CHAPTER 137

Nanoemulsions and cyclodextrins inclusion complexes as methods of protection of essential oils for the development of bioherbicides

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ABSTRACT
Essential oils are a source of promising bioactive compounds for the development of environmentally friendly herbicides. However, the low solubility, high volatility, and degradability make it impossible to apply essential oils under field conditions. Encapsulation in nanoemulsions and inclusion complexes can overcome these disadvantages by increasing the stability of these products. Recent studies have shown the ability of some protected essential oils to suppress weed development. This chapter reviews the processes of encapsulation and use of these essential oils. Inclusion complexes can be prepared using oligosaccharides such as cyclodextrins by the molecular inclusion method, while nanoemulsions can be obtained through low and high-energy methods. The development of nanoemulsions and inclusion complexes can increase the solubility and physicochemical stability of essential oils, providing stable formulations for use in agricultural fields.

Keywords: Allelopathy, Bioherbicides, Cyclodextrins, Encapsulation.

1 INTRODUCTION

1.1 ESSENTIAL OILS

Essential oils (OEs) are complex mixtures of volatile compounds synthesized by the secondary metabolism of aromatic plants, being found in flowers, fruits, seeds, leaves, stems, and roots (AUMEERUDDY-ERALFI et al., 2018; HANIF et al., 2019). They are produced and stored by secretory structures located on the surface (idioblasts, cavities, and secretory ducts) or within plant tissues (trichomes, epidermal cells, and osmophores) (SVOBODA and SVOBODA, 2000; AUMEERUDDY-ELFI, et al., 2018).

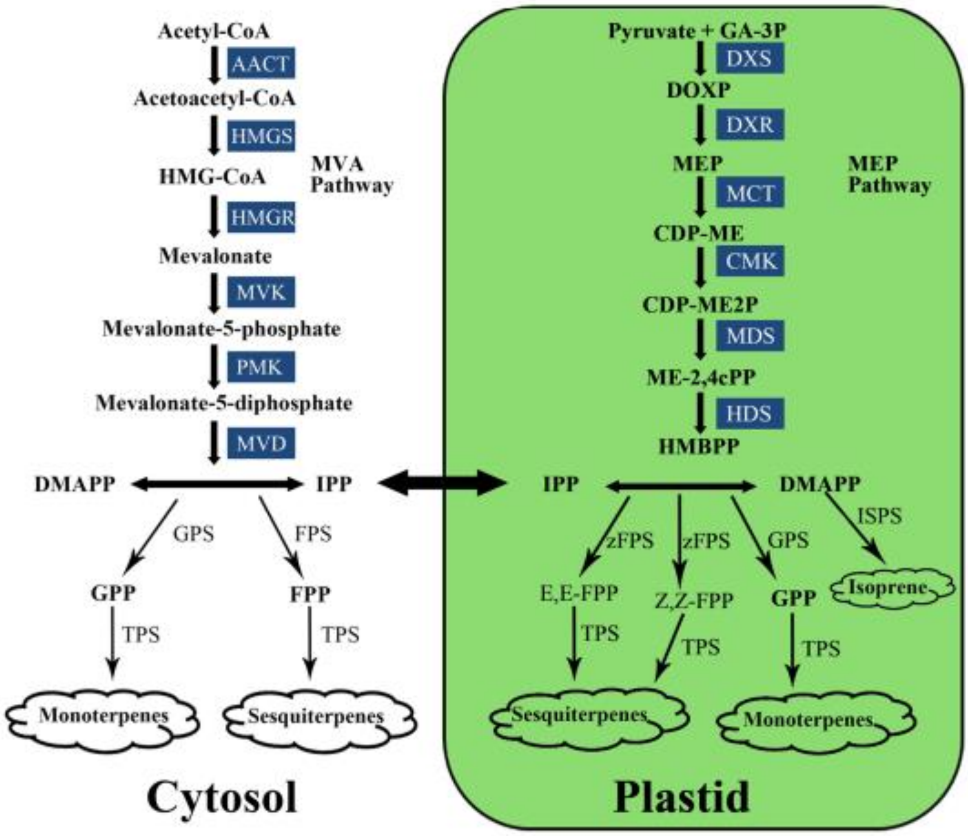
Formed by more than 300 volatile and semivolatile organic compounds of low molecular weight, the main constituents of OEs comprise terpenoids (monoterpenes, sesquiterpenes, and their oxygenated derivatives) and phenylpropanoids. Aliphatic alcohols, esters, aldehydes, fatty acids, hydrocarbons, sterols, waxes, flavonoids, carotenoids, nitrogen derivatives, and sulfur can also be found in the volatile and non-volatile fraction of OEs (DHIFI et al., 2016; HANIF et al., 2019).

Terpenoids and phenylpropanoids are synthesized by distinct biosynthetic pathways and by different primary metabolic precursors. Phenylpropanoids are synthesized through the shikimic acid pathway. Its main precursors are cinnamic acid and p-hydroxycinnamic acid, derivatives of phenylalanine and tyrosine, respectively. Terpenoids are derived from the condensation of a five-carbon isoprene unit, isopentenyl diphosphate (IPP), with its allyl isomer, dimethylallyl diphosphate (DMAPP). In higher plants, IPP is synthesized by two separate compartments via two independent pathways: the mevalonate pathway (MVA) (cytoplasm) and the mevalonate-independent pathway (deoxylulose phosphate) (MEP) (chloroplasts). In the MVA pathway, IPP is formed through mevalonic acid resulting from the condensation of three acetyl-CoA molecules. In the MEP pathway, IPP is formed from the condensation of pyruvate and glyceraldehyde-3-phosphate. The condensation of an IPP unit with its isomer, DMAPP, produces a geranyl diphosphate (GPP), an immediate precursor to monoterpenes (C_{10}). Condensation of GPP with IPP leads to farnesyl diphosphate (FPP), the immediate precursor of sesquiterpenes (C_{15}), and condensation of FPP with IPP results in geranylgeranyl diphosphate (GGPP), the precursor of diterpenes (C_{20}) (ZUZARTE and SALGUEIRO, 2015; ABBAS et al., 2017) (Figure 1).

The chemical composition of OEs is strongly influenced by physiological, environmental, and genetic factors. The oil extracted from different plant organs of the same plant may have different chemical compositions, as well as, may vary according to the ontogenesis of the individual. Geographic location, climatic conditions, soil type, nutrient and water availability, and sunlight levels are other factors that cause qualitative and quantitative differences in OEs. Genetic differences explain the distinct chemotypes between populations of the same species (TAIZ and ZEIGER 2010; DHIFI et al., 2016; SALGUEIRO et al., 2010).

OEs, as seen earlier, are rich in terpenoids. These bioactive components are involved in various physiological processes and ecological functions of plants in nature. The genes that synthesize terpenes have differential expression in response to biotic and abiotic environmental factors, being produced according to the specific needs of plants (GERSHENZON and DUDAREVA, 2007). Terpenoids are involved in mediating plant-organism and plant-environment interactions, such as (i) attraction of seed disseminators and pollinators; (ii) attraction of herbivores for protection from aggressive natural enemies; (iii) sending attack messages and initiating defense responses in neighbors of unattacked plants; (iv) protection against the attack of pathogens; (v) protection against environmental stresses (thermotolerance and photoprotection); (vi) allelopathic activity against neighboring competing or parasitic plants (ABBAS et al., 2017) (Figure 2).

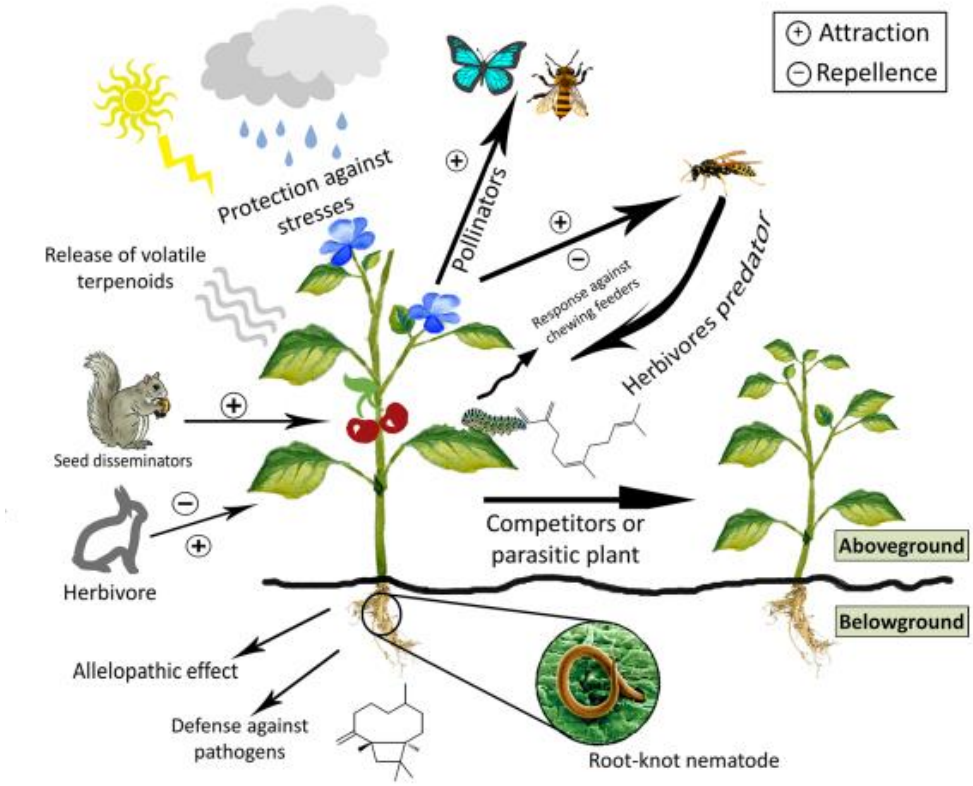
Figure 1. Terpenoid biosynthesis pathway in plants.



Source: Abbas et al. (2017)

Terpenoids are antiseptic, anti-inflammatory, bactericidal, and antiviral (HANIF et al., 2019). In this context, the variability of bioactive compounds and biocides in essential oils makes them interesting for the pharmaceutical, sanitary, cosmetic, agricultural, and food industries (BUCHBAUER, 2000). Studies indicate cytotoxic, antiparasitic (BEZERRA et al., 2022), antimicrobial, antioxidant (COIMBRA et al., 2022; CHEBBAC et al., 2022; ABD-ELGAWAD et al., 2022), antifungal (MOUMNI et al., 2021; ROANA et al., 2021), antiviral (SOBRINHO et al., 2021), anti-inflammatory (SANTOS et al., 2021), insecticide (ZIMMERMANN et al., 2021; CHEN et al., 2021); repellent (LUO et al., 2022) and phytotoxic (POURESMAEIL et al., 2022; HAN et al., 2021; Jiang et al., 2021a; MAHANTA et al., 2022) from several OEs.

Figure 2. Functions of terpenoids in mediating interactions between plants and the environment.



Source: Abbas et al. (2017).

1.2 HERBICIDAL ACTIVITY OF ESSENTIAL OILS

In recent years the allelopathic activity of OEs has been applied and directed to the control and management of weeds as an alternative to synthetic herbicides, harmful to the environment and human health (HAN et al., 2021, ISSA et al., 2020; Singh et al., 2020). Evaluated as herbicides and pesticides of the next generation, natural compounds are promising sources for the development of ecologically correct herbicides, since they are biodegradable, exhibit relatively low toxicity to non-target organisms, present high structural diversity, and new molecular target sites, offer low risk of resistance induction (DAYAN et al., 2009; DAYAN & DUKE, 2014; ISMAN, 2015; PAVELA & BENELLI, 2016).

OEs of species such as *Ambrosia artemisiifolia L.*, Vitex negundo *L.*, *Artemisia fragrans* Willd., *Mentha longifolia* (L.) Huds, *Thymus proximus* Serg., have been reported for their ability to suppress different weed species through inhibition of germination, seedling growth (coleoptile and root length), and photosynthetic pigments. In addition to these effects, changes in the activity of photosynthetic machinery enzymes (catalase, peroxidase, ascorbate peroxidase, and superoxide dismutase), oxidative stress induction, and leaf wilting have also been observed (HAN et al., 2021; ISSA et al., 2021; POURESMAEIL et al., 2020; Singh et al., 2020; ZHOU et al., 2021).

Although the amount of research pointing to the bioherbicidal potential of OEs is extensive, only six commercial products containing OEs and/or their active compounds are commercially available. They are: GreenMatch (55% d-limonene), Matratec [50% clove oil (*Syzygium aromaticum* L.)], WeedZap [45% clove oil + 45% cinnamon oil (*Cinnamomum zeylanicum Blume*)], GreenMatch EX [50% lemongrass oil (*Cymbopogon citratus* (DC.) Stapf)], Avenger Weed Killer (70% d-limonene) and Weed Slayer (6% eugenol) (VERDEGUER et al., 2020).

The low conversion of laboratory studies into practical applications is linked to physicochemical characteristics of OEs that hinder and make their use by industries difficult and unfeasible (KRZYZOWSKI et al., 2020; TUREK & STINTZING, 2013; PAVELA & BENELLI, 2016; CAMPOLO et al., 2020). The OEs have little solubility in water which hinders their distribution in agricultural fields, in addition, the direct application of these products on the crops can cause severe desiccation of the crops to be protected, harming the quality and productivity of the non-target plants (KRZYZOWSKI et al., 2020; DE ALMEIDA et al., 2010; IBÁÑEZ & BLÁZQUEZ, 2020).

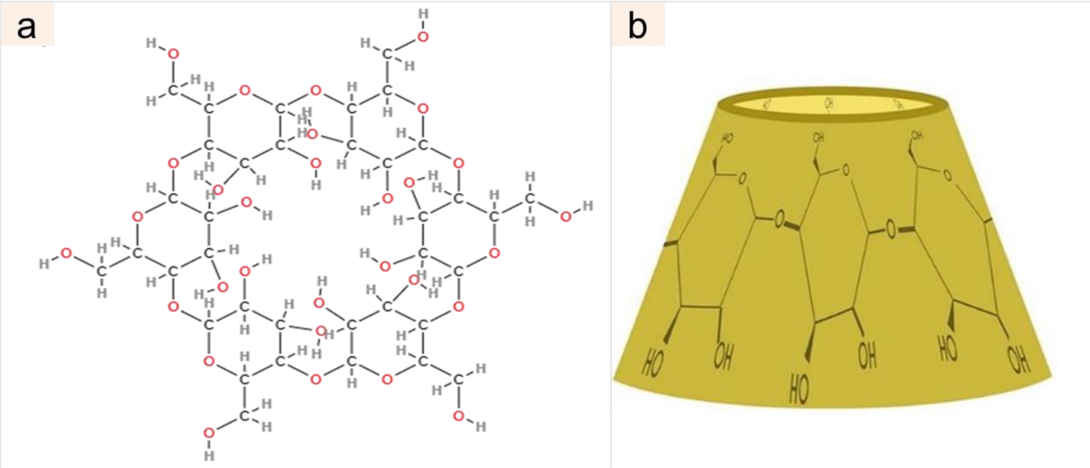
Limiting aspects such as volatility, degradability, and self-oxidation make OEs unstable physically. Exposure to heat, moisture, light, and oxygen causes them to degrade easily. Although this ensures the non-persistence and non-accumulation of their residues in the environment and agricultural production, the low persistence of OEs can reduce their effectiveness against pests (TUREK & STINTZING, 2013; PAVELA & BENELLI, 2016; CAMPOLO et al., 2020), such as weeds. To overcome these problems, encapsulation through cyclodextrins and nanoemulsion inclusion complexes may be the key to overcoming the disadvantages of using OEs.

Encapsulation is a process where bioactive components are retained within a coating matrix of synthetic material or natural polymer, leading to the formation of small capsules on the order of nanometers (RODRÍGUEZ et al., 2016; SCHMITT & TURGEON, 2011). This process protects the active ingredients of the core from external environmental conditions through their slow release, decreasing their reactivity and increasing their stability over time (GHARSALLAOUI et al., 2007).

2 CYCLODEXTRINS INCLUSION COMPLEX

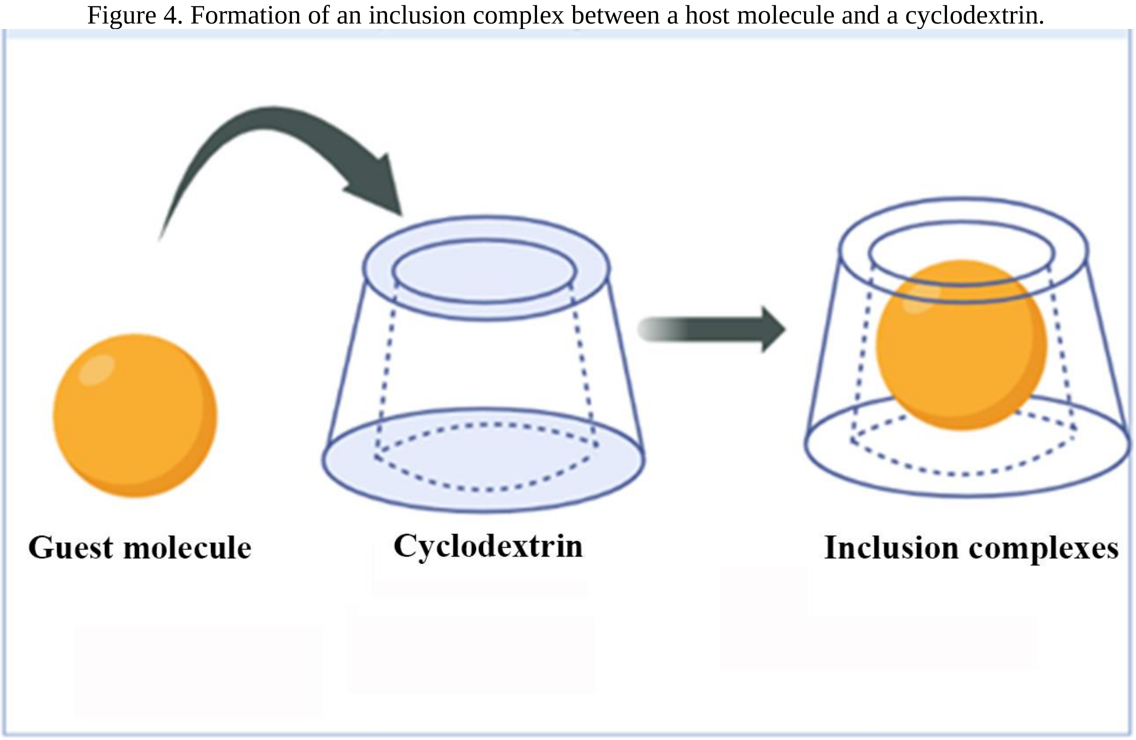
Cyclodextrins (CyDs) are cyclic oligosaccharides of glucose (α-d-glucopyranose) produced from enzymatic conversion, degradation, and cyclization of starch (SILVA et al., 2020). Known as Schardinger's dextrins, cyclo maltose, and cyclo amylase, CyDs have a three-dimensional, toroidal-cone-shaped, hollow, truncated structure. In them, groups of secondary hydroxyls appear on the widest edge of the cone, attached to the C-2 and C-3 atoms, while the primary hydroxyls are located in the narrowest part, attached to C6. The outer face of the CyDs contains the CH, H-1, H-2, and H-4 groups and the hydroxy groups of the α-d-glucopyranose that are oriented towards the outside of the cone. The inner part is lined by rings of CH groups (H-3 and H-5) and by a ring containing hydrocarbons and ether interconnected by glycosidic bonds with oxygen (O-4 and O-5) (Figure 3). In this sense, the surface of the CyDs is hydrophilic, while the cavity of the cone with lower polarity is hydrophobic (CABRAL-MARQUES et al., 1994). This amphipathic property of CyDs allows the entrapment of hydrophobic organic molecules in their cavity, which in aqueous solutions form a host-host system, called the inclusion complex (SUGANYA; ANURADHA, 2017) (Figure 4).

Figure 3. Chemical structure of cyclodextrin. a) structural formula. b) geometric shape.



Source: a) Loren Cristina Vasconcelos and Luiza Alves Mendes.

Figure 4. Formation of an inclusion complex between a host molecule and a cyclodextrin.

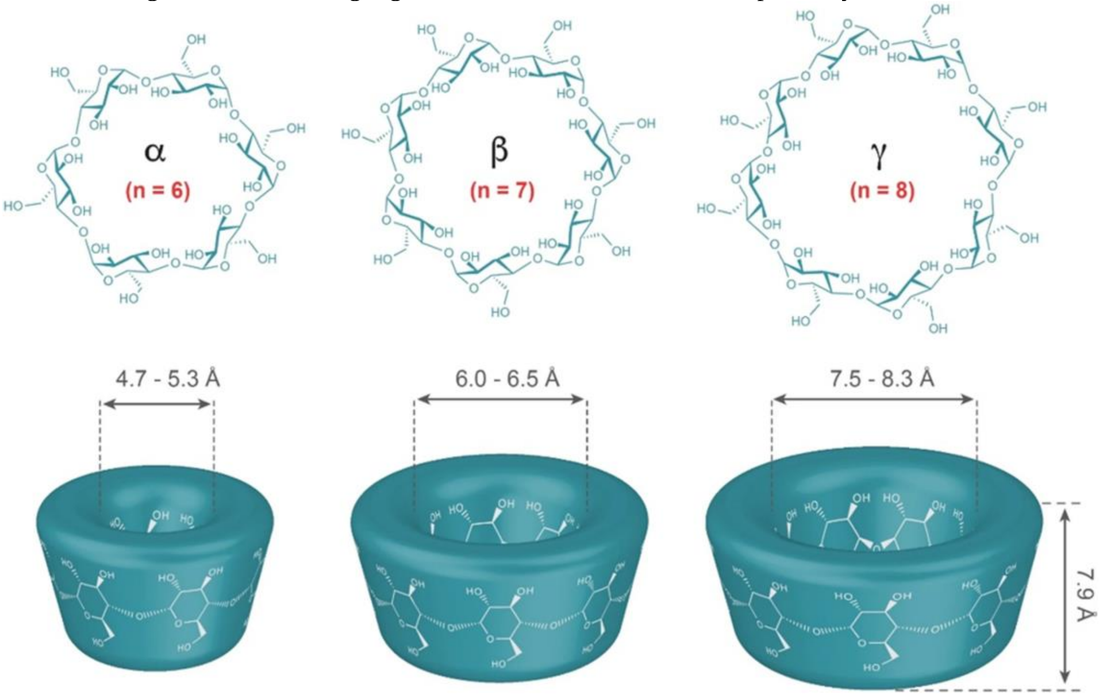


Source: Loren Cristina Vasconcelos.

The formation of CyDs inclusion complexes is equivalent to molecular encapsulation, since the host molecules are isolated from each other and remain dispersed at the molecular level in an oligosaccharide matrix (MARQUES et al., 2010). The encapsulation by CyDs modifies several physicochemical properties of the encapsulated host molecule bringing advantages such as increased solubility of poorly soluble compounds; increased chemical and physical stability against decomposition, oxidation, hydrolysis, or evaporation loss; control of sublimation and volatility; physical separation of incompatible compounds; transformation of liquid compounds into crystalline form (powder); increased effectiveness and potency of the compound; reduction of the toxicity of the material; control of material release; and modification of flavors and odors (MARQUES et al., 2010; WADHWA et al., 2017).

CyDs can be categorized into natural and chemically modified (semisynthetic) cyclodextrins (SZEJTLI and OSA et al., 1996). Among the natural ones, the most common forms include α, β and γ cyclodextrins, they are differentiated based on the number of units of α-d-glucopyranose, presenting six, seven, and eight units of α-d-glucopyranose, respectively (DEL VALLE et al., 2004) (Figure 5).

Figure 5. Schematic representations of the three main cyclodextrins, namely α-cyclodextrin, β-cyclodextrin, and γ-cyclodextrin, containing six, seven, and eight glucose units in their structure, respectively, and their dimensions.



Source: Crini and Aleya (2021).

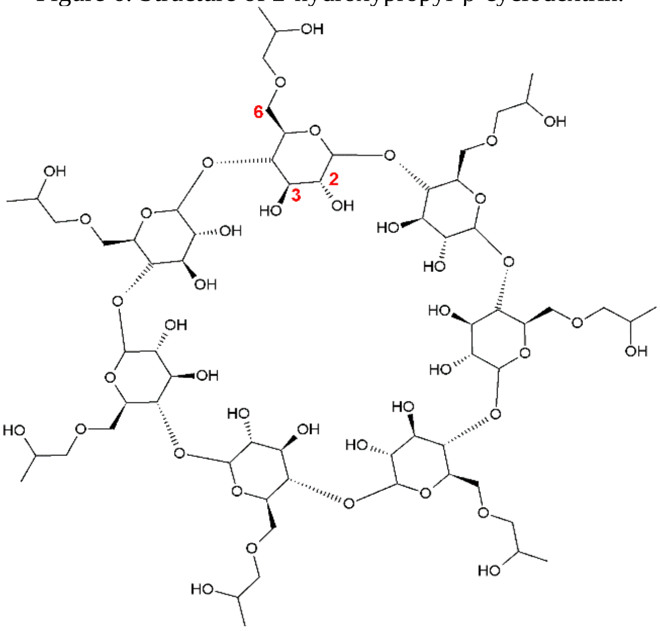
β-cyclodextrin is the most widely used natural cyclodextrin due to the proper size of its cavity and its ability to improve the solubility and stability of different compounds, as well as having no mutagenic effects and having a low cost (WADHWA et al., 2017; BEZERRA et al., 2020). However, just like the other natural cyclodextrins, it has low solubility. To overcome this deficiency, functional groups such as hydroxyalkyl and methyl are introduced in the CyDs of origin (SZENTE and SZEJTLI, 1999), forming the semisynthetic CyDs. Among the semisynthetic CyDs, 2-hydroxypropyl-β-cyclodextrin (HPβCD), sulfo-butyl-ether-β-CD (SBβCD) and randomized methyl-β-CD (RMβCD) are the most used. These CyDs, in addition to having greater solubility, have improved physicochemical properties, presenting greater inclusion capacity and greater stability than natural cyclodextrins (DE LIMA; ALVES; SANTANA, 2012; BREWSTER; LOFTSSON, 2007).

2.1 2-HIDROXIPROPIL-B-CICLODEXTRINA

2-Hydroxypropyl-β-cyclodextrin (HPβCD) is a derivative of β-cyclodextrin obtained by replacing –OH groups with hydroxypropyl groups on the outer surface (CUI; SIVA; LIN, 2019). The addition of hydroxypropyl groups usually occurs on the 6 carbon attached to the primary hydroxyls, extending the hydrophobic region of the cavity. This modification increases the interaction with the host molecule, thus obtaining a greater complexation during the formation of the inclusion complexes (JANSOOK; OGAWA; LOFTSSON, 2018; BREWSTER; LOFTSSON, 2007). The OH groups of carbons 2 and 3 of β-cyclodextrin are also available for structural modifications without the cavity structure being lost (MacNicol; McKendrick; Wilson, 1978) (Figure 6). HPβCD has a solubility 20 times higher than natural β-cyclodextrin, in addition to having greater selectivity for guest molecules and higher complexation efficiency (CUI; SIVA; LIN, 2019).

Inclusion complexes with HPβCD have been used for the encapsulation of various essential oils to increase solubility, reduce volatility, increase release time and physicochemical stability of their volatile and lipophilic compounds (CUI; SIVA; LIN, 2019; HOU et al., 2022; YUAN et al., 2019; YUAN; WANG; CUI, 2019; ZANG; YUAN; SUI, 2018; Jiang et al., 2021b).

Figure 6. Structure of 2-hydroxypropyl-β-cyclodextrin.



Source: Luiza Alves Mendes.

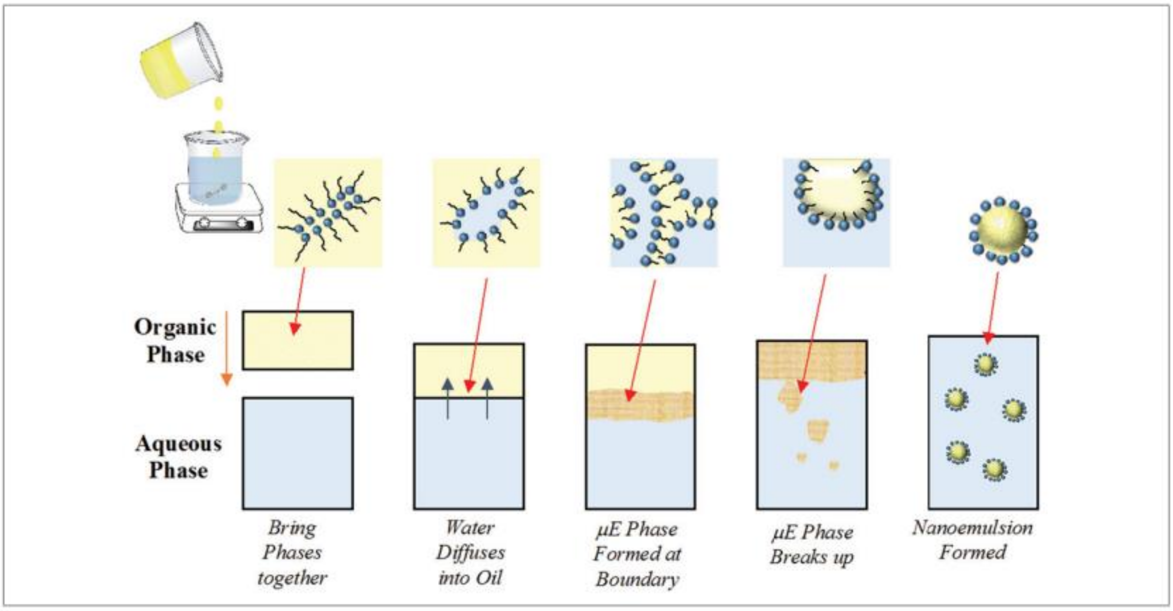
Recently, a study evaluated the formation of inclusion complexes of cuminaldehyde, a major constituent of *Cuminum cyminum essential* oil (cumin), with HPβCD through ultrasound. The results showed that the phenyl ring with the aldehyde group of cuminaldehyde was deeply inserted into the hydrophobic cavity of HPβCD, increasing its bioavailability. The inclusion of cuminaldehyde promoted greater solubility in water, greater thermal stability, and greater antibacterial activity against *Escherichia coli* and *Staphylococcus aureus* (CUI; SIVA; LIN, 2019). In another study, encapsulation of nanoemulsions of *Cinnamomum verum essential* oil (cinnamon) in HPβCD reduced the size of nanoemulsion particles, improved controlled essential oil release, and increased antibacterial activity. HPβCD also increased the stability of nanoemulsions under different temperatures (HOU et al., 2021). Studies with the essential oils of *Illicium verum* Hooker f., *Eucalyptus staigeriana*, and *Lavandula angustifolia* encapsulated in HPβCD showed that components of essential oils were preferentially included in the complex, presenting differences in content regarding the pure essential oil. Selective encapsulation promoted by HPβCD improved the thermal stability, controlled release, and antimicrobial activity of essential oils (YUAN et al., 2019; YUAN; WANG; CUI, 2019; ZANG; YUAN; SUI, 2018). Jiang et al. (2021) when preparing complexes for the inclusion of *Melaleuca alternifolia* essential oil with HPβCD by the ultrasonic method achieved an encapsulation efficiency greater than 80%. The inclusion complex showed greater stability, in addition to the long-lasting controlled release and antifungal properties against the growth of *M. fructicola*, which causes brown rot in peach fruits.

El-Alam et al. (2020) tested for the first time the phytotoxic activity of nine essential oils encapsulated with HPβCD in vitro bioassays with *Lolium perenne L. and* Lactuca sativa *L.* Encapsulation with HPβCD reduced the volatility of essential oils, however, there was no significant improvement in their phytotoxic efficiency. Although the in vitro results were not expressive, analyses under greenhouse or field conditions are essential to determine the efficiency of the controlled release of these essential oils. Published studies evaluating inclusion complexes formed by β-cyclodextrins and HPβCD have shown the efficiency of these encapsulations in increasing water solubility, thermostability, and oxidation resistance, as well as promoting the controlled release of synthetic herbicides such as bentazon, cyanazine, and diuron (AZZALI et al., 2019; Yañez et al., 2012; Gao et al., 2020; GAO et al., 2019). These results indicate that inclusion complexes are interesting and effective systems for the release of bioactive compounds in agriculture. In this sense, the advantages provided by the formation of HPβCD inclusion complexes can be explored and applied to improve essential oils with herbicidal activity, since studies with this purpose of application are scarce in the literature.

2.2 NANOEMULSIONS

Nanoemulsions (NEs), also known as ultrafine emulsions, mini emulsions, and submicron emulsions, consist of two immiscible liquids where droplets of size from 20 to 500 nm of one liquid phase (dispersed phase) are suspended in the other liquid phase (continuous phase) (SHARMA; VISHT; Kulkari, 2010; SOLANS and SOLÉ, 2012; GANTA et al., 2014; KOMAIKO; MCCLEMENTS, 2016; LU et al., 2012). A typical nanoemulsion consists of three main parts: oil, water, and an emulsifier/surfactant. Surfactants act in the dispersed phase of the emulsion. By decreasing interfacial tension and surface energy per unit area between the organic (oil) and aqueous (water) phases, they cause a rupture of the droplets and hinder their regrouping (coalescence), leading to the formation of extremely small droplets. Surfactants are also involved in the stabilization of NEs through repulsive electrostatic forces and steric impediment (MASON et al., 2006; MAA and HSU, 1999) (Figure 7).

Figure 7. Nanoemulsions. Schematic representation of the basic mechanism for the formation of nanoemulsions.

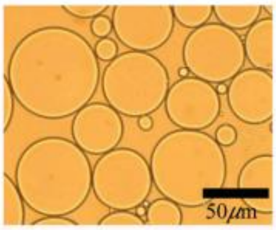
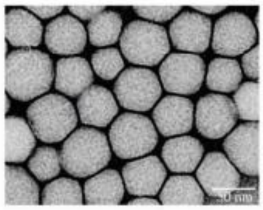
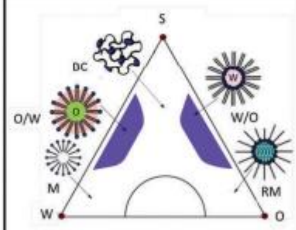


Source: Komaiko and McClements (2016).

NEs can be classified into three types: oil in water (O/A), when oil is defined as the dispersed phase and water is defined as the continuous phase; water in oil (A/O), when the opposite occurs; double or bi continuous (A/O/A or O/A/O) when the oil and the aqueous phase are separated by the surfactant layer (DEVARAJAN and RAVICHANDRAN, 2011; TAYEB and SAINSBURY, 2018). O/A emulsions are used to encapsulate and protect lipophilic active agents, such as sterols and carotenoids, while A/O emulsions encapsulate hydrophilic compounds, such as polyphenols (ZUIDAM and SHIMONI, 2010).

Droplet size and stability characteristics are responsible for differentiating between classical emulsions (macroemulsions), microemulsions, and nanoemulsions. Nanoemulsions, as well as macroemulsions, are thermodynamically unstable, that is, in a certain period the separation of phases occurs. However, unlike macroemulsions, their small size makes them kinetically stable over long timescales, thus being less subject to physical change. Microemulsions, although small in size (10 to 100 nm), are thermodynamically stable in composition, being susceptible to temperature changes and physical changes in general (MCCLEMENTS, 2012; ANTON and VANDAMME, 2011; GUPTA et al., 2016) (Figure 8).

Figure 8. Comparison of macroemulsions, nanoemulsions, and microemulsions about size, shape, stability, preparation method, and polydispersity.

	macroemulsions	nanoemulsions	microemulsions
			
size	1-100 µm	20-500 nm	10-100 nm
shape	spherical	spherical	spherical, lamellar
stability	thermodynamically unstable, weakly kinetically stable	thermodynamically unstable, kinetically stable	thermodynamically stable
method of preparation	high & low energy methods	high & low energy methods	low energy method
polydispersity	often high (>40%)	typically low (<10-20%)	typically low (<10%)

Source: Gupta et al. (2016).

NEs are manufactured from two main approaches based on energy input: high-energy methods and low-energy methods. High-energy methods use mechanical devices to generate high-shear forces by dividing the disperse into small droplets. These methods usually include high-pressure homogenization and ultrasound. In high-pressure homogenization, the mixture of oil, water, and surfactant is pushed through a small opening, which results in the acceleration of the fluid as it enters the system. Subsequent accelerations and decelerations result in high tangential and shear forces forcing the droplets to split into smaller segments (GUPTA et al., 2016; GUPTA, 2020; SANGUANSRI et al., 2013; Shubert et al., 2003; JAFARI et al., 2017). In the ultrasound method, the electrical input is converted into pressure fluctuations that generate cavitation bubbles. When cavitation bubbles, shear forces are generated by breaking the droplets into smaller segments (ABBAS et al., 2013; HAKANSSON and RAYNER, 2018).

Low-energy methods harness the internal chemical potential of the system to formulate NEs. When the system undergoes a phase inversion in response to changes in composition or temperature and undergoes a state of low interfacial tension the macroemulsions are broken down into smaller droplets, forming the NES. The main low-energy methods are emulsion inversion point (EIP) and phase inversion temperature (PIT). In IPE the A/O and O/A emulsions undergo a phase change (A/O to O/A or O/A to A/O) at room temperature when the second liquid phase is introduced. The lack of affinity of the surfactant with the initial phase of the mixture causes it to migrate toward the interface when the second phase is added, thus occurring the transformation of the emulsion from A/O to O/A or O/A to A/O. In the PIT method, the components are mixed at a temperature higher than the phase inversion temperature of the mixture, being then cold. When passing through the phase inversion temperature occurs the transformation of the mixture from A/O to O/A or from O/A to A/O (GUPTA et al., 2016; GUPTA, 2020; JAFARI et al., 2017).

NEs have been widely used as carrier agents and nanoencapsulators of OEs. Studies report the ability of NEs to improve the chemical stability of OEs by reducing volatilization and degradation processes such as oxidation (KREUTZ et al., 2021; FLORES et al., 2011; PAVONI et al., 2020). Increased bioactivity, stability, and controlled release of volatile compounds from OEs are also reported (CHUESIANG; SANGUANDEEKUL; SIRIPATRAWAN, 2021; CHU et al., 2020; Moghimi et al., 2016; OSANLOO et al., 2017; OSANLOO et al., 2018; SEIBERT et al., 2018; MANSOURI et al., 2020).

Recently, NEs with peppermint essential oil (*Mentha* × *piperita* L.) prepared by the high-energy emulsification method (ultrasonication) were evaluated for their bioherbicidal activity in seeds and young plants of corn (Zea mays *L.) and its weed rice grass* (Echinochloa crus-galli) (L.) Beauv.). Calorimetric analyses showed that the rice grass seedlings treated with the NEs presented lower thermal power curves than the control seedlings. According to the authors, the changes in the heat emission patterns of rice grass point to the inhibitory effect of peppermint NEs on metabolism, which explains the changes found in the content and carbohydrate composition of the seeds (storage material used in the early stages of germination). Herbicides effects such as leaf necrosis and reduction of length, shoot dry mass, relative water content in cells, and fluorescent chlorophyll a were also expressive in rice grass seedlings, indicating a greater susceptibility of rice grass to NEs when compared to corn. The NEs of peppermint essential oil showed good physicochemical characteristics and stability, exhibiting droplets of very small size (around 100 nm) and a very low polydispersity index after 30 days of storage (RYS et al., 2022).

NEs of fennel essential oil (*Foeniculum vulgare* Mill.) prepared by the ultrasonic emulsification method also demonstrated bioherbicidal properties on *Phalaris minor* Retz., *Avena ludoviciana* Durieu, *Rumex dentatus L* . and *Medicago denticulata* Willd, wheat weeds (*Triticum aestivum* L.). The NEs were more effective than the emulsion of the essential oil *of T. aestivum*, causing greater inhibitory effect on the germination and growth of seedlings, increased permeability and leakage of electrolytes by the membrane. The increased activities of the enzymes catalase and peroxidase also indicate that NEs caused oxidative stress due to overproduction of reactive oxygen species and free radicals. Additionally, fennel essential oil NEs were stable to centrifugation, dilution, and 30-day storage at room temperature (KAUR et al., 2021).

Similar results were found in the weeds *Amaranthus retroflexus* and *Chenopodium album* treated with NEs of the essential oil of garden safety (*Satureja hortensis*) obtained by the low energy method (catastrophic phase inversion). Under laboratory conditions, the NEs inhibited according to the increase in concentrations the germination percentage, the germination speed index and the length of the root and shoot of *A. retroflexus* and *C. album.* Under greenhouse conditions both weeds (stages of 2-4 true leaves) sprayed with the NEs of *S. hortensis* had an increase in the percentage of lethality and the reduction of fresh weight, dry weight, leaf area, length of the primary root and length of the primary aerial part of the seedlings. The total chlorophyll content also showed a decreasing pattern in a dose-dependent manner. In addition to these effects, a significant decline in membrane integrity and increased electrolyte leakage were also observed. The NEs of *S. hortensis* showed low polydispersity index and good stability at room temperature in 30 days, with droplet size below 130 nm (HAZRATI et al., 2017). Given these results, the formulation of nanoemulsions, in addition to overcoming the low solubility in water, is a promising and efficient approach to decrease volatility and increase the bioactivity and physical stability of OEs with herbicidal properties, thus contributing to the sustainable management of weeds in organic agricultural systems.

REFERENCES

Abbas, F., Ke, Y., Yu, R., Yue, Y., Amanullah, S., Jahangir, M. M., & Fan, Y. (2017). Volatile terpenoids: multiple functions, biosynthesis, modulation and manipulation by genetic engineering. *Planta*, *246*(5), 803–816. https://doi.org/10.1007/s00425-017-2749-x

Abbas, S., Hayat, K., Karangwa, E., Bashari, M., & Zhang, X. (2013). An Overview of Ultrasound-Assisted Food-Grade Nanoemulsions. *Food Engineering Reviews*, *5*(3), 139–157. https://doi.org/10.1007/s12393-013-9066-3

Abd-ElGawad, A. M., El-Amier, Y. A., Bonanomi, G., Gendy, A. E.-N. G. E., Elgorban, A. M., Alamery, S. F., & Elshamy, A. I. (2022). Chemical Composition of Kickxia aegyptiaca Essential Oil and Its Potential Antioxidant and Antimicrobial Activities. *Plants*, *11*(5), 594. https://doi.org/10.3390/plants11050594

Anton, N., & Vandamme, T. F. (2010). Nano-emulsions and Micro-emulsions: Clarifications of the Critical Differences. *Pharmaceutical Research*, *28*(5), 978–985. https://doi.org/10.1007/s11095-010-0309-1

Aumeeruddy-Elalfi, Z., Lall, N., Fibrich, B., Blom van Staden, A., Hosenally, M., & Mahomoodally, M. F. (2018). Selected essential oils inhibit key physiological enzymes and possess intracellular and extracellular antimelanogenic properties in vitro. *Journal of Food and Drug Analysis*, *26*(1), 232–243. https://doi.org/10.1016/j.jfda.2017.03.002

Azzali, A., D'Agostino, S., & Fabrizia Grepioni. (2021). Tuning the Solubility of the Herbicide Bentazon: from Salt to Neutral and to Inclusion Complexes. *ACS Sustainable Chem. Eng.*, *9*(37), 12530–12539. https://doi.org/10.1021/acssuschemeng.1c02749

Bezerra, F. M., Lis, M. J., Firmino, H. B., Dias da Silva, J. G., Curto Valle, R. de C. S., Borges Valle, J. A., Scacchetti, F. A. P., & Tessaro, A. L. (2020). The Role of β-Cyclodextrin in the Textile Industry—Review. *Molecules*, *25*(16), 3624. https://doi.org/10.3390/molecules25163624

Bezerra, J. N., Gomez, M. C. V., Rolón, M., Coronel, C., Almeida-Bezerra, J. W., Fidelis, K. R., Menezes, S. A. de, Cruz, R. P. da, Duarte, A. E., Ribeiro, P. R. V., Brito, E. S. de, Coutinho, H. D. M., Morais-Braga, M. F. B., & Bezerra, C. F. (2022). Chemical composition, Evaluation of Antiparasitary and Cytotoxic Activity of the essential oil of Psidium brownianum MART EX. DC. *Biocatalysis and Agricultural Biotechnology*, *39*, 102247. https://doi.org/10.1016/j.bcab.2021.102247

Brewster, M. E., & Loftsson, T. (2007). Cyclodextrins as pharmaceutical solubilizers. *Advanced Drug Delivery Reviews*, *59*(7), 645–666. https://doi.org/10.1016/j.addr.2007.05.012

Buchbauer, G. (2000). The Detailed Analysis of Essential Oils Leads to the Understanding of their Properties a. *Chemical Weekly-Bombay*, *45*(49), 163–165.

Cabral-Marques, H. (1994). Structure and properties of cyclodextrins: inclusion complex formation. *Rev. Port. Farm*, *44*, 2.

Campolo, O., Giunti, G., Laigle, M., Michel, T., & Palmeri, V. (2020). Essential oil-based nano-emulsions: Effect of different surfactants, sonication and plant species on physicochemical characteristics. *Industrial Crops and Products*, *157*, 112935. https://doi.org/10.1016/j.indcrop.2020.112935

Chebbac, K., Ghneim, H. K., El Moussaoui, A., Bourhia, M., El Barnossi, A., Benziane Ouaritini, Z., Salamatullah, A. M., Alzahrani, A., Aboul-Soud, M. A. M., Giesy, J. P., & Guemmouh, R. (2022). Antioxidant and Antimicrobial Activities of Chemically-Characterized Essential Oil from Artemisia aragonensis Lam. against Drug-Resistant Microbes. *Molecules*, *27*(3), 1136. https://doi.org/10.3390/molecules27031136

Chen, Y., Luo, J., Zhang, N., Yu, W., Jiang, J., & Dai, G. (2021). Insecticidal activities of Salvia hispanica L. essential oil and combinations of their main compounds against the beet armyworm Spodoptera exigua. *Industrial Crops and Products*, *162*, 113271. https://doi.org/10.1016/j.indcrop.2021.113271

Chu, Y., Gao, C., Liu, X., Zhang, N., Xu, T., Feng, X., Yang, Y., Shen, X., & Tang, X. (2020). Improvement of storage quality of strawberries by pullulan coatings incorporated with cinnamon essential oil nanoemulsion. *LWT*, *122*, 109054. https://doi.org/10.1016/j.lwt.2020.109054

Chuesiang, P., Sanguandeekul, R., & Siripatrawan, U. (2021). Enhancing effect of nanoemulsion on antimicrobial activity of cinnamon essential oil against foodborne pathogens in refrigerated Asian seabass (Lates calcarifer) fillets. *Food Control*, *122*, 107782. https://doi.org/10.1016/j.foodcont.2020.107782

Coimbra, A., Ferreira, S., & Duarte, A. P. (2022). Biological properties of Thymus zygis essential oil with emphasis on antimicrobial activity and food application. *Food Chemistry*, *393*, 133370. https://doi.org/10.1016/j.foodchem.2022.133370

Crini, G., & Aleya, L. (2021). Cyclodextrin applications in pharmacy, biology, medicine, and environment. *Environmental Science and Pollution Research*. https://doi.org/10.1007/s11356-021-16871-2

Cui, H., Siva, S., & Lin, L. (2019). Ultrasound processed cuminaldehyde/2-hydroxypropyl-β-cyclodextrin inclusion complex: Preparation, characterization and antibacterial activity. *Ultrasonics Sonochemistry*, *56*, 84–93. https://doi.org/10.1016/j.ultsonch.2019.04.001

Dayan, F. E., Cantrell, C. L., & Duke, S. O. (2009). Natural products in crop protection. *Bioorganic & Medicinal Chemistry*, *17*(12), 4022–4034. https://doi.org/10.1016/j.bmc.2009.01.046

Dayan, F. E., & Duke, S. O. (2014). Natural Compounds as Next-Generation Herbicides. *Plant Physiology*, *166*(3), 1090–1105. https://doi.org/10.1104/pp.114.239061

De Almeida, L. F. R., Frei, F., Mancini, E., De Martino, L., & De Feo, V. (2010). Phytotoxic Activities of Mediterranean Essential Oils. *Molecules*, *15*(6), 4309–4323. https://doi.org/10.3390/molecules15064309

De Lima, J. R., Santos, D., & De Santana, D. (2012). Complexos de inclusão como estratégia para veiculação de óleos essenciais. *Revista Brasileira de Farmácia*, *93*, 4.

Del Valle, E. M. Martin. (2004). Cyclodextrins and their uses: a review. *Process Biochemistry*, *39*(9), 1033–1046. https://doi.org/10.1016/s0032-9592(03)00258-9

Devarajan, V., & Ravichandran, V. (2011). Nanoemulsions: As modified drug delivery tool. *International Journal of Comprehensive Pharmacy*, *2*, 1–6.

Dhifi, W., Bellili, S., Jazi, S., Bahloul, N., & Mnif, W. (2016). Essential oils' chemical characterization and investigation of some biological activities: A critical review. *Medicines*, *3*(4), 25. https://doi.org/10.3390/medicines3040025

El-Alam, I., Raveau, R., Fontaine, J., Verdin, A., Laruelle, F., Fourmentin, S., Chahine, R., Makhlouf, H., & Lounès-Hadj Sahraoui, A. (2020). Antifungal and Phytotoxic Activities of Essential Oils: In Vitro Assays and Their Potential Use in Crop Protection. *Agronomy*, *10*(6), 825. https://doi.org/10.3390/agronomy10060825

Flores, F. C., Ribeiro, R. F., Ourique, A. F., Rolim, C. M. B., Silva, C. de B. da, Pohlmann, A. R., Beck, R. C. R., & Guterres, S. S. (2011). Nanostructured systems containing an essential oil: protection against volatilization. *Química Nova*, *34*, 968–972. https://doi.org/10.1590/S0100-40422011000600010

Ganta, S., Talekar, M., Singh, A., Coleman, T. P., & Amiji, M. M. (2014). Nanoemulsions in Translational Research—Opportunities and Challenges in Targeted Cancer Therapy. *AAPS PharmSciTech*, *15*(3), 694–708. https://doi.org/10.1208/s12249-014-0088-9

Gao, S., Jiang, J., Li, X., Liu, Y., Zhao, L., Fu, Y., & Ye, F. (2020). Enhanced physicochemical properties and herbicidal activity of an environment-friendly clathrate formed by β-cyclodextrin and herbicide cyanazine. *Journal of Molecular Liquids*, *305*, 112858. https://doi.org/10.1016/j.molliq.2020.112858

Gao, S., Jiang, J.-Y., Liu, Y.-Y., Fu, Y., Zhao, L.-X., Li, C.-Y., & Ye, F. (2019). Enhanced Solubility, Stability, and Herbicidal Activity of the Herbicide Diuron by Complex Formation with β-Cyclodextrin. *Polymers*, *11*(9), 1396. https://doi.org/10.3390/polym11091396

Gershenzon, J., & Dudareva, N. (2007). The function of terpene natural products in the natural world. *Nature Chemical Biology*, *3*(7), 408–414. https://doi.org/10.1038/nchembio.2007.5

Gharsallaoui, A., Roudaut, G., Chambin, O., Voilley, A., & Saurel, R. (2007). Applications of spray-drying in microencapsulation of food ingredients: An overview. *Food Research International*, *40*(9), 1107–1121. https://doi.org/10.1016/j.foodres.2007.07.004

Gupta, A. (2019). Chapter 21 - Nanoemulsions. In E. J. Chung, L. Leon, & C. Rinaldi (Eds.), *Nanoparticles for Biomedical Applications: Fundamental Concepts, Biological Interactions and Clinical Applications* (pp. 371–384). Elsevier. https://www.sciencedirect.com/science/article/abs/pii/B9780128166628000217

Gupta, A., Eral, H. B., Hatton, T. A., & Doyle, P. S. (2016). Nanoemulsions: formation, properties and applications. *Soft Matter*, *12*(11), 2826–2841. https://doi.org/10.1039/c5sm02958a

Håkansson, A., & Rayner, M. (2018). General Principles of Nanoemulsion Formation by High-Energy Mechanical Methods. In S. M. Jafari & D. J. McClements (Eds.), *Nanoemulsions: Formulation, Applications, and Characterization* (pp. 103–139). Academic Press,. https://doi.org/10.1016/b978-0-12-811838-2.00005-9

Han, C., Shao, H., Zhou, S., Mei, Y., Cheng, Z., Huang, L., & Lv, G. (2021). Chemical composition and phytotoxicity of essential oil from invasive plant, Ambrosia artemisiifolia L. *Ecotoxicology and Environmental Safety*, *211*, 111879. https://doi.org/10.1016/j.ecoenv.2020.111879

Hanif, M. A., Nisar, S., Khan, G. S., Mushtaq, Z., & Zubair, M. (2019). Essential Oils. *Essential Oil Research*, 3–17. https://doi.org/10.1007/978-3-030-16546-8_1

Hazrati, H., Saharkhiz, M. J., Niakousari, M., & Moein, M. (2017). Natural herbicide activity of Satureja hortensis L. essential oil nanoemulsion on the seed germination and morphophysiological features of two important weed species. *Ecotoxicology and Environmental Safety*, *142*, 423–430. https://doi.org/10.1016/j.ecoenv.2017.04.041

Hou, K., Xu, Y., Cen, K., Gao, C., Feng, X., & Tang, X. (2021). Nanoemulsion of cinnamon essential oil Co-emulsified with hydroxypropyl-β-cyclodextrin and Tween-80: Antibacterial activity, stability and slow release performance. *Food Bioscience*, *43*, 101232. https://doi.org/10.1016/j.fbio.2021.101232

Ibáñez, M. D., & Blázquez, M. A. (2020). Phytotoxic effects of commercial essential oils on selected vegetable crops: Cucumber and tomato. *Sustainable Chemistry and Pharmacy*, *15*, 100209. https://doi.org/10.1016/j.scp.2019.100209

Isman, M. B. (2015). A renaissance for botanical insecticides? *Pest Management Science*, *71*(12), 1587–1590. https://doi.org/10.1002/ps.4088

Issa, M., Chandel, S., Pal Singh, H., Rani Batish, D., Kumar Kohli, R., Singh Yadav, S., & Kumari, A. (2020). Appraisal of phytotoxic, cytotoxic and genotoxic potential of essential oil of a medicinal plant Vitex negundo. *Industrial Crops and Products*, *145*, 112083. https://doi.org/10.1016/j.indcrop.2019.112083

Jafari , S. M., Paximada, P., Mandala, I., Assadpour, E., & Mehrnia, M. A. (2017). 2 - Encapsulation by nanoemulsions. In *Nanoencapsulation technologies for the food and nutraceutical industries* (pp. 36–73). Academic Press. https://doi.org/10.1016/B978-0-12-809436-5.00002-1

Jansook, P., Ogawa, N., & Loftsson, T. (2018). Cyclodextrins: structure, physicochemical properties and pharmaceutical applications. *International Journal of Pharmaceutics*, *535*(1-2), 272–284. https://doi.org/10.1016/j.ijpharm.2017.11.018

Jiang, C., Zhou, S., Liu, L., Toshmatov, Z., Huang, L., Shi, K., Zhang, C., & Shao, H. (2021a). Evaluation of the phytotoxic effect of the essential oil from Artemisia absinthium. *Ecotoxicology and Environmental Safety*, *226*, 112856. https://doi.org/10.1016/j.ecoenv.2021.112856

Jiang, S., Zhao, T., Wei, Y., Cao, Z., Xu, Y., Wei, J., Xu, F., Wang, H., & Shao, X. (2021b). Preparation and characterization of tea tree oil/ hydroxypropyl-β-cyclodextrin inclusion complex and its application to control brown rot in peach fruit. *Food Hydrocolloids*, *121*, 107037. https://doi.org/10.1016/j.foodhyd.2021.107037

Kaur, P., Gupta, S., Kaur, K., Kaur, N., Kumar, R., & Bhullar, M. S. (2021). Nanoemulsion of Foeniculum vulgare essential oil: A propitious striver against weeds of Triticum aestivum. *Industrial Crops and Products*, *168*, 113601. https://doi.org/10.1016/j.indcrop.2021.113601

Komaiko, J. S., & 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. https://doi.org/10.1111/1541-4337.12189

Kreutz, T., Carneiro, S. B., Soares, K. D., Limberger, R. P., Apel, M. A., Veiga-Junior, V. F., & Koester, L. S. (2021). Aniba canelilla (Kunth) Mez essential oil-loaded nanoemulsion: Improved stability of the main constituents and in vitro antichemotactic activity. *Industrial Crops and Products*, *171*, 113949. https://doi.org/10.1016/j.indcrop.2021.113949

Krzyżowski, M., Baran, B., Łozowski, B., & Francikowski, J. (2020). The role of dilution mediums in studies of fumigant insecticidal activity of essential oils. *Journal of Pest Science*, *93*(4), 1119–1124. https://doi.org/10.1007/s10340-020-01241-7

Lu, Y., Qi, J., & Wu, W. (2012). Absorption, Disposition and Pharmacokinetics of Nanoemulsions. *Current Drug Metabolism*, *13*(4), 396–417. https://doi.org/10.2174/138920012800166544

Luo, D.-Y., Yan, Z.-T., Che, L.-R., Zhu, J. J., & Chen, B. (2022). Repellency and insecticidal activity of seven Mugwort (Artemisia argyi) essential oils against the malaria vector Anopheles sinensis. *Scientific Reports*, *12*(1). https://doi.org/10.1038/s41598-022-09190-0

Maa, Y.-F., & Hsu, C. C. (1999). Performance of Sonication and Microfluidization for Liquid–Liquid Emulsification. *Pharmaceutical Development and Technology*, *4*(2), 233–240. https://doi.org/10.1081/pdt-100101357

MacNicol, D. D., McKendrick, J. J., & Wilson, D. R. (1978). Clathrates and molecular inclusion phenomena. *Chemical Society Reviews*, *7*(1), 65–87. https://doi.org/10.1039/CS9780700065

Mahanta, B. P., Kemprai, P., Bora, P. K., Lal, M., & Haldar, S. (2022). Phytotoxic essential oil from black turmeric (Curcuma caesia Roxb.) rhizome: Screening, efficacy, chemical basis, uptake and mode of transport. *Industrial Crops and Products*, *180*, 114788. https://doi.org/10.1016/j.indcrop.2022.114788

Mansouri, S., Pajohi-Alamoti, M., Aghajani, N., Bazargani-Gilani, B., & Nourian, A. (2020). Stability and antibacterial activity of

Thymus daenensis

L. essential oil nanoemulsion in mayonnaise. *Journal of the Science of Food and Agriculture*, *101*(9), 3880–3888. https://doi.org/10.1002/jsfa.11026

Marques, H. M. C. (2010). A review on cyclodextrin encapsulation of essential oils and volatiles. *Flavour and Fragrance Journal*, *25*(5), 313–326. https://doi.org/10.1002/ffj.2019

Mason, T. G., Wilking, J. N., Meleson, K., Chang, C. B., & Graves, S. M. (2007). Nanoemulsions: formation, structure, and physical properties. *Journal of Physics: Condensed Matter*, *19*(7), 079001. https://doi.org/10.1088/0953-8984/19/7/079001

McClements, D. J. (2012). Nanoemulsions versus microemulsions: terminology, differences, and similarities. *Soft Matter*, *8*(6), 1719–1729. https://doi.org/10.1039/c2sm06903b

Moghimi, R., Ghaderi, L., Rafati, H., Aliahmadi, A., & McClements, D. J. (2016). Superior antibacterial activity of nanoemulsion of Thymus daenensis essential oil against E. coli. *Food Chemistry*, *194*, 410–415. https://doi.org/10.1016/j.foodchem.2015.07.139

Moumni, M., Romanazzi, G., Najar, B., Pistelli, L., Ben Amara, H., Mezrioui, K., Karous, O., Chaieb, I., & Allagui, M. B. (2021). Antifungal Activity and Chemical Composition of Seven Essential Oils to

Control the Main Seedborne Fungi of Cucurbits. *Antibiotics*, *10*(2), 104. https://doi.org/10.3390/antibiotics10020104

Osanloo, M., Amani, A., Sereshti, H., Abai, M. R., Esmaeili, F., & Sedaghat, M. M. (2017). Preparation and optimization nanoemulsion of Tarragon (Artemisia dracunculus) essential oil as effective herbal larvicide against Anopheles stephensi. *Industrial Crops and Products*, *109*, 214–219. https://doi.org/10.1016/j.indcrop.2017.08.037

Osanloo, M., Hassan Sereshti, H., Sedaghat, M. M., & Amani, A. (2018). Nanoemulsion of Dill essential oil as a green and potent larvicide against Anopheles stephensi. *Environmental Science and Pollution Research*, *25*(7), 6466–6473. https://doi.org/10.1007/s11356-017-0822-4

Pavela, R., & Benelli, G. (2016). Essential Oils as Ecofriendly Biopesticides? Challenges and Constraints. *Trends in Plant Science*, *21*(12), 1000–1007. https://doi.org/10.1016/j.tplants.2016.10.005

Pavoni, L., Perinelli, D. R., Bonacucina, G., Cespi, M., & Palmieri, G. F. (2020). An Overview of Micro- and Nanoemulsions as Vehicles for Essential Oils: Formulation, Preparation and Stability. *Nanomaterials*, *10*(1), 135. https://doi.org/10.3390/nano10010135

Pouresmaeil, M., Sabzi-Nojadeh, M., Movafeghi, A., Aghbash, B. N., Kosari-Nasab, M., Zengin, G., & Maggi, F. (2022). Phytotoxic activity of Moldavian dragonhead (Dracocephalum moldavica L.) essential oil and its possible use as bio-herbicide. *Process Biochemistry*, *114*, 86–92. https://doi.org/10.1016/j.procbio.2022.01.018

Roana, J., Mandras, N., Scalas, D., Campagna, P., & Tullio, V. (2021). Antifungal Activity of Melaleuca alternifolia Essential Oil (TTO) and Its Synergy with Itraconazole or Ketoconazole against Trichophyton rubrum. *Molecules*, *26*(2), 461. https://doi.org/10.3390/molecules26020461

Rodríguez, J., Martín, M. J., Ruiz, M. A., & Clares, B. (2016). Current encapsulation strategies for bioactive oils: From alimentary to pharmaceutical perspectives. *Food Research International*, *83*, 41–59. https://doi.org/10.1016/j.foodres.2016.01.032

Rys, M., Miastkowska, M., Sikora, E., Łętocha, A., Krajewska, A., & Synowiec, A. (2022). Bio-Herbicidal Potential of Nanoemulsions with Peppermint Oil on Barnyard Grass and Maize. *Molecules*, *27*(11), 3480. https://doi.org/10.3390/molecules27113480

Salgueiro, L., Martins, A. P., & Correia, H. (2010). Raw materials: the importance of quality and safety. A review. *Flavour and Fragrance Journal*, *25*(5), 253–271. https://doi.org/10.1002/ffj.1973

Sanguansri, L., Oliver, C. M., & Leal-Calderon, F. (2013). Nanoemulsion technology for delivery of nutraceuticals and functional-food ingredients. In *Bio-nanotechnology: a revolution in food, biomedical and health sciences* (pp. 667–696). John Wiley & Sons.

Santos, S. M. dos, de Oliveira Junior, P. C., de Matos Balsalobre, N., Kassuya, C. A. L., Cardoso, C. A. L., Pereira, Z. V., Silva, R. M. M. F., & Formagio, A. S. N. (2021). Variation in essential oil components and anti-inflammatory activity of Allophylus edulis leaves collected in central-western Brazil. *Journal of Ethnopharmacology*, *267*, 113495. https://doi.org/10.1016/j.jep.2020.113495

Schmitt, C., & Turgeon, S. L. (2011). Protein/polysaccharide complexes and coacervates in food systems. *Advances in Colloid and Interface Science*, *167*(1-2), 63–70.

https://doi.org/10.1016/j.cis.2010.10.001

Schubert, M. A., & Müller-Goymann, C. C. (2003). Solvent injection as a new approach for manufacturing lipid nanoparticles – evaluation of the method and process parameters. *European Journal of Pharmaceutics and Biopharmaceutics*, *55*(1), 125–131. https://doi.org/10.1016/S0939-6411(02)00130-3

Seibert, J. B., Rodrigues, I. V., Carneiro, S. P., Amparo, T. R., Lanza, J. S., Jean, Henrique, G., & Orlando, S. (2019). Seasonality study of essential oil from leaves of Cymbopogon densiflorus and nanoemulsion development with antioxidant activity. *Flavour and Fragrance Journal*, *34*, 1.

Sharma, N., Bansal, M., Visht, S., Sharma, P., & Kulkarni, G. (2010). Nanoemulsion: A new concept of delivery system. *Chronicles of Young Scientists*, *1*, 2.

Silva, A. F. R., Monteiro, M., Resende, D., Braga, S. S., Coimbra, M. A., Silva, A. M. S., & Cardoso, S. M. (2020). Inclusion Complex of Resveratrol with γ-Cyclodextrin as a Functional Ingredient for Lemon Juices. *Foods*, *10*(1), 16. https://doi.org/10.3390/foods10010016

Singh, N., Singh, H. P., Batish, D. R., Kohli, R. K., & Yadav, S. S. (2020). Chemical characterization, phytotoxic, and cytotoxic activities of essential oil of Mentha longifolia. *Environmental Science and Pollution Research*, *27*(12), 13512–13523. https://doi.org/10.1007/s11356-020-07823-3

Sobrinho, A. C. N., de Morais, S. M., Marinho, M. M., de Souza, N. V., & Lima, D. M. (2021). Antiviral activity on the Zika virus and larvicidal activity on the Aedes spp. of Lippia alba essential oil and β-caryophyllene. *Industrial Crops and Products*, *162*, 113281. https://doi.org/10.1016/j.indcrop.2021.113281

Solans, C., & Solé, I. (2012). Nano-emulsions: Formation by low-energy methods. *Current Opinion in Colloid & Interface Science*, *17*(5), 246–254. https://doi.org/10.1016/j.cocis.2012.07.003

Suganya, V., & Anuradha, V. (2017). Microencapsulation and nanoencapsulation: a review. *Int. J. Pharm. Clin. Res*, *9*, 3.

Svoboda, K. P., & Svoboda, T. G. (2000). *Secretory structures of aromatic and medicinal plants: a review and atlas of micrographs* (P. M. Syred, Ed.). CABI Direct. https://www.cabdirect.org/cabdirect/abstract/20000315078

Szejtli, J., & Osa, T. (1996). *Comprehensive supramolecular chemistry, vol. 3, pargamon*. Elsevier, Oxford.

Szente, L., & Szejtli, J. (1999). Highly soluble cyclodextrin derivatives: chemistry, properties, and trends in development. *Advanced Drug Delivery Reviews*, *36*(1), 17–28. https://doi.org/10.1016/s0169-409x(98)00092-1

Taiz, L., & Zeiger, E. (2010). *Plant Physiology* (5th ed.). Sinauer Associates Inc.

Tayeb, H. H., & Sainsbury, F. (2018). Nanoemulsions in drug delivery: formulation to medical application. *Nanomedicine*, *13*(19), 2507–2525. https://doi.org/10.2217/nnm-2018-0088

Turek, C., & Stintzing, F. C. (2013). Stability of Essential Oils: A Review. *Comprehensive Reviews in Food Science and Food Safety*, *12*(1), 40–53. https://doi.org/10.1111/1541-4337.12006

Verdeguer, M., Sánchez-Moreiras, A. M., & Araniti, F. (2020). Phytotoxic Effects and Mechanism of Action of Essential Oils and Terpenoids. *Plants*, *9*(11), 1571. https://doi.org/10.3390/plants9111571

Wadhwa, G., Kumar, S., Chhabra, L., Mahant, S., & Rao, R. (2017). Essential oil–cyclodextrin complexes: an updated review. *Journal of Inclusion Phenomena and Macrocyclic Chemistry*, *89*(1-2), 39–58. https://doi.org/10.1007/s10847-017-0744-2

Yáñez, C., Cañete-Rosales, P., Castillo, J. P., Catalán, N., Undabeytia, T., & Morillo, E. (2012). Cyclodextrin Inclusion Complex to Improve Physicochemical Properties of Herbicide Bentazon: Exploring Better Formulations. *PLoS ONE*, *7*(8), e41072. https://doi.org/10.1371/journal.pone.0041072

Yuan, C., Thomas, D. S., Hook, J. M., Qin, G., Qi, K., & Zhao, J. (2019). Molecular Encapsulation of Eucalyptus staigeriana Essential Oil by Forming Inclusion Complexes with Hydroxypropyl-β-Cyclodextrin. *Food and Bioprocess Technology*, *12*(8), 1264–1272. https://doi.org/10.1007/s11947-019-02291-1

Yuan, C., Wang, Y., Liu, Y., & Cui, B. (2019). Physicochemical characterization and antibacterial activity assessment of lavender essential oil encapsulated in hydroxypropyl-beta-cyclodextrin. *Industrial Crops and Products*, *130*, 104–110. https://doi.org/10.1016/j.indcrop.2018.12.067

Zhang, G., Yuan, C., & Sun, Y. (2018). Effect of Selective Encapsulation of Hydroxypropyl-β-cyclodextrin on Components and Antibacterial Properties of Star Anise Essential Oil. *Molecules*, *23*(5), 1126. https://doi.org/10.3390/molecules23051126

Zhou, S., Han, C., Zhang, C., Kuchkarova, N., Wei, C., Zhang, C., & Shao, H. (2021). Allelopathic, Phytotoxic, and Insecticidal Effects of Thymus proximus Serg. Essential Oil and Its Major Constituents. *Frontiers in Plant Science*, *12*. https://doi.org/10.3389/fpls.2021.689875

Zimmermann, R. C., Aragão, C. E. de C., Araújo, P. J. P. de, Benatto, A., Chaaban, A., Martins, C. E. N., Amaral, W. do, Cipriano, R. R., & Zawadneak, M. A. C. (2021). Insecticide activity and toxicity of essential oils against two stored-product insects. *Crop Protection*, *144*, 105575. https://doi.org/10.1016/j.cropro.2021.105575

Zuidam, N. J., & Shimoni, E. (2009). Overview of Microencapsulates for Use in Food Products or Processes and Methods to Make Them. *Encapsulation Technologies for Active Food Ingredients and Food Processing*, 3–29. https://doi.org/10.1007/978-1-4419-1008-0_2

Zuzarte, M., & Salgueiro, L. (2015). Essential Oils Chemistry. *Bioactive Essential Oils and Cancer*, 19–61. https://doi.org/10.1007/978-3-319-19144-7_2