, 2018).

The thermodynamic efficiency of HPVH is low, it requires high energy input, in consequence, the temperature of the product increases. This might have negative effects on the emulsion or on any molecules carried in the emulsion, such as nutrients or pharmaceuticals (Hakansson, 2018).

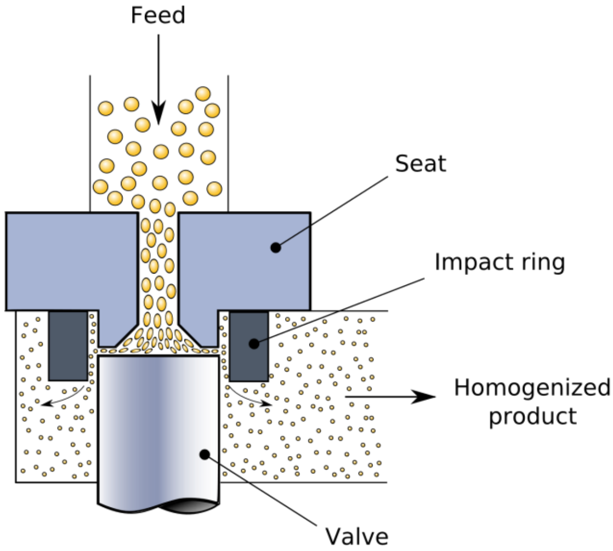

Fig. 9. Schematic illustration of high-pressure valve homogenization. (Daniele Pugliesi, *11/1/22 via Wikimedia Commons, Creative Commons Attribution).*

The mechanism of this method is the following: first, a coarse emulsion is created, the pre-emulsion, which feeds the homogenizer. Then, this is pumped into the HPVH, and it passes through the valve. Each valve or stage – being normal for a HPVH to have one or two stages– has an annular seat, a cylindrical forcer, and an annular impact ring. The pre-emulsion passes through the gap formed between the seat and the forcer, and enters the outlet chamber, the space formed by the seat, the forcer, and the impact ring. In this action is when the breakup of the droplets occurs because of the pressure, giving rise to a homogenous emulsion (Fig. 9) (Hakansson, 2018). One or more stages can be added to the design (Fig. 10), and some studies have shown that two-stage improves the thermodynamic efficiencies compared to one-stage (Mohr, 1987).

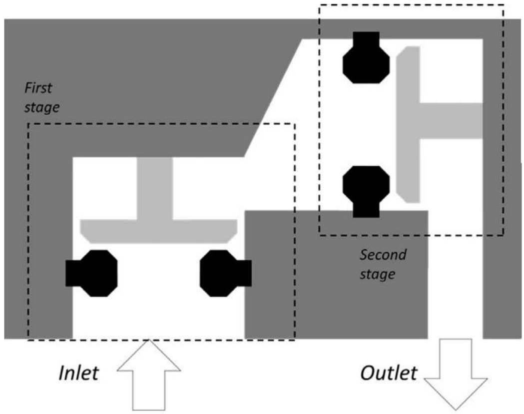

Fig. 10. Schematic illustration of high-pressure homogenizer with two valves mounted in series. (*Image extracted from Hakansson, 2018).*

4.1.2. Microfluidization

Microfluidization is a very usual method. The way this method works is quite similar to high-pressure valve homogenization. It consists of a pump that forces a coarse emulsion into two channels, at the exit of this, there is an impact zone, where the channels meet, and the droplets collide. The crash makes the droplets burst into small sized droplets (Fig. 11) (Hakansson and Rayner, 2018).

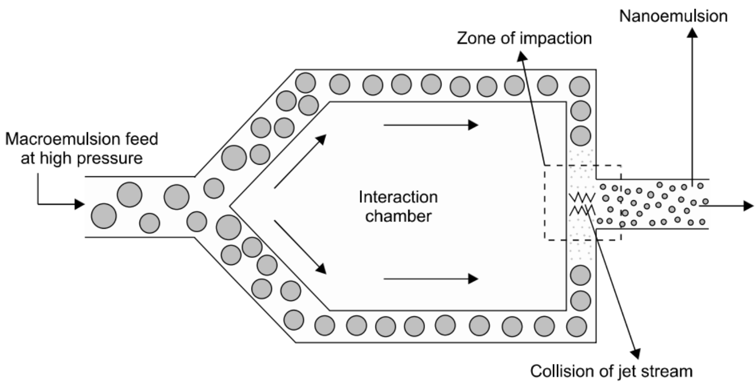

Fig. 11. Schematic illustration of microfluidization method. (*Image extracted from Kumar et al., Prev. Nutr. Food Sci., **2019**, 24(3), 225-234).*

Microfluidization presents a main advantage; it can control the emulsion droplet size, which makes this method very useful in industry operations (Villalobos-Castillejos et al., 2018). There is relationship between the droplet size and the pressure or the number of passes, among others. It is that to reduce the droplet size, it is necessary an increase of the pressure, an increase of the number of passes, an increase of the concentration of emulsifier or a decrease of the ratio of viscosities of dispersed and continuous phases. Furthermore, this method permits the production of both O/W and W/O nanoemulsions (Villalobos-Castillejos et al., 2018), as well as nanoemulsions together with various materials such as bioactive lipids, nutraceuticals, antioxidants, flavours, colours, and vitamins (McClements et al., 2007; Weiss et al., 2009).

A pneumatic pump is responsible for providing the coarse emulsion to the interaction chamber at very high pressures of up to 30 000 psi (200 MPa). Inside the interaction chamber the sample is pushed through two channels that have been engineered to be between 75 to 100 µm in nominal dimension. Here the pre-emulsion is accelerated, it can reach velocities of up to 500 meters per second, and then collide the two channels, generating shear, impact, and energy dissipation forces, which create fine droplets (Microfluidics, 2014).

4.1.3. Ultrasonication

In contradistinction to the just explained methods, ultrasonication does not accelerate the coarse emulsion to break it up. It consists of ultrasonic sound waves that are capable of break the droplets. The energy necessary for the breakup comes from inertial cavitation (Hakansson and Rayner, 2018).

This method is appropriate to food, pharmaceutical, and petroleum industries applications due to its capacity to produce very fine droplet nanoemulsions with narrow particle size distributions (Gharibzahedi and Jafari, 2018; Canselier et al., 2002; Sivakumar et al., 2014). The principal reason for the reduction of droplet size is the cavitation phenomenon (Canselier et al., 2002). An increase on the sonication time improves the efficiency of emulsification process, and decreases the droplet size (Landfester et al., 2004).

The audible sound range is extended between 16 Hz and 16 kHz. Frequencies above these belong to the ultrasonic range. Only powerful ultrasound 16 kHz to 1 MHz is capable to interact with matter such as producing cavitation phenomenon. Frequency range used in ultrasonication for the formation of nanoemulsions is between 20 to 100 kHz, which is the most powerful in ultrasound range (Fig. 12) (Canselier et al., 2002).

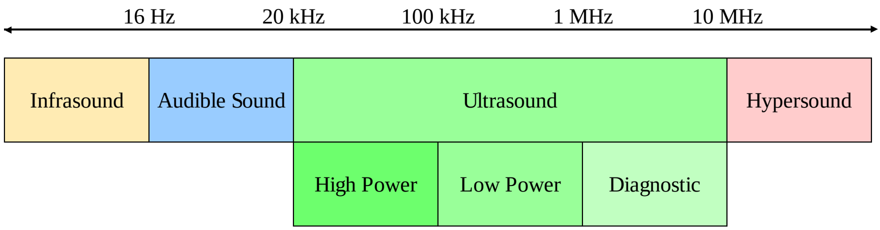

Fig. 12. Sound frequency range diagram *(Based on Canselier et al.,2002).*

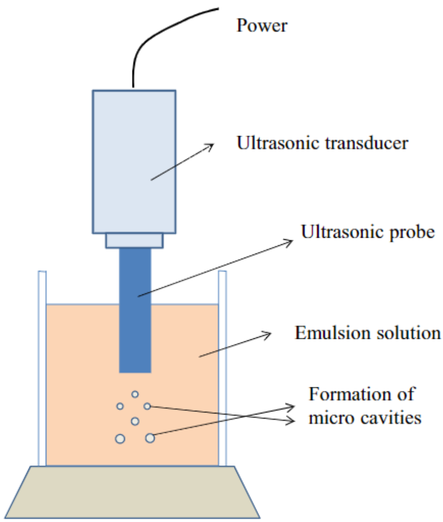

Fig. 13. Schematic illustration of ultrasonication set up. (*Image extracted from Carpenter and Saharan, Ultrasonics Sonochemistry, **2017**, 35, 422-430).*

Sound waves are generated using piezotransmitter. Piezotransmitters convert electric voltage into mechanical vibrations (Hakansson and Rayner, 2018). Vibrations are amplified by a metal horn and propagated in the form of acoustic waves through a probe attached to the horn. When the probe is submerged into the coarse emulsion, the waves induce acoustic pressure.

The gas microbubbles dissolved into the continuous phase grow because of rectified diffusion. As they grow in size, they suffer contractions and expansions, and finally implode generating high shear forces that breaks the emulsion droplets (Fig. 13) (Abbas et al., 2013; Gharibzahedi and Jafari, 2018).

4.1.4. Rotor-stator emulsification

The formation of a pre-emulsion is needed in some methods. It is made, most of the time, by rotor-stator emulsification, which consists of a perforated stator screen around of a rotor that spins (Fig. 14) (Hakansson and Rayner, 2018).

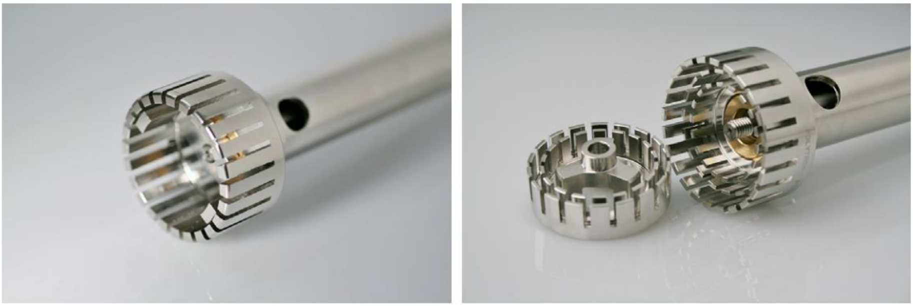

Fig. 14. Rotor-stator mixers. Rotor-stator mixer mounted (left) and rotor separated from the stator (right). (*Image extracted from van der Schaaf and Karbstein, 2018).*

This method is usually utilised to prepare coarse emulsions. Nevertheless, rotor-stator devices can reach the nano range but droplet sizes below 1 μm are very difficult to reach with one-step production (Jafari et al., 2007). That is why it may be combined to other methods to achieve the nanoemulsion droplet size easily (van der Schaaf and Karbstein, 2018).

One part of the device moves while the other part is motionless. The coarse emulsion enters the device axially, then, it is accelerated tangentially, and finally, it is impelled radially by the rotor through the slots of the stator screen, which generate intense hydrodynamic stresses (Fig. 15) (Hakansson and Rayner, 2018). To break the coarse emulsion droplets are necessary high stresses that may deform long enough the droplets. When stresses pass a critical value, droplets are broken, and emulsion droplet size is minimized (Gharibzahedi and Jafari, 2018).

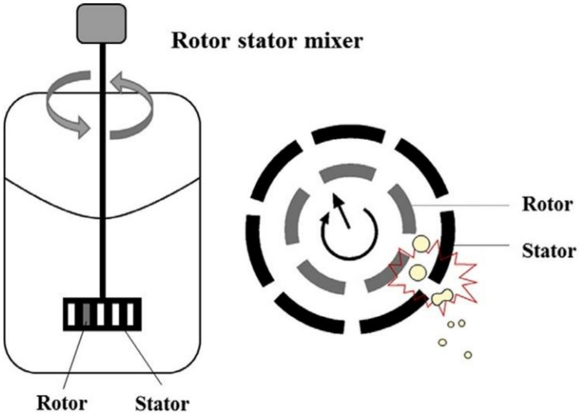

Fig. 15. Schematic illustration of a rotor-stator mixer *(Image extracted from Hakansson and Rayner, 2018).*

4.1.5. Membrane emulsification

Membrane emulsification techniques are in the line that separates high-energy from low-energy methods. They require more energy than phase inversion techniques but less than other high-energy methods. They are finally categorized as high-energy methods due to their similitudes in the mechanism of emulsification with those methods (Hakansson and Rayner, 2018).

The two main advantages of this method are the little energy it requires compared to the other high-energy methods, and the relatively uniform drop distributions it can obtain (Hakansson and Rayner, 2018).

In direct membrane emulsification, there are two liquids separated at the beginning by a porous membrane. The liquid of the dispersed phase is pressed across and passes through the membrane into the other liquid, the continuous phase. While the liquid passes through the orifices, the liquid in the transversal flow applies a laminar shear force, creating fine droplets of dispersed phase (Fig. 16) (Hakansson and Rayner, 2018). In premix membrane emulsification, a coarse emulsion is already prepared, and it is forced to pass through the membrane. This makes the droplet size to reduce (Fig. 16) (Vladisavljevic, 2018).

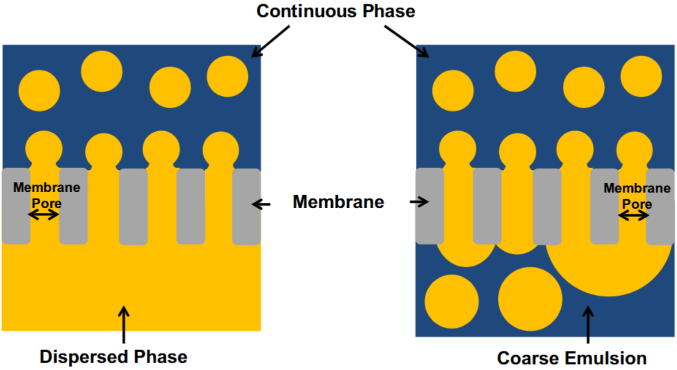

Fig. 16. Schematic illustration of membrane emulsification methods. Direct membrane emulsification (left) and premix membrane emulsification (right) *(Image extracted from Piacentini et al., Journal of Membrane Science, **2014**, 468, 410-422).*

4.2. LOW-ENERGY METHODS

Low-energy methods are preferred over high-energy methods when spending little energy and using a method of a gentle nature with non-destructive processes are wanted. However, these methods used for the formation of nanoemulsions require high concentration of surfactants, so they are shorter used than high-energy ones (Sugumar et al., 2016).

Methods such as emulsion inversion point, spontaneous emulsification, phase inversion temperature (PIT) or phase inversion composition (PIC) use energy generated by alterations in their composition or environment conditions. Phase inversion temperature or composition methods rely on transitional phase inversion (TPI) while emulsion inversion point relies on catastrophic phase inversion (CPI) (McClements, 2011). Generally, TPI methods create smaller droplets than CPI ones, in spite of the fact that CPI methods are normally simpler to perform (Perazzo and Preziosi, 2018).

4.2.1. Phase inversion temperature

In phase inversion temperature (PIT) method, one type of emulsion is induced to transform to another, from O/W to W/O or from W/O to O/W, by changing the temperature, which makes the physicochemical properties of the surfactants change and redistribute. This method is based on the change of optimum curvature and solubility (McClements, 2011).

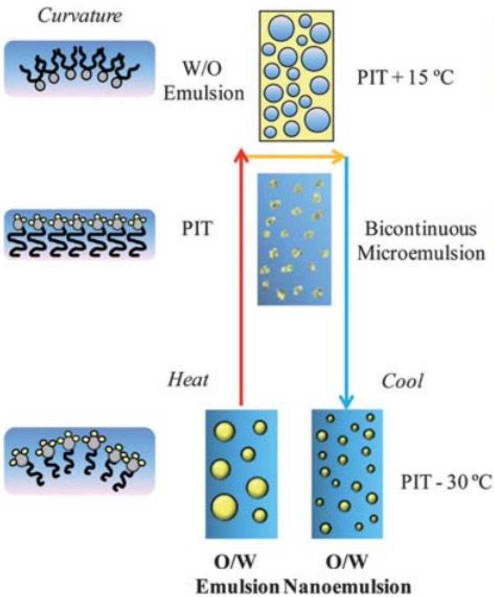

Fig. 17. Schematic illustration of phase inversion temperature method. (*Image extracted from McClements, 2011).*

Surfactant molecules are associated forming monolayers that have a curvature. Its geometry depends on $\upsilon/a_0 l$, known as the "packing parameter", p, which depends on volume, υ, and length, l, of hydrophobic tail and cross-sectional area, a_0, of hydrophilic head-group of the surfactant (Israelachvili, 2011).

The curvature of a monolayer is optimum when the molecules can be packed efficiently, and the free energy of the monolayer is the minimum. For $p < 1$, the optimum surfactant curvature is convex and tends to form O/W emulsions, for $p > 1$ the optimum curvature is concave and tends to form W/O emulsions, and for $p = 1$ the optimum curvature is no-curvature and forms bicontinuous or liquid crystalline systems (McClements, 2011).

Non-ionic surfactants change their solubility with temperature changes. When the temperature is lower than the phase inversion temperature (PIT), the hydrophilic head-group of the surfactant is hydrated and becomes more soluble in water. When temperature increase and surpass the PIT, the head-group is gradually dehydrated and solubility in water decreases. At the PIT, the surfactant has almost equal solubility in oil and water phases (McClements, 2011).

To get small droplets, such as a nanoemulsion, an emulsion at a temperature moderately above the PIT may be quickly cooled to a temperature bellow the PIT (Fig. 17) (McClements, 2011). The resulting nanoemulsions are kinetically stable for months (Anton and Vandamme, 2009).

4.2.2. Phase inversion composition

The phase inversion composition (PIC) method is analogue to PIT but with composition altered to change the emulsion type (Fig. 18). To invert an O/W emulsion that is stabilized with an ionic surfactant by PIC method, it is necessary to add salt, which screen the electrical charge of the surfactant head-groups and, consequently, packing parameter changes from $p < 1$ to $p > 1$, this generates a W/O emulsion. On the other side, to invert a W/O emulsion that have a high salt concentration, it is necessary to dilute it adding water, which reduce the ionic strength below a critical level, the packing parameter changes from $p > 1$ to $p < 1$, and a O/W is formed (McClements, 2011; Sugumar et al., 2016).

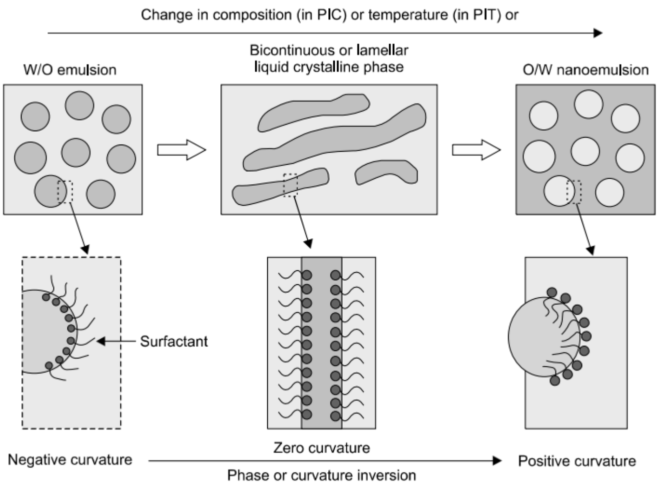

Fig. 18. Schematic illustration of phase inversion composition and temperature methods. (*Image extracted from Kumar et al., Prev. Nutr. Food Sci., **2019**, 24(3), 225-234).*

4.2.3. Spontaneous emulsification

In this method nanoemulsion is formed by mixing two liquids, usually an organic and an aqueous liquid. It works because of the movement of solvents or surfactants that are miscible with the aqueous phase from the organic phase to the aqueous phase. The way it is implemented can change due to the composition of the organic and aqueous phases, the environmental conditions, and the mixing conditions (McClements, 2011).

It starts with the mix of the two phases, which are thermodynamically stable alone. However, when the liquids are mixed, they are brought to a non-equilibrium state (Anton and Vandamme, 2009). Then, the component that is water-miscible and, initially, is in the organic phase, moves from one phase to the other. This movement creates a large turbulent force at the oil-water interface and its velocity creates a large increase in the oil-water interfacial area, which induces a spontaneous formation of droplets (Fig. 19) (McClements, 2011). To get nano-scaled droplets, the quantity of water-miscible component needs to be much bigger than oil in the organic phase before mixing, so then, the water-miscible component diffusion is faster and the turbulence generated form, in that way, very small droplets (Anton and Vandamme, 2009).

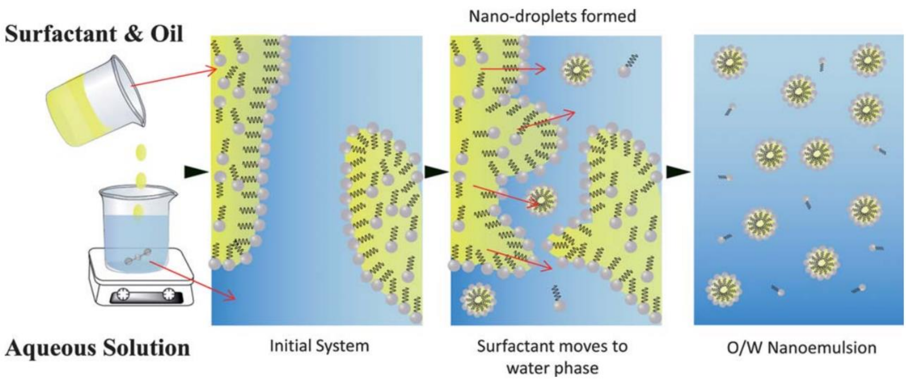

Fig. 19. Schematic illustration of spontaneous emulsification method. (*Image extracted from McClements, 2011).*

4.2.4. Emulsion inversion point

In this method, the emulsion changes from one type to another through a catastrophic phase inversion (CPI). This type of inversion is caused by the increase in the volume of the dispersed phase (Sajjadi et al., 2004). The emulsion is inverted due to the increasing amount of dispersed phase, which becomes the continuous phase after the inversion (Sajjadi et al., 2003). Generally, the emulsion is inverted from W/O to O/W by the addition of big amounts of water.

Abnormal emulsions have as the dispersed phase the phase with the surfactant dissolved. They are extremely unstable, nevertheless they can be created under continuous agitation by adding the dispersed phase to the continuous phase. By adding huge quantities of dispersed phase, the abnormal emulsion is destabilized, giving rise to a normal emulsion, this is called the catastrophic phase inversion (Sajjadi et al., 2003).

Water is added to a W/O emulsion with high oil-to-water ratio. It is continuously stirring. At a critical point, water is so concentrated that is packed tightly together and causes the phase inversion to occur. The emulsion changes from W/O to O/W (Fig. 20). The droplet size depends on the stirring speed and the rate of water addition (McClements, 2011). Mixtures of surfactants, rather than just one type, can be used to produce nanoemulsions (Perazzo and Preziosi, 2018).

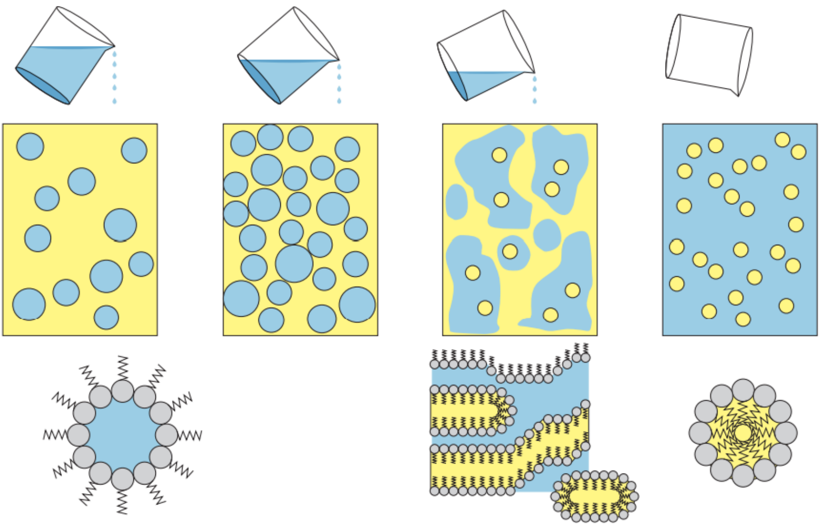

Fig. 20. Schematic illustration of emulsion inversion point method. (*Image extracted from Cardoso-Ugarte and Jiménez-Munguía, 2015).*

5. APPLICATIONS

Although nanoemulsions have applications in wider technological areas, the principal applications of nanoemulsions are in food, beverage, and pharmaceutical industries. Here, the relationship between the nanoemulsion microstructure and their physicochemical properties become important (McClements, 2011). This study is focused on food industry applications.

The nanoemulsion particle size is measured with dynamic or static light scattering analysis, the morphology of droplets can be studied through electron microscopic technique. Physical stability of the nanoemulsion system can be studied with environmental stress conditions, such as pH, temperature, and salt. Chemical stability can be studied using either high-performance liquid chromatography or spectroscopic analysis upon accelerated aging conditions (Sugumar et al., 2016).

One of the most important applications of nanoemulsions is their utilization in transparent or cloudy products, almost without changing the appearance of the product. This is possible when the droplet size is much smaller than wavelength of light, because light is not highly scattered, and the appearance is clearly. To obtain an optical transparent nanoemulsion, the majority of droplets of the system should have a size below a critical value, which is, according to Wooster et al. (2008), a diameter below 80 nm. To have the major quantity of the droplets below that size, the mean size of the nanoemulsion must be very small and the distribution of droplet size must be narrow (McClements, 2011). It is also important that the particle size is maintained and that the nanoemulsion is not destabilized, in this way it is avoided that the nanoemulsion becomes cloudy. By optimizing the composition and the process conditions, transparent nanoemulsions can be obtained by high and low energy methods.

Other application found is about their ability to enhance the bioavailability of lipophilic components encapsulated within the droplets. The smaller the droplet, the higher bioavailability. This is, in part, due to the biggest surface area that small droplets have, which permits a faster digestion, so the content is released and absorbed quickly. In other part, it can also be due to its small size, which allows it to penetrate layers of mucosa. Finally, it may also be due to the fact that the water solubility of the lipophilic components increases as the droplet size decreases, which could improve absorption (McClements, 2011). In this way, the improvement in the availability of curcumin when it is incorporated in nanoemulsions could be explained (Huang et al., 2010; Wang et al., 2008).

Sometimes, applications with nanoemulsions overtake those of emulsions since they may have better gravitational stability although they are more susceptible to Ostwald ripening due to their reduced particle size. It may also be that nanoemulsions find other applications due to a different texture, given by the rheology of the nanoemulsion, which depends on its microstructure.

Nanoemulsions have the ability to encapsulate and release components, if functional components such as vitamins, flavours, antimicrobials, and antioxidants are incorporated, this ability becomes a great application. Essential oils (EO), which can be encapsulated into nanoemulsions, are natural concentrated aromatic and volatile liquids obtained from the extraction by distillation of different parts of a plant, such as seeds, flowers, leaves, roots, or whole plant (Sugumar et al., 2016). EOs are utilized in medicine, cosmetic, and food applications due to their smell and antimicrobial properties (Tajkarimi et al., 2010).

There is an increasing interest in the use of essential oils as natural antimicrobials and preservatives incorporated on nanoemulsions because it is an alternative to synthetic food additives (Seow et al., 2014). Nanoemulsions containing essential oils present positive effects against specific types of bacteria, some studies are shown in Table 1.

Table 1. Antibacterial and antifungal essential oil studies.

Essential oil	Emulsifier	Type of bacteria or yeast	Reference
Peppermint oil	Modified starch	Gram positive bacteria (*Listeria monocytogenes and Staphylococcus aureus*)	Liang et al., 2012
Thyme oil	Tween 80, T80	Yeasts (*Zygosaccharomyces bailli, Saccharomyces cerevisiae, Brettanomyces bruxellensis, and Brettanomyces naardenensis*)	Ziani et al., 2011
Orange oil	Tween 80	Yeast (*Saccharomyces cerevisiae*)	Sugumar et al., 2015
Cinnamon oil	Tween 80	Gram positive bacteria (*Bacillus cereus*)	Ghost et al., 2013b
Sesame oil	Tween 20, Tween 80	Gram positive bacteria (*Staphylococcus aureus*)	Ghost et al., 2013a

EOs do not have a specific mechanism to perform antimicrobial actions, but rather have different ways of interacting with cells (Fig. 21), based on molecular hydrophobicity (Salvia-Trujillo et al., 2015). The phenolic components of EOs are the main ones in charge of interacting with the cell membrane, since they interact with the lipids that form it, making it more permeable, breaking homeostasis, and causing the leakage of ions and cytoplasmic content (Seow et al., 2014).

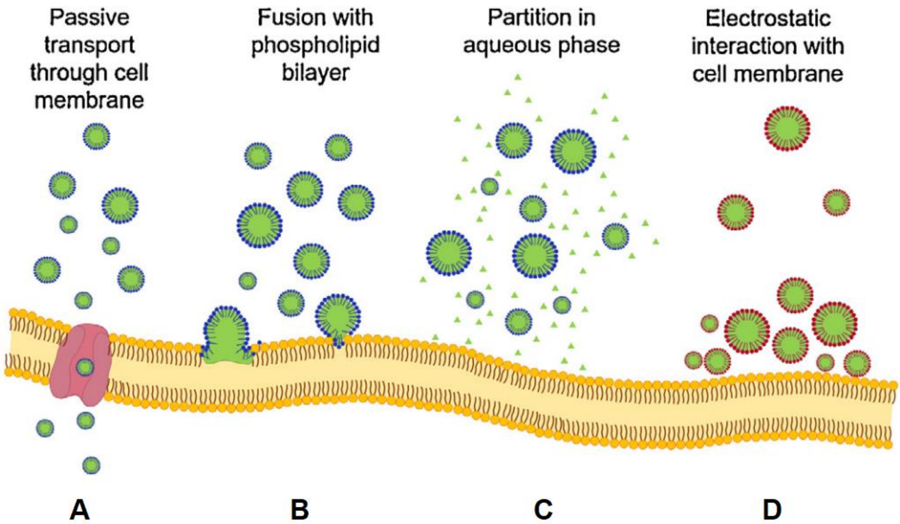

Fig. 21. Schematic illustration of the mechanisms of interaction of nanoemulsions with essential oils with microbial cell membranes. (*Image extracted from Donsì and Ferrari, 2016).*

Encapsulation of EOs improves their physicochemical stability and their solubility with food matrices, resulting in better interaction with microbial cells (Donsì and Ferrari, 2016). The small nanoemulsion droplets are capable to pass through porin proteins of bacteria cell membrane (Fig. 21A) (Nazzaro et al., 2013). Emulsifier droplets may fusion with the phospholipid bilayer of cell membranes, this allows the release of EOs at desired sites (Fig. 21B) (Donsì and Ferrari, 2016). Li et al. (2015) demonstrate the antimicrobial activity against *E. coli* with Tween 80 or SDS as emulsifiers of eugenol, and then, Majeed et al. (2016) also demonstrate the antimicrobial activity against *E. coli*, *S. aureus*, and *L. monocytogenes* with Tween 80 or modified starch as emulsifiers of thyme oil. The nanoemulsion droplets act as a storage for essential oils, so they can be released prolongedly in time, due to dynamic equilibrium between the dispersed and continuous phases of EOs molecules (Fig. 21C) (Donsì and Ferrari, 2016). When droplets are charged positively, they are attracted to the negatively charged cell membranes, so EOs are nearer to the desired sites (Fig. 21D) (Chang et al., 2015).

5.1. EDIBLE COATINGS

Nanoemulsion based edible coatings have been successfully introduced in food industry due to their benefits in preservation and enhancement of the shelf-life of food. The preservation of food using films or coatings has been done for centuries, for example the use of wax to cover fruits or soymilk films to wrap vegetables (Gennadios and Weller, 1991). When a food is covered to preserve it, it is a great advantage that what covers the food is edible. That is why all components formulated in edible nanoemulsion coatings should be generally recognized as safe (GRAS) (Flores-López et al., 2015).

There are a few differences between films and coatings: films are a type of packaging that is prepared apart and then it is attached to the food, while coatings are solutions applied to the surface of food and that remain attached to the food (Montero et al., 2016). There are different methods to cover foods, which depend on the surface of the food and the purpose of the coating, these are dipping, brushing, spraying, and panning. The easiest way to apply the coating is directly from solution. The affinity of the coating for the product is indispensable for the adhesion of both parts to occur and be durable (Flores-López et al., 2015).

The edible coatings are fine layers that covers the food and present various advantages. These coatings can act as carriers for food additives such as antioxidants and antimicrobials. They can create a protective barrier between food and air, creating an artificial environment that reduces respiration rate and moisture loss (Chaudhary et al., 2020).

Although the nanoemulsion based edible coatings have a large number of benefits, they also have certain disadvantages. Some coatings can generate unpleasant odours once applied to the product due to deterioration. The coatings can have a non-transparent appearance which makes the product less attractive to the consumer. Some coatings may have a final sticky texture, which would also make it unpleasant for de consumer. When the coating is drying, it is difficult for the layer to remain homogeneous, so in some locations of the product there will be less coating than in others, in these parts it may not be enough coating to develop its function. Sometimes, a coating requires large amounts of bioactive agents to release them effectively.

A component often added in edible nanoemulsion coatings is chitosan. It is a biopolymer with antimicrobial and antifungal properties. It is obtained from chitin, a substance found in the exoskeleton of crustaceans, crabs, and shrimps, and it can also be obtained from the cell walls of fungi (Chaudhary et al., 2020). Chitosan combined with other substances, such as essential oils, resins, polysaccharides, and proteins, improve its functional properties (Horison et al., 2019).

Essential oils have shown antibacterial activity when they have been used into nanoemulsions, that is why they have also been used in nanoemulsion coatings for foods, presenting, in the same way, inhibition of bacterial growth.

Mohammadi et al. (2015) proved the effectivity of different nanoemulsion coatings to improve the shelf-life of cucumber during storage. Two types of coating were compared with the uncoated control vegetable during 21 days at cold storage temperature of 10±1 °C. The coating with chitosan nanoparticles (CSN) and the one with *Cinnamomum zeylanicum* essential oil (CEO)-loaded CSN were very effective for inhibiting total aerobic bacteria, yeasts, and mould growth, although the CEO-CSN was more effective than pure CSNs coating. It can be easily observed in Fig. 22 that the control cucumbers were dehydrated and very contaminated by mould at the 15th day, whereas coated vegetables maintained a good appearance almost until the 21st day, although the CEO-CSN coated cucumbers present a better green coloured appearance and are more hydrated than the CSN ones. The coatings contained CEO which is an essential oil that can be applied as an antioxidant, bactericidal, antifungal, and enhancer of flavours and aromas. It is obtained from cinnamon, which is a popular spice with a characteristic smell (Cardoso-Ugarte et al., 2016).

Horison et al. (2019) proved the effect of chitosan combined with nutmeg seed oil, which have antimicrobial properties, as components of edible nanoemulsion coating. They made the nanoemulsions using the rotor-stator method and the high-pressure valve homogenization method and observed the growth of microbes and mould-yeast in strawberry fruit stored at 10 °C for 5 days. The results showed that coated fruit presented less microbial and fungal activities than uncoated fruit. The droplets made with HPVH method were smaller, so their surface area was larger which increased the effectiveness in inhibiting microbial and mould-yeast growth on strawberries compared to rotor-stator method nanoemulsion.

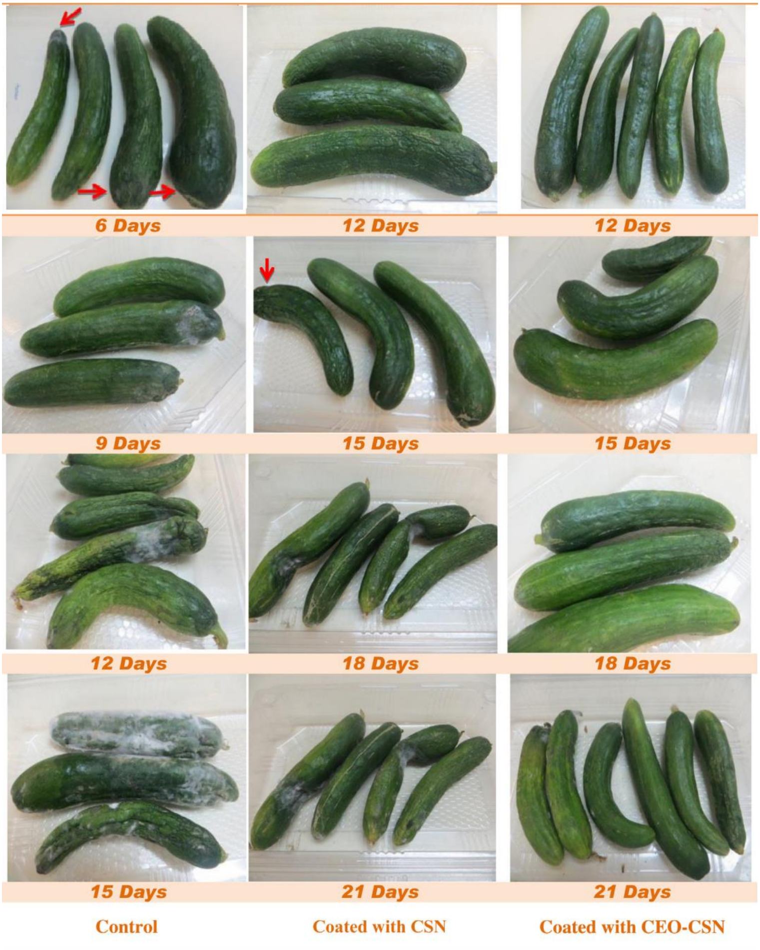

Fig. 22. Appearance of control and coated cucumbers during the experiment. (*Image extracted from Mohammadi et al., 2015).*

Kim et al. (2014) proved the effect of lemongrass oil incorporated in nanoemulsions against *Salmonella typhimurium* and *Escherichia coli* in grape berries. They compared the stability of different nanoemulsion formation methods: vortex mixing, high-shear probe mixing, and dynamic high pressure. The high pressure one created the most stable nanoemulsion and presented the smaller droplets (Fig. 23), so it was the method used during the experimentation. The coating made the surface of the fruit shinier and did not greatly affect the flavour. After 28 days stored at 4 and 25 °C, the coating showed antimicrobial properties against *S. typhimurium* and *E. coli*. The coating was also effective to reducing weight loss, firmness, and presented antioxidant activity.

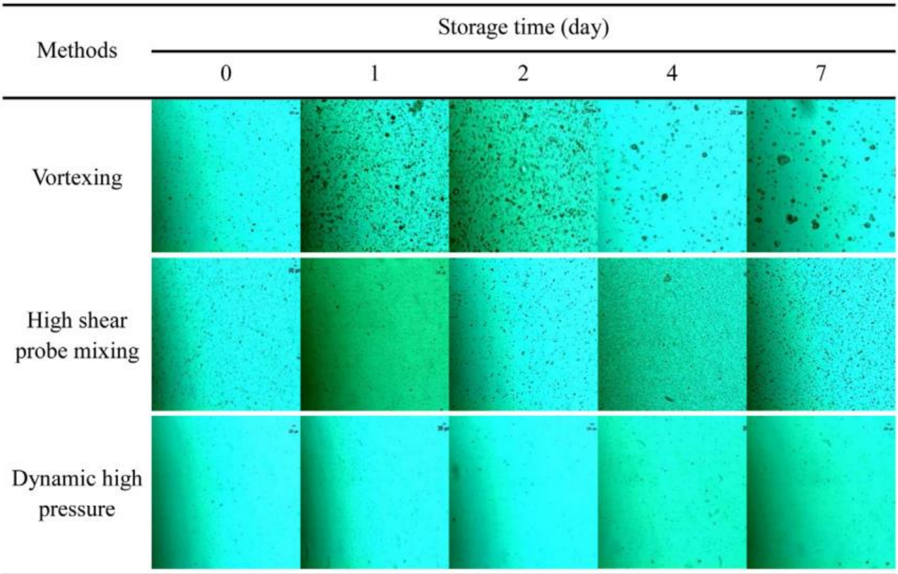

Fig. 23. Micrographs of nanoemulsions containing lemongrass oil produced by different methods at 25 °C for 7 days. (*Image extracted from Kim et al., 2014).*

More applications of edible nanoemulsion coatings are shown in Table 2. In all these applications the coatings increased the shelf-life of food products. The efficacy of edible coatings for the inhibition of bacteria is reaffirmed. The coatings did not significantly affect the appearance and firmness of food. Some of these studies provided scientific basis for the use of edible coatings as an alternative preservation treatment for fresh-cut fruits. The components forming the studied coatings meet the vegan requirements.

Table 2. Applications of edible coatings on fruits.

Main ingredients	Food products	Properties of coating	References
Alginate, chitosan and *Flourensia cernua*	Tomato	Reduced the weight loss, microbial growth, gas exchange and ethylene production. Maintained firmness and colour.	Salas-Méndez et al., 2019
Alginate and chitosan	Fresh-cut melon	Good adhesion to melon matrix. Reduced the bacteria, yeast, and fungi growth. Maintained the attractive appearance.	Poverenov et al., 2014
Sodium chlorite and chitosan	Fresh-cut d'Anjou pears	Maintained firmness and did not affect weight loss, prevented the browning reaction. Effective inhibition of *E. coli*.	Xiao et al., 2011
Apple pectin, potassium sorbate, sodium benzoate, and nisin	Fresh-cut 'Rojo Brillante' persimmon	Inhibited browning and growth of bacteria, maintained acceptable flavour. No growth of moulds and yeasts.	Sanchís et al., 2016
Gelatine, chitosan, and guar gum	Barhi date	Delayed the ripening and extended shelf-life of the fruit. No affected significantly the organoleptic characteristics.	Abu-Shama et al., 2020
Guar gum, aloe vera, and *Spirulina platensis*	Mango	Reduced respiration rate and weight. Maintained firmness and the ascorbic acid content of the fruit.	Ebrahimi and Rastegar, 2020
Chitosan and chlorogenic acid	Peach	Increased the antioxidant activity. Reduced weight loss and respiration rate. Maintained firmness, soluble solids, titratable acidity, and ascorbic acid contents of the fruit.	Jiao et al., 2019

6. CONCLUSIONS

Most of the properties of nanoemulsions are determined by the characteristics of the droplet and its size. Nanoemulsions present some advantages over emulsions because of their smaller droplet size: they do not scatter light strongly so they can be from transparent to opaque depending on the droplet radius, and they are highly stable to gravitational separation and droplet aggregation, although they are not so stable to Ostwald ripening.

High-energy methods are the most utilized in industry to produce nanoemulsions and the high-pressure valve homogenization method is currently the most widely used to produce nanoemulsions.

Nanoemulsions have a large number of applications, of which the most prominent are in food, pharmaceutical, and beverage industries, due to their bioavailability and transparency. Edible nanoemulsion coatings are capable of improving preservation and shelf-life of food and reducing the growth of bacteria and fungi. Essential oils incorporated in the coatings present antibacterial and antifungal activities. One of the most used ingredients is chitosan, due to the good results it shows in terms of the growth of bacteria and fungi. Most of the coatings applied to food products do not significantly affect the appearance and the firmness of food.

REFERENCES AND NOTES

1. Abbas, S.; Hayat, K.; Karangwa, E.; Bashari, M.; Zhang, X. An Overview of Ultrasound-Assisted Food-Grade Nanoemulsions. Food Engineering Reviews. **2013**. 5(3), 139-157.
2. Abu-Shama, H.S.; Abou-Zaid, F.O.F.; El-Sayed, E.Z. Effect of using edible coatings on fruit quality of Barhi date cultivar. Scientia Horticulturae. **2020**. 265. 109262.
3. Acevedo-Fani, A.; Soliva-Fortuny, R.; Martín-Belloso, O. Nanoemulsions as Edible Coatings. Current Opinion in Food Science. **2017**. 15, 43-49.
4. Anton, N.; Vandamme, T.F. The universality of low-energy nano-emulsification. International Journal of Pharmaceutics. **2009**. 377, 142-147.
5. Canselier, J.P.; Delmas, H.; Wilhelm, A.M., Abismail, B. Ultrasound emulsification – an overview. J. Dispers. Sci. Technol. **2002**. 23, 333-349.
6. Cardoso-Ugarte, G.A.; Jiménez-Munguía, M.T. Nanoemulsiones en alimentos: preparación y aplicaciones. Temas Selectos de Ingeniería de Alimentos. **2015**. 9, 15-24.
7. Cardoso-Ugarte, G.A.; López-Malo, A.; Sosa-Morales, M.E. Cinnamon (*Cinnamonum zeylanicum*) essential oils. Essential Oils in Food Preservation, Flavor and Safety. **2016**. 339-347.
8. Chang, Y.; McLandsborough, L.; McClements, D.J. Fabrication, stability, and efficacy of dual-component antimicrobial nanoemulsions: essential oil (thyme oil) and cationic surfactant (lauric arginate). Food Chem. **2015**. 172, 298-283.
9. Chaudhary, S.; Kumar, S.; Kumar, V.; Sharma, R. Chitosan nanoemulsions as advanced edible coatings for fruits and vegetables: Composition, fabrication and developments in last decade. International Journal of Biological Macromolecules. **2020**. 152, 154-170.
10. Donsì, F.; Ferrari, G. Essential oil nanoemulsions as antimicrobial agents in food. Journal of Biotechnology. **2016**. 233, 106-120.
11. Ebrahimi, F.; Rastegar, S. Preservation of mango fruit with guar-based edible coatings enriched with Spirulina platensis and Aloe vera extract during storage at ambient temperature. Scientia Horticulturae. **2020**. 265. 109258.
12. Flores-López, M.L.; Cerqueira, M.A.; Rodríguez, D.J., & Vicente, A.A. Perspectives on Utilization of Edible Coatings and Nano-laminate Coatings for Extension of Postharvest Storage of Fruits and Vegetables. Food Engineering Reviews. **2015**, 8, 292-305.
13. Gennadios, A.; Wellet, C.L. Edible films and coatings from soy milk and soy protein. Cereal Foods World. **1991**. 36, 1004-1009.
14. Gharibzahedi, S.M.T; Jafari, S.M. Chapter 9 – Fabrication of nanoemulsions by ultrasonication. Jafari, S.M.; McClements, D.J. *Nanoemulsions*. Academic Press. **2018**. 233-285.
15. Ghost, V.; Mukherjee, A.; Chandrasekaran, N. Eugenol-loaded antimicrobial nanoemulsion preserves fruit juice against microbial spoilage. Colloids Surf. B: Biointerfaces. **2013a**. 114, 392-397.
16. Ghost, V.; Saranya, S.; Mukherjee, A.; Chandrasekaran, N. Cinnamon oil nanoemulsion formulation by ultrasonic emulsification: investigation of its bactericidal activity. J. Nanosci. Nanotechnol. **2013b**. 13, 114-122.
17. Hakansson, A. Chapter 7 – Fabrication of nanoemulsions by high-pressure valve homogenization. Jafari, S.M.; McClements, D.J. *Nanoemulsions*. Academic Press. **2018**. 175-206.
18. Hakansson, A.; Rayner, M. Chapter 5 – General principles of nanoemulsion formation by high-energy mechanical methods. Jafari, S.M.; McClements, D.J. *Nanoemulsions*. Academic Press. **2018**. 103-139.
19. Horison, R.; Sulaiman, F.O.; Alfredo, D.; Wardana, A.A. Physical characteristics of nanoemulsion from chitosan/nutmeg seed oil and evaluation of its coating against microbial growth on strawberry. Food Research. **2019**. 3(6), 821-827.

20. Huang, Q.; Yu, H.; Ru, Q. Bioavailability and delivery of nutraceuticals using nanotechnology. J. Food Sci. **2010**. 75 (1), R50-R57.
21. Israelachvili, J.N. Intermolecular and Surface forces, 3rd ed. Academic Press. **2011**. 1-674.
22. Jafari, S.M.; Assadpoor, E.; He, Y.; Bhandari, B. Re-coalescence of emulsion droplets during high-energy emulsification. Food Hydrocoll. **2008**. 22 (7), 1191-1202.
23. Jafari, S.M.; He, Y.; Bhandari, B. Production of sub-micron emulsions by ultrasound and microfluidization techniques. J. Food. Eng. **2007**. 82 (4), 478-488.
24. Jiao, W.; Shu, C.; Li, Cao, J.; Fan, X.; Jiang, W. Preparation of a chitosan-chlorogenic acid conjugate and its application as edible coating in postharvest preservation of peach fruit. Postharvest Biology and Technology. **2019**. 154, 129-136.
25. Kanasaki, Y.N.; Sagawa, N.; Deguchi, S. Two-step droplet formation in monodisperse nanodroplet generation in hydrothermal solution as revealed by spontaneous transformation of nanodroplets to swollen micelles in octane in water nanoemulsions. Journal of Colloid and Interface Science. **2021**. 221-226.
26. Kim, I.H.; Oh, Y.A.; Lee, H.; Song, K.B.; Min, S.C. Grape berry coatings of lemongrass oil-incorporating nanoemulsion. LWT – Food Science and Technology. **2014**. 58, 1-10.
27. Landfester, K.; Eisenblätter, J.; Rothe, R. Preparation of polymerizable miniemulsions by ultrasonication. JCT Research. **2004**. 1, 65-68.
28. Lee, S.J.; McClements, D.J. Fabrication of protein-stabilized nanoemulsions using a combined homogenization and amphiphilic solvent dissolution/evaporation approach. Food Hydrocoll. **2010**. 560-569.
29. Leong, T.S.H.; Wooster, T.J.; Kentish, S.E.; Ashokkumar, M. Minimising oil droplet size using ultrasonic emulsification. Ultrasonics Sonochemistry. **2009**. 16 (6), 721-727.
30. Li, W.; Chen, H.; He, Z.; Han, C.; Liu, S.; Li, Y. Influence of surfactant and oil composition on the stability and bacterial activity of eugenol nanoemulsions. Food Sci. Technol. **2015**. 62, 39-47.
31. Liang, R; Xu, S.; Shoemaker, C.F.; Li, Y.; Zhong, F.; Huang, Q. Physical and antimicrobial properties of peppermint oil nanoemulsions. J. Agric. Food Chem. **2012**. 60, 7548-7555.
32. Majeed, H.; Liu, F.; Hategekimana, J.; Sharif, H.R.; Qi, J.; Ali, B.; Bian, Y.Y.; Ma, J.; Yokoyama, W.; Zhong, F. Bactericidal action mechanism of negatively charged food grade clove oil nanoemulsions. Food Chem. **2016**. 197, 75-83.
33. Mason, T.G.; Wilking, J.N.; Meleson, K.; Chang, C.B.; Graves, S.M. Nanoemulsions: Formation, Structure, and Physical Properties. J. Phys.: Condens. Matter. **2006**. 18, R635-R666.
34. McClements, D.J. Advances in Colloid and Interface Science. Theoretical Prediction of Emulsion Color. Elsevier Science. **2002**. 63-89.
35. McClements, D.J. Edible Nanoemulsions: Fabrication, Properties, and Functional Performance. Soft Matter. **2011**. 7, 2297-2316.
36. McClements, D.J. Food Emulsions. Principles, Practices, and Techniques, 3rd ed.; CRC Press. **1999**. 1-27.
37. McClements, D.J.; Decker, E.A.; Weiss, J. Emulsion-based delivery systems for lipophilic bioactive components. J. Food Sci. **2007**. 72 (8), R109-R124.
38. McClements, D.J.; Jafari, S.M. Chapter 1 – General Aspects of Nanoemulsions and Their Formulation. Jafari, S.M.; McClements, D.J. *Nanoemulsions*. Academic Press. **2018**. 3-20.
39. Microfluidics. Microfluidizer Processor User Guide. **2014**. Available from: https://www.alfatest.it/keyportal/uploads/2017-microfluidics-chamber-user-guide.pdf . Accessed 3 December 2021.
40. Mohammadi, A.; Hashemi, M.; Hosseini, S.M.; Chitosan nanoparticles loaded with *Cinnamomum zeylanicum* essential oil enhance the shelf life of cucumber during cold storage. Postharvest Biology and Technology. **2015**. 110, 203-213.
41. Mohr, K. High-pressure Homogenization. Part II. The influence of cavitation on liquid-liquid dispersion in turbulence fields of high energy density. J. Food Eng. **1987**. 6 (4), 311-324.

42. Montero García, M.P.; Gómez-Guillén, M.C.; López-Caballero, M.E., & Barbosa-Cánovas, G.V. Edible Films and Coatings: Fundamentals and Applications, 1st ed.; CRC Press. Taylor & Francis Group. **2016**. 3-23.
43. Nazzaro, F.; Fratianni, F.; De Martino, L.; Coppola, R.; De Feo, V. Effect of essential oils on pathogenic bacteria. Pharmaceuticals Basel. **2013**. 6, 1451-1474.
44. Perazzo, A.; Preziosi, V. Chapter 3 – Catastrophic phase inversion techniques for nanoemulsification. Jafari, S.M.; McClements, D.J. *Nanoemulsions*. Academic Press. **2018**. 53-76.
45. Poverenov, E.; Danino, S.; Horev, B.; Granit, R.; Vinokur, Y; Rodov, V. Layer-by-Layer Electrostatic Deposition of Edible Coating on Fresh Cut Melon Model: Anticipated and Unexpected Effects of Alginate–Chitosan Combination. Food and Bioprocess Technology. **2014**. 7, 1424-1432.
46. Quemada, D.; Berli, C. Energy of interaction in colloids and its implications in rheological modelling. Adv. Colloid Interf. Sci. **2002**. 98, 51-85.
47. Rao, A. Rheology of fluid, semisolid, and solid foods: principles and applications. Springer Science & Business Media. **2013**. 1-15.
48. Rehman, A.; Qunyi, T.; Sharif, H.R.; Korma, S.A.; Karim, A.; Manzoor, M.F.; Mehmood, A.; Iqbal, M.W.; Raza, H.; Ali, A.; Mehmood, T. Biopolymer based nanoemulsion delivery system: An effective approach to boost the antioxidant potential of essential oil in food products. Carbohydrate Polymer Technologies and Applications. **2021**. 2, 1-16.
49. Sajjadi, S.; Jahanzad, F.; Yianneskis, M. Catastrophic phase inversion of abnormal emulsions in the vicinity of the locus of transitional inversion. Colloids and Surfaces A: Physicochem. Eng. Aspects. **2004**. 240, 149-155.
50. Sajjadi, S.; Jahanzad, F.; Yianneskis, M.; Brooks, B.W. Phase inversion in abnormal O/W/O emulsions. Ing. Eng. Chem. Res. **2003**. 42, 3571-3577.
51. Salas-Méndez, E.J.; Vicente, A.; Pinheiro, A.C.; Ballesteros, L.F.; Silva, P.; Rodríguez-García, R.; Hernández-Castillo, F.D.; Díaz-Jiménez, M.L.V.; Flores-López, M.L.; Villarreal-Quintanilla, J.A.; Peña-Ramos, F.M.; Carrillo-Lomelí, D.A.; de Rodríguez, D.J. Application of edible nanolaminate coatings with antimicrobial extract of *Flourensia cernua* to extend the shelf-life of tomato (*Solanum lycopersicum L.*) fruit. Postharvest Biology and Technology. **2019**. 150, 19-27.
52. Salvia-Trujillo, L.; Rojas-Graü, A.; Soliva-Fortuny, R.; Martín-Belloso, O. Physicochemical characterization and antimicrobial activity of food-grade emulsions and nanoemulsions incorporating essential oils. Food Hydrocolloids. **2015**. 43, 547-556.
53. Sanchís, E.; González, S.; Ghidelli, C.; Sheth, C.C.; Mateos, M.; Palou, L.; Pérez-Gago, M.B.P. Browning inhibition and microbial control in fresh-cut persimmon (Diospyros kaki Thunb. cv. Rojo Brillante) by apple pectin-based edible Coatings. Postharvest Biology and Technology. **2016**. 112, 186-193.
54. Seow, Y.X.; Yeo, C.R.; Chung, H.L.; Yuk, H.G. Plant essential oils as active antimicrobial agents. Crit. Rev. Food Sci. Nutr. **2014**. 54, 625-644.
55. Sivakumar, M.; Tang, S.Y.; Tan, K.W. Cavitation technology – a greener processing technique for the generation of pharmaceutical nanoemulsions. Ultrason. Sonochem. **2014**. 21, 2069-2083.
56. Sugumar, S.; Singh, S.; Mukherjee, A.; Chandrasekaran, N. Nanoemulsion of orange oil with non-ionic surfactant produced emulsion using ultrasonication technique evaluating against food spoilage yeast. Appl. Nanosci. **2015**. 6, 113-120.
57. Sugumar, S; Ghosh, V.; Mukherjee, A.; Chandrasekaran, N. Chapter 9 – Essential oil-based nanoemulsion formation by low- and high-energy methods and their application in food preservation against food spoilage microorganisms. Preedy, V.R. *Essential oils in food preservation, flavor and safety*. Academic Press. **2016**. 93-100.
58. Tadros, T.F. Rheology of dispersions: principles and applications. Wiley-VCH Verlag GmbH & Co. KGaA. **2010**. 1-199.
59. Tajkarimi, M.M; Ibrahim, S.A.; Cliver, D.O. Antimicrobial herb and spice compounds in food. Food Control. **2010**. 21, 1199-1218.

60. Taylor, P. Ostwald ripening in emulsions. Adv. Colloid Interf. Sci. **1998**. 75, 107-163.
61. van der Schaaf, U.S.; Karbstein, H.P; Chapter 6 – Fabrication of nanoemulsions by rotor-stator emulsification. Jafari, S.M.; McClements, D.J. *Nanoemulsions*. Academic Press. **2018**. 141-174.
62. Velikov, K.P.; Pelan, E. Colloidal Delivery Systems for Micronutrients and Nutraceuticals. Soft Matter. **2008**. 4, 1964-1980.
63. Villalobos-Castillejos, F.; Granillo-Guerrero, V.G.; Leyva-Daniel, D.E.; Alamilla-Beltrán, L.; Gutiérrez-López, G.F.; Monroy-Villagrana, A.; Jafari, S.M. Chapter 8 – Fabrication of nanoemulsions by microfluidization. Jafari, S.M.; McClements, D.J. *Nanoemulsions*. Academic Press. **2018**. 207-232.
64. Vladisavljevic, G.T. Chapter 10 – Fabrication of nanoemulsions by membrane emulsification. Jafari, S.M.; McClements, D.J. *Nanoemulsions*. Academic Press. **2018**. 287-346.
65. Walstra, P. Principles of emulsion formation. Chemical Engineering Science. **1993**. 48 (2), 333-349.
66. Wang, X; Jiang, Y.; Wang, Y.W.; Huang, M.T.; Ho, C.T.; Huang, Q. Enhancing anti-inflammation activity curcumin through O/W nanoemulsions. Food Chemistry. **2008**. 108, 419-424.
67. Weiss, J.; Gaysinsky, S.; Davidson, M.; McClements, D.J. Nanostructured encapsulation systems: food antimicrobials. Global Issues in Food Science and Technology. **2009**. 425-479.
68. Wooster, T.J.; Golding, M.; Sanguansri, P. Impact of oil type on nanoemulsion formation and Ostwald ripening stability. Langmuir. **2008**. 24, 12758-12765.
69. Xiao, Z.; Luo, Y.; Luo, Y.; Wang, Q. Combined effects of sodium chlorite dip treatment and chitosan coatings on the quality of fresh-cut d'Anjou pears. Postharvest Biology and Technology. **2011**. 62, 319-326.
70. Zhang, Z.; McClements, D.J. Chapter 2 – Overview of nanoemulsion properties: stability, rheology, and appearance. Jafari, S.M.; McClements, D.J. *Nanoemulsions*. Academic Press. **2018**. 21-49.
71. Ziani, K.; Chang, Y.H.; McLandsborough, L.; McClements, D.J. Influence of surfactant charge on antimicrobial efficacy of surfactant-stabilized thyme oil nanoemulsions. J. Agric. Food Chem. **2011**. 59, 6247-6255.

Acronyms

CEO: *Cinnamomum zeylanicum* essential oil

CSN: chitosan nanoparticles

CPI: catastrophic phase inversion

EO: essential oil

GRAS: generally recognized as safe

HPVH: high-pressure valve homogenization

O/W: oil-in-water

O/W/O: oil-in-water-in-oil

PIC: phase inversion temperature

PIT: phase inversion composition

TPI: transitional phase inversion

W/O: water-in-oil

W/O/W: water-in-oil-in-water