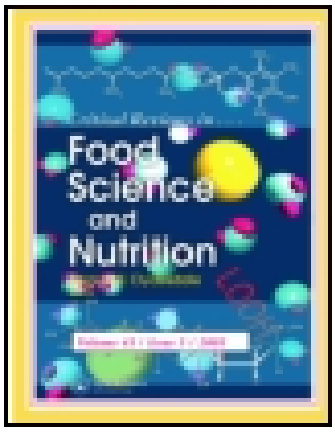

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Encapsulation of vegetable oils as source of omega-3 fatty acids for enriched functional foods

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Abstract

Polyunsaturated omega-3 fatty acids (PUFAs), a functional component present in vegetable oils, are generally recognized as being beneficial to health. Omega-3 PUFAs are rich in double bonds and unsaturated in nature; this attribute makes them highly susceptible to lipid oxidation and unfit for incorporation into long-shelf life foods. The microencapsulation of oils in a polymeric matrix (mainly polysaccharides) offers the possibility of controlled release of the lipophilic functional ingredient and can be useful for the supplementation of foods with PUFAs. The present article paper provides a literature review of different vegetable sources of omega-3 fatty acids**,** the functional effects of omega-3 fatty acids, different microencapsulation methods that can possibly be used for the encapsulation of oils, the properties of vegetable oil microcapsules,

the effect of encapsulation on oxidation stability and fatty acid composition of vegetable oils, and the incorporation of long-chain omega-3 polyunsaturated fatty acids in foods.

Keywords

encapsulation, vegetable oils, omega 3 fatty acids, functional food.

Introduction

Many components naturally present in vegetable oils have been shown to have beneficial properties. Once isolated and concentrated, a number of these compounds have proven effective in treating a wide range of conditions ranging from irritable bowel syndrome to chronic liver disease. Similarly, many of the fatty acids and other compounds present in vegetable oils have long been known to benefit our health. There is clearly great potential for developing functional vegetable oils (Riechart, 2002). The number of active ingredients so far identified in oil seeds is impressive. Many of these compounds make it through to the final salad or cooking oil whilst others may be partially or wholly removed during the oil-refining process. Vitamin E is a powerful antioxidant and vegetable oils are a major dietary source of this vitamin. Each fatty acid also has its own specific properties (De Deckere and Verschuren, 2000). Linoleic acid is a polyunsaturated fatty acid with cholesterol-lowering properties and a-linolenic acid is also linked to heart health. Ricinoleic acid is the active ingredient in castor oil and is a powerful stimulant laxative, whilst γ-linolenic acid provides the main benefits of evening primrose oil, used, among other things, to treat breast pain and atopic eczema. Phytosterols are found in vegetable oils, particularly germ oils. Margarines fortified with sterols have recently hit the headlines because their cholesterol-lowering capacity is as effective as many drugs (Law, 2000).

It is now also suggested that natural levels of phytosterols found in many vegetable oils (maize oil: 968 mg/100g, wheat germ oil: 553 mg/100g and olive oil: 221 mg/100g) may also make a significant contribution to cholesterol lowering (Ostlund et al., 2002). Plenty of other beneficial compounds are also extracted and concentrated from byproducts of the refining process,

including ß-carotene, Vitamin K, phosphatidylcholine, which is used in the treatment of liver conditions, and phosphatidylserine, which is mainly used to prevent brain deterioration (Riechart, 2002). Since many compounds in oil seeds have already proven nutritional benefits, there are great possibilities for using them to develop new functional vegetable oils. Vegetable oils containing enhanced levels of beneficial active ingredients could have a substantial impact on human health considering the amount of cooking and salad oils consumed in most industrialized countries (Riechart, 2002).

The incorporation of functional ingredients in a given food system and the processing and handling of such foods are associated with nutritional challenges for their healthy delivery. The extreme sensitivity of oils to oxidation can easily lead to the development of off-flavors and cause significant loss of product quality, stability, nutritional value and bioavailability, and the overall acceptability of the food product. Consequently, microencapsulation has been successfully used to encapsulate oils in order to prevent oxidation and to improve stability and bioavailability (Wakil et al., 2010). Microencapsulation is one example of technology that has the potential to meet the challenge of successfully incorporating and delivering functional ingredients into a range of food types.

Omega-3 fatty acid sources and its components

Omega-3 fatty acids (also known as n-3 fatty acids) are a group of naturally occurring lipids, which are present in high concentrations in certain fishes and plants (Cunnane et al., 1995). The term omega-3 (n-3, ω-3) signifies that the first double bond exists at the third carbon-carbon bond from the terminal methyl end (ω) of the carbon chain. There are three most common

omega-3 fatty acids: alpha linolenic acid (ALA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). ALA is present in certain vegetable oils (flaxseed and canola), whereas EPA and DHA are present in fish and in more concentrated amounts in fish oil (Mantzioris et al., 2000). They are found in varying ratios in fish, particularly in cold-water and pelagic species such as menhaden, mackerel, oil sardine and salmon (Stensby, 1969). The origin of the omega-3 fatty acids found in these species of fishes is the chloroplasts of marine phytoplankton and algae (Stamey, 2010; Cohen, 1995). Fish consumption and omega-3 supplementation have attracted considerable interest in the past few decades in relation to their health benefits. Fish oils provide a source of EPA and DHA, two fatty acids now recognized as an important part of the human diet (Calder, 2006). EPA and DHA are highly unsaturated fatty acids synthesized from ALA and other fatty acids in the omega-3 pathway (Figure 1).

The major sources of these omega-3 fatty acids are oily fish species including salmon, mackerel and herring (Strobel et al., 2012). Fisheries are currently producing the maximum fish stocks per annum in order to supply fish for human consumption, as well as supplying feed for industrial fish farms and fish oil supplements, resulting in a substantial effect on fish levels and the possibility of extinction (Dulvy et al., 2003). However, an expansive literature indicates that omega-3 fish oils are crucial dietary components. In order to protect fish species and the oceans' ecosystems, alternative sources of long-chain polyunsaturated fatty acids are required. Currently explored alternatives include plant oils with high omega-3 content, the use of stearidonic acid and algae oils (Lenihan-Geels et al., 2013).

Over thousands of years of an agriculture-based existence, the dietary ratio n-3:n-6 remained stable at about l. However, in the past 100 years changes in the food supply and dietary habits have caused this ratio to fall dramatically to less than 0.1. Within a range of total caloric intake of 2000–2500 Kcal, a proper safety ratio of oleic acid (OA):linoleic acid (LA):ALA could be expressed as 11–16:4–6:1. Recent data show that a diet including about 13% of OA in the total caloric intake could provide protection against the occurrence of new cardiovascular events, but an increase of OA intake to more than 20% could limit this beneficial intake by inducing an increase of low-density lipoprotein in blood (Stuchlík and Žák, 2002). Today this consumption ratio is about 10–20:1, indicating a deficiency in n-3 polyunsaturated fatty acids (PUFAs) compared with the diet on which humans evolved and their genetic patterns were established. The n-3 and n-6 fatty acids are not interconvertible in the human body, and therefore, appropriate amounts of both acids need to be considered in making dietary recommendations. The food industry is already taking steps to return n-3 PUFAs to the food supply by enriching various products with safety sources of n-3 fatty acids (Simopoulos et al., 1999). The richest vegetable sources of the n-3 PUFA series, represented by LNA (Table 1), seem to be *Dracocephalum moldavica*, *Linum usitatissimum*, *Perilla frutescens*, *Salvia hispanica* and *Aleurites moluccana* (Stuchlík and Žák, 2002).

Vegetables oils as obtained from *Dracocephalum moldavica* contain 61% of linolenic acid (Dziki et al., 2013). *Linum usitatissimum* oil contains a high proportion of unsaturated fatty acids (88.97%) with 11.01% of saturated fatty acids; predominantly in the composition of unsaturated fatty acids is linolenic acid (55-65%) (Popa et al., 2012). The major fatty acids of the *Perilla frutescens* oil are linolenic (58.0%), linoleic (14.0%) and oleic acids (19.0%) (Shin and Kim,

1994). *Salvia hispanica* oil contains a high proportion of unsaturated fatty acids (89.29%) with 10.71% of saturated fatty acids; predominantly in the composition of unsaturated fatty acids is linolenic acid (51.0%) (Segura-Campos et al., 2014). *Aleurites moluccana* oil contains a high proportion of unsaturated fatty acids like linolenic (30%), oleic (28%) and linoleic (40%) (Stuchlík and Žák, 2002).

The *Dracocephalum moldavica* plant is used in particular for its content of essential oil in all above-ground parts; however, the seeds with about 15% of vegetable oil contain 61% of LNA. Vegetable LNA thus serves as the higher member precursor of the n-3 series of fatty acids well known from fish oils. For example, LNA can be converted in humans to EPA or DHA. Omega-3 unsaturated fatty acids are nutritionally important for good health and are especially beneficial for individuals suffering from coronary heart disease, diabetes and immune response disorders (Djordjevic et al., 2004; McClements et al., 2007). An adequate ratio or balance between ω-6 and ω-3 FAs is important for the prevention and treatment of cardiovascular diseases. For the secondary prevention of cardiovascular disease, a ratio of 4:1 has been associated with a 70% reduction in total mortality (Gómez-Candela et al., 2011).

Functional effects of omega-3 fatty acids

Functional foods must be safe according to all standards assessing food risk. However, the concept of risk versus benefit cannot be applied in such a straightforward manner as it is for drugs. Finally, long-term consequences of interactions between functional food components and functions in a body and interactions between components must be carefully monitored (Diplock et al., 1999). The major problems associated with functional foods concern intake and effect. In

order to determine that a food has an effect beyond its nutritional attributes it is necessary to confirm that the target population is exposed to food that contains a putative active agent and that this particular component possesses a desired effect. Since the diet of most adults is complex and varied, the correlation between distinct effects and specific dietary constituents is difficult to confirm in free-living individuals (Southon, 2000).

More recent studies assessed that omega-3 polyunsaturated fatty acid supplementation could be helpful against many inflammatory diseases. The key link between polyunsaturated fatty acids (PUFAs) and inflammation is that eicosanoids, which are among the mediators and regulators of inflammation, are generated from 20-carbon PUFAs. Because inflammatory cells typically contain a high proportion of the n-6 PUFA arachidonic acid (20:4n-6) and low proportions of other 20-carbon PUFAs, arachidonic acid is usually the major substrate for eicosanoid synthesis. Eicosanoids, which include PGs, thromboxanes, leukotrienes (LTs) and other oxidized derivatives, are generated from arachidonic acid by the metabolic processes summarized in Figure 2 (Calder, 2006).

Although their action in antagonizing arachidonic acid metabolism is a key antiinflammatory effect of n-3 PUFAs, these fatty acids have several other antiinflammatory effects that might result from altered eicosanoid production or might be independent of this. For example, studies have shown that, when consumed in sufficient quantities, dietary fish oil results in decreased leukocyte chemotaxis, decreased production of reactive oxygen species and proinflammatory cytokines, and decreased adhesion molecule expression (Table 2) (Calder, 2001).

Methods for oil encapsulation

One way of protecting or delivering oils is by encapsulating them in a matrix that acts as a barrier (Ye et al., 2009). The encapsulation of oils in the form of oil-core capsules is used in agriculture, pharmaceuticals, foods, cosmetics and fragrances, and many other industries. Commonly, oils are encapsulated for controlled release, masking off-flavors or colors, increasing shelf life stability, evaporation and incompatibilities with reactive substances, easy handling by converting sticky oils to free-flowing particles, and for easy storage of dry particles compared to frozen oils (Abang et al., 2012). Microencapsulation of omega-3 oils minimizes oxidative deterioration and allows their use in stable and easy-to-handle form (Kaushik et al, 2014). Oils can be encapsulated in two structures: beads or capsules. A capsule consists of a well-defined core and an envelope, whilst a bead is made of a continuous phase of one or more miscible polymers in which encapsulants are dispersed (Mathiowitz et al., 1999).

Different encapsulation methods are used to produce these two structures. Oils have been entrapped in beads using spray-drying and freeze-drying (Dzondo-Gaget et al., 2005; Jafari et al., 2008), emulsification/internal gelation (Ribeiro et al., 1999), cocrystallization (Beristain et al., 1996), emulsion extrusion (Yilmaz et al., 2001), solvent evaporation (Aliabadi et al., 2007), emulsification (Miyazawa et al., 2000) and ionic gelation (Chan, 2011). On the other hand, oils can be encapsulated in capsules using a coacervation method (Katona et al., 2010), extrusion through concentric nozzles (Wyss et al., 2004), condensation and interfacial polymerization (Bouchemal et al., 2006), and microfluidic devices (Ren et al., 2010). The key parameter in any of these processes is the selection of wall material (Kaushik et al, 2014).

Spray-draying

Spray-drying is a low-cost microencapsulation technology and the most commonly used in the food industry. This technique has been widely used for drying heat-sensitive foods, pharmaceuticals and other substances, because of the rapid evaporation of the applied solvent from the droplets. Spray-drying involves atomization of the feed (dispersion or emulsion) using a pressure nozzle or a centrifugal wheel into a hot medium (typically air), resulting in rapid evaporation of water (Figure 3).

It involves the transformation of a feed from a liquid form (solution, dispersion or paste) into a powder by spraying the feed into a hot drying medium (Zuidam and Shimoni, 2010). Dzondo-Gaget et al. (2005) used spray-drying and freeze-drying to encapsulate oil extracted from safou fruit. The storage stability was evaluated determining parameters such as: iodine value, peroxide value and the content of thiobarbituric reactive species for a period of two months, at four temperatures from 4 to 50 °C. According to the authors, the freeze-dried particles were more efficient against oil oxidation than spray-dried particles.

Emulsification/internal gelation

Microspheres of less than 100 μm can be produced by the emulsification/internal gelation technique (Poncelet, 2001) by dispersing an alginate solution containing an insoluble calcium salt into an oil. Gelation is achieved by gentle acidification with an oil-soluble acid that causes calcium ion release. Small-sized alginate microspheres may be an adequate system for delivering diverse ingredients since they permit a predictable gastrointestinal transit time and an increased surface area for interaction with the intestinal epithelium. Alginate bioadhesive properties will be favored by a more intimate contact between intestinal mucosa and smaller microspheres (Silva et

al., 2006). Using this method, Ribeiro et al. (1999) encapsulated drugs dissolved in soy oil in chitosan-coated alginate microspheres.

Cocrystallization

This process consists of introducing the compounds into a saturated solution of sucrose (syrup). The spontaneous crystallization of this syrup is realized at high temperatures (above 120 °C) and with a low degree of humidity. The crystal structure of sucrose is modified and small crystal aggregates (less than 30 μm) trapping the active molecule are formed (Beristain et al., 1996). The main advantages of the cocrystallization technique are that the granular product obtained possesses a very low hygroscopicity, a good fluidity and a better stability. Beristain et al. (1996) encapsulated orange peel oil by cocrystallization using sucrose syrups. According to these authors, at proportions of 100 to 250 g oil/kg of sugar it was possible to obtain encapsulation capacities greater than 90%. When oil was encapsulated without antioxidants a sensory evaluation detected oxidized flavors in oils after storage at 35 °C for 1 d. When BHA was added to the oil prior to cocrystallization, no signs of oxidized flavors were detected after two months of storage at ambient temperature. Furthermore, the cocrystallization offers a good economic alternative and remains a flexible technique because of its simplicity (Munin and Edwards-Lévy, 2011).

Emulsion extrusion

Emulsion extrusion is considered to be the most common approach of microencapsulation and might be achieved by emulsifying or dispersing the hydrophobic components in an aqueous solution where gelation occurs (ionotropic or thermal) (Yuliani et al., 2006). Using this method,

Yilmaz et al. (2001) encapsulated sunflower oil in a starch matrix. The authors observed that the average size of the dispersed oil droplets decreased as the hydrophobic-hydrophilic balance value increased, and this was explained by the observed decrease in the interfacial surface tension between the starch melt and the oil phase. Average sizes of oil droplets also decreased with increasing screw speed, increasing melt temperature and decreasing throughput. The screw configurations also affected the average sizes of dispersed oil droplets. By using emulsion extrusion for microencapsulation, a broad selection of polymer coatings ("shell") and methods of deposition are available, which are easily adaptable to large-scale production. Alginates are natural, commonly used as wall materials since they show high toughness and have considerable effects on the mechanical stability of beads (Figure 4) (Soliman et al., 2013).

Solvent evaporation

In the solvent evaporation method, three phases are present. They are core, coat material and liquid manufacturing vehicle (LMV). Initially coat material will be dissolved in a volatile solvent, which is not soluble in the LMV phase. A core material to be encapsulated, dissolved or dispersed in the coating polymer solution is added to the liquid manufacturing vehicle phase with agitation, and the mixture is heated to evaporate the solvent for polymer. Here the coat material shrinks around the core material and encapsulates the core (Jyothi et al., 2010). Mineral and vegetable oils could be used as LMV allowing the encapsulation of hydrophobic drugs, vitamins, peptides, proteins or food additives like pigments (Tiwari and Verma, 2011).

Emulsification

Colloidal particles with a hollow interior represent a special class of core-shell particles. They often exhibit properties that are substantially different from those of other particles (e.g., their low density, large specific surface area, stability and surface permeability), thus making them attractive from both a scientific and technological viewpoint. Hollow particles represent a distinct class of materials that are of interest in the fields of medicine, pharmaceutics, materials science and the paint industry (Miyazawa et al., 2000). They find diverse applications, including encapsulation of products (for the controlled release of drugs, cosmetics, inks and dyes), protection of sensitive components (such as enzymes, proteins and oils), ultrasound contrast agents, catalysts, coatings, composite materials, artificial cells and fillers (Zoldesi et al., 2007). Chan et al. (2000) encapsulated wheat germ oil using sodium alginate by the emulsification method. According to these authors the encapsulation efficiency and oil content of wheat germ oil increased with an increase in oil load. The mean size of the microspheres increased sharply at a high oil load of 250% wt/wt. The microspheres were larger, spherical and had more vesicles within, as oil load increased. The emulsification method developed was successfully applied to wheat germ oil, with a maximum encapsulation efficiency of approximately 88%.

Ionic gelation and inverse gelation

The conventional method of producing calcium alginate beads through ionic gelation is by dropping an alginate solution into a calcium chloride solution. If the procedure is inversed, that is, a calcium chloride solution is dropped into an alginate solution, aqueous-core calcium alginate capsules are produced (Sasaki et al., 2008). Recently, an inverse gelation technique was used to produce oil-core capsules with a polysaccharide gel membrane. In this method, an

emulsion comprising oil and calcium chloride solution is added drop wise to an aqueous gelling solution of ionic polysaccharide (Figure 5) (Abang et al., 2012).

Fujiwara et al. (2013) obtained microcapsules containing stigmasterol through a one-stage process using the gelation technique. These authors employed a blend of polymers of sodium alginate, starch and chitosan as the coating material, and canola oil as hydrophobic solvent for the stigmasterol. Resultant microcapsules were spherical, averaging 1.4 mm in size. Encapsulation efficiency was 90.42% and method yield 94.87%. The amount of stigmasterol in the oil recovered from microcapsules was 9.97 mg/g. This technique proved feasible for the microencapsulation of stigmasterol as functional ingredient.

Coacervation method

This was the first reported process to be adapted for the industrial production of microcapsules. Currently, two methods of coacervation are available, namely simple and complex processes. The mechanism of microcapsule formation for both processes is identical, except for the way in which the phase separation is carried out. In simple coacervation, a desolvation agent is added for phase separation, whereas complex coacervation involves complexation between two oppositely charged polymers (Figure 6). The three basic steps in complex coacervation are: (i) formation of three immiscible phases; (ii) deposition of the coating; and (iii) rigidization of the coating (Jyothi et al., 2010).

Katona et al. (2010) used this method to obtain emulsions of sunflower oil in ternary hydroxypropylmethyl cellulose (0.7%)/sodium carboxymethyl cellulose (0.3%)/sodium dodecylsulfate (0.00–2.00 %) mixtures. These authors investigated different viscoelastic

properties at the oil in water interface in order to prepare oil-containing microcapsules with a coacervate shell of different properties. Deposition and stability of the coacervate shell depend on dodecyl sulfate concentration in emulsion. Powder of microcapsules was obtained by spray drying, and barrier properties of microcapsules' shell were investigated by oil extraction. The amount of extracted oil correlates with viscoelastic properties of corresponding coacervate.

Extrusion through concentric nozzles

This method is based on the simultaneous dropping of an oil droplet and the shell material through a vibrating concentric nozzle in a stream of cooling oil. The hell material may consist of gelatin or other polysaccharides and glycerol (as a plasticizer), and the fat content of the obtained particle is approximately 90 wt%. The obtained capsules may have a relatively high oil load of about 70–90 wt% and a particle size of between 0.8 and 8.0 mm (Wyss et al., 2004). Mostafa et al. (2013) encapsulated (gelatin as shell material) vegetable the oil blends ratio rich in omega-3 fatty acids using this method. The oil blends ratios were: 19 olive oil: 1 flaxseed oil (OF) and 9 canola oil: 1 flaxseed oil (CF-2). The prepared capsules were packed in a brown glass jar and kept at 22 °C for 6 months. The oil blend CF-2 exhibited the higher values of peroxide (9.58 meq O_2/kg) and thiobarbituric acid (0.498 absorbance/kg). Because both blends containing flaxseed oil, the differences changes in the rate of the oil stability inside the capsules could be attributed to the other oil type in such blends. Canola oil had more unsaturated fatty acids especially linolenic acid than olive oil rich in oleic acid and nearly free from linolenic one.

Condensation and interfacial polymerization

Condensation and interfacial polymerization methods entail polymerization or cross-linking at the continuous/noncontinuous interface of monomers or components dissolved in one or more phases. The dispersed capsules may be dried to provide a powder (Bouchemal et al., 2006). Shell materials commonly used include polyesters, polyurethanes, polyureas, polycyanoacrylates and melamine-formaldehyde resins. Materials used to form, cross-link or harden the shell must be screened for any unwanted reaction with the core ingredients as well as for their toxicological potential; residual monomer content can pose a problem as may the presence of residual solvent (Whateley, 1996). Prasetya and Hasokowati (2010) encapsulated a pesticide solution dissolved in refined palm oil using urea-formaldehyde (UF) resin. According to the authors the UF polymerization reaction takes place simultaneously in the solution and at the microcapsule surface. UF reaction in the solution produced UF polymer micro particles, while UF reaction at the microcapsule surface forms microcapsule shell. The UF polymer micro particles precipitate in the form of fine powder, attach to the microcapsule surface. Based on oil and resin efficiencies as well as microcapsule characteristics (diameter from 20 μm-around 220 μm), the process was best conducted at 50 °C, 30 min of homogenization and 3 h of microencapsulation time.

Microfluidic devices

The liquid coating is sprayed onto the particles and the rapid evaporation helps in the formation of an outer layer on the particles. The thickness and formulations of the coating can be obtained as desired. Different types of fluid-bed coaters include top spray, bottom spray and tangential spray. In the top spray system the coating material is sprayed downwards on to the fluid bed so

that as the solid or porous particles move to the coating region they become encapsulated (Ren et al., 2010).

Properties of vegetable oils microcapsules

Spray-drying and coacervation are the two most frequently reported methods for the encapsulation of vegetable oils. The first method is widely used for drying heat-sensitive foods, pharmaceuticals and other substances, because of the rapid evaporation of the applied solvent from the droplets. The second method was the first reported process to be adapted for the industrial production of microcapsules. Sometimes both methods are used consecutively for the obtaining of oil microcapsules.

Yu et al. (2012) obtained olive oil capsules by complex coacervation using gelatin and acacia gum as wall materials, and glutaric dialdehyde as cross-linking agent. The authors evaluated process parameters, such as the dosage of the cross-linking agent, concentration of the wall materials, pH value and the ratio between core and wall materials. The optimum process parameters were as follows: The dosage of the cross-linking agent was 3 mL, the concentration of wall materials was 3%, the pH value of coacervation was 4.0 and the ratio of core/wall material was 1:1. In order to obtain microcapsules with good mobility and dispersal, a spray-drying process was used by the authors to dry the product. The capsules had a mean particle size of 6 μm and a loading oil rate of 60%. At 80 °C the time and retention rate that the microcapsules had a fast release stage in the first five hours and then remained almost steady. The higher decomposition temperature (180 °C) of the microcapsules was obtained under higher concentration of wall materials. This is due to the fact that the wall materials play an important

role in the heat-resistant microcapsule. According to Yu et al. (2012), using the method of complex coacervation, the first elastic layer of the microcapsule was obtained, and then the second glass layer was obtained through spray-drying. The two double-wall constructures resulted in the good heat-resistant property of microcapsules.

In another study, Devi et al. (2012) encapsulated olive oil by complex coacervation using gelatin, sodium alginate and glutaraldehyde as cross-linking agents. For these authors the optimum ratio between gelatin A-sodium alginate and pH to form the maximum coacervate complex was found to be 3.5:1 and 3.5:8, respectively. In these conditions the oil load, the oil content and the encapsulation efficiency were 188%, 58% and 89%, respectively. Through scanning electron microscopy analysis the formation of free-flowing spherical microcapsules of different sizes was confirmed. With the increase of the amount of polymer concentration, the size of the microcapsules increased. According to Devi and Maji (2010), this might be due to the increase of the thickness of the wall of the microcapsules. The Fourier transform infrared and thermogravimetric analysis carried out by the authors did not exhibit any remarkable interaction between olive oil and gelatin-sodium alginate complex. This indicates that the encapsulated material retains all its properties.

A major property of encapsulated powders manufactured for consumer use is their ease of reconstitution. The reconstitution process in water can be divided into four steps: wetting, submersion, dispersion and dissolving (Schubert et al., 2003). Wettability is understood as the ability of a bulk powder to imbibe a liquid under the influence of capillary forces. Generally, it depends on powder particle size, density, porosity, surface charge, surface area and the presence

of amphipathic substances. Fast wetting is also favored by large particles of high porosity (Domian, 2005). In this sense, Domian and Wqsak (2008) encapsulated rapeseed oil by spray-drying using maltodextrin and acacia gum. The emulsions were dried at a constant temperature of inlet and outlet air – 200 °C and 100 °C, respectively – and at a spray disk speed reaching 34,000 rpm. The capsules were characterized by a regular and spherical shape, a smooth surface with visible cavities, and constituted powder particles with sizes ranging from 10 to 90 μm. The encapsulation yield ranged between 71% and 94% and the encapsulation efficiency ranged between 83% and 95%. According to the authors, directly after drying, the powders demonstrated low water activity, i.e. aw of 0.04–0.15, at a moisture content w of 2.5–4.8 %. Loose and tapped bulk density as well as a porosity of loosely poured and packed bed reached ρL 430–489 kg/m^3 and ρT 695–765 kg/m^3 as well as εL 0.60–0.65 and εT 0.35–0.45, respectively. Particles of microencapsulated oil powder were characterized by density ρ ranging from 1184 to 1288 kg/m^3. The authors concluded that spray-drying microencapsulation of rapeseed oil onto a maltodextrin carrier with the addition of acacia gum enabled the obtaining of a powdered product with complete water reconstitution and very poor wettability and flowability.

Oxidation stability and fatty acid composition of encapsulated vegetable oils

Lipid oxidation in microencapsulated lipids is of paramount importance because it may result in a loss of nutritional value and the development of flavors unacceptable to consumers in a significant number of products such as infant formulas, bakery products, milk powders, dried eggs, and dehydrated soups and sauces (Velasco et al., 2003). In this sense, Dzondo-Gaget et al.

(2005) encapsulated oil extracted from the safou fruit by using spray-drying and freeze-drying. These authors evaluated the storage stability by determining parameters such as iodine value, peroxide value and the content of thiobarbituric reactive species in the oil for a period of two months, at four storage temperatures from 4 to 50 °C. According to the authors, the freeze-dried particles were more efficient against oil oxidation than spray-dried particles. In another study, Beristain et al. (1996) encapsulated orange peel oil by cocrystallization using sucrose syrups. According to these authors, at proportions of 100 to 250 g oil/kg of sugar it was possible to obtain encapsulation capacities greater than 90%. When oil was encapsulated without antioxidants a sensory evaluation detected oxidized flavors in oils after storage at 35 °C for one day. When BHA was added to the oil prior to cocrystallization, no signs of oxidized flavors were detected after two months of storage at ambient temperature. This suggests that encapsulation with the addition of antioxidant additives could prolong the storage time of the encapsulated oils.

Calvo et al. (2010) encapsulated extra virgin olive oils by spray-drying using sodium caseinate and lactose as wall materials. The spray-drying conditions were: The pressure of compressed air for the flow of the spray was 5 bars. The inlet and outlet air temperatures were maintained at 165 and 80 °C, respectively, with a feed rate of 540 mL/h. With these conditions the oil encapsulation yield was 53%. For the extra virgin olive oil that had highest amounts of total phenolic contents (459.64 mg caffeic acid/kg oil), oxidative stability index (83.89 h), and a ratio between C18:1 and C18:2 (12.82), the fatty acid profile was unaltered after the microencapsulation process.

Fatty acid profile and Vitamin E contents could be used as parameters to evaluate the nutritional value and to regulate the marketing of oil-encapsulated base products. In this sense, Hirashima et al. (2013) evaluated the identity (fatty acid profile) and the compliance with nutritional labeling (fatty acid and Vitamin E contents) of 21 commercial encapsulated oils. Samples included: flaxseed oil (6), evening primrose (5), safflower (8), borage (1) and blackcurrant (1). Nine samples were adulterated (five samples of safflower oil, three of flaxseed oil and one of evening primrose). Among them, three flaxseed and two safflower oil samples were probably adulterated by the addition of soybean oil. Only two samples presented all values in compliance with nutritional labeling (one safflower oil sample and one borage oil sample). The results show that a continuous monitoring of commercialized encapsulated oils is necessary.

It is difficult to foresee the rate of oxidation in heterogeneous systems due to the high number of variables involved. In particular, the evolution of oxidation in the noncontinuous or dispersed lipid phase may become very complex due to the heterogeneity in the lipid droplets isolated one from another in the matrix. Consequently, different oxidation rates can occur in different droplets. However, after extraction of the encapsulated fraction, a continuous oily phase is analyzed and substantial information on the oxidation in the different droplets is lost (Velasco et al., 2003). Some authors have reported that oxidation proceeded more rapidly in freeze-dried samples than in spray-dried samples, attributing such results to the greater surface area of the former (Fioriti et al., 1975; Taguchi et al., 1992). Other researchers have found the opposite (Desobry et al., 1997; Stapelfeldt et al., 1999), even starting from samples with similar microencapsulation efficiency (Desobry et al., 1997), then attributing the lower oxidative stability of spray-dried samples to the high temperatures used during the atomization process.

Minemoto et al. (2001) compared freeze-drying with hot-air drying at 50 °C, finding freeze-dried samples more resistant to oxidation, even though they showed lower microencapsulation efficiency. Moreover, this research group suggested that the effect of the drying method might be closely related to the type of encapsulating agent and oxidative conditions. According to de Barros-Fernandes et al. (2013) the use of higher temperatures and low feed flow rate contributed to the decrease of particle moisture content and, in general, it was related to the variation of the hygroscopicity and wettability of the resulting powders. A higher moisture content may lead to oxidation of the encapsulated material.

Incorporation of long chain omega-3 polyunsaturated fatty acids in foods

Fortification of commonly consumed food products with n-3 long-chain omega-3 polyunsaturated fatty acids in foods is considered an innovative way of providing health benefits to people without major alteration to their dietary habits (Garg et al., 2006). An alternative to increasing consumption would be to supplement with ω-3 daily foodstuffs such as margarine, eggs and their products, pasta, sauces, juices, meat, and milk and dairy products – the so-called functional foods (Gómez-Candela et al., 2011). There is a restriction in the level of oil fortified in different products such as bakery, dairy and other frozen foods as oils are highly susceptible to oxidation during storage (Willumsen, 2006). Fatty acid present in oil would undergo oxidative deterioration leading to degradation of the original long carbon chain to yield highly reactive intermediate lipid radicals and ultimately potentially unhealthy small molecules. It has been found that hydroperoxides, which are the primary products of lipid oxidation, are considered to be toxic for human health (Oarada, 1988). As the number of double bonds in a fatty acid

increases, so does the rate of oxidation. This makes polyunsaturated lipids such as omega-3 fats highly susceptible to oxidation (Keogh et al., 2001). Oil encapsulation may be useful to retard lipid autooxidation and increase the range of applications where otherwise oil could not be used (Calvo et al., 2010).

Rubilar et al. (2012) encapsulated linseed oil as a source of omega-3 fatty acids by spray-drying. These authors evaluated the influence of gum arabic, maltodextrin and mixtures of both in a ratio of 3:2 as wall material, the concentration of wall material (25 and 30 % in 100 g of emulsion), and the concentration of oil (14 and 20 % in 100 g of emulsion) in the encapsulation of linseed oil. The spray-drying conditions were: an air input temperature of 140 °C, an outlet temperature of 95 °C, with a drying airflow of 73 m^3/h and a feed rate of 5.3 g/min. The maximum encapsulation efficiency was obtained with a mixture of both wall materials of 3:2, at a concentration of 30% with 14% of oil. The encapsulated oil exhibited an induction time and an oxidative stability index at 100 °C of 2.83 h and 3.78 h, respectively. According to these authors, the spray-drying conditions (temperature) did not affect the linseed oil's chemical stability. However, the amounts of oleic and linoleic acid decreased by almost 50%. Microcapsules of linseed oil were added to a soup formulation in a proportion of 14%; this proportion provides approximately 80% of the recommended daily intake of α-linolenic acid (1 g/day) according to the International Society for the Study of Fatty Acids and Lipids. The oxidative stability and the shelf life of oil extracted from the soup exhibited an induction time of 14.85 h and the stability time was 15.61 h. The determined shelf life was 8.78 months.

Conclusion

Encapsulation methods such as spray-drying and coacervation have allowed the obtaining of microcapsules of diverse vegetable oils. A variety of other methods are in development including spray chilling, extrusion coating and liposome entrapment. The key parameter in any of these processes is the selection of wall material. For spray dried emulsions and complex coacervates protein or polysaccharides are primarily used as shell material, although complex coacervation is currently commercially limited to gelatin. When vegetable oils are encapsulated oxidation is reduced, allowing storage for longer periods. Similarly, the composition of fatty acids is not significantly affected. Strategies involving the use of novel microencapsulation techniques along with the use of a combination of antioxidants could increase the stability of such products. Omega-3 PUFA-enriched foods should be stable and convenient, have an acceptable taste/flavor, and be without heavy price premiums in order to achieve an effective population-wide increase in consumption of these bioactive fatty acids. Further studies on the interaction of the oils in foods, oxidative stability, bioavailability and nutritional impact are needed.

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Table 1. Composition of vegetable oils rich in n-3 fatty acids.

Vegetable oil	Oil content in seed (g/100 g)	$C_{18:3}$ (n-3) (%)	$C_{18:1}$ (n-9) (%)	$C_{18:2}$ (n-6) (%)
Dracocephalum moldavica	20	61	10	18
Linum usitatissimum	31-55	55-65	6-20	7-14
Perilla frutescens	35	58	19	14
Salvia hispanica	30	51	12	19
Aleurites moluccana	30	39	28	40

Table 2. Summary of the antiinflammatory effects of long-chain n-3 fatty acids (Calder, 2001).

Antiinflammatory effect	Mechanism likely to be involved
Decreased generation of arachidonic acid derived eicosanoids (with inflammatory actions).	Decreased arachidonic acid in cell membrane phospholipids; inhibition of arachidonic acid metabolism; decreased induction of COX-2, 5-LOX, and 5-LOX activating protein.
Increased generation of EPA-derived eicosanoids (with less inflammatory actions than those produced from arachidonic acid).	Increased content of EPA in cell membrane phospholipids.
Increased generation of EPA and DHA-derived resolving (with antiinflammatory actions).	Increased content of EPA and DHA in cell membrane phospholipids.
Decreased generation of inflammatory cytokines (TNF-α, IL-1β, IL-6, and IL-8).	Decreased activation of NFκB (via decreased phosphorylation of IκB); activation of PPARγ; altered activity of other transcription factors; differential effects of arachidonic acid vs EPA derived eicosanoids.
Decreased expression of adhesion molecules.	Decreased activation of NFκB (via decreased phosphorylation of IκB); altered activity of other transcription factors.
Decreased leukocyte chemotaxis.	Not clear; perhaps decreased expression of receptors for some chemoattractants.
Decreased generation of reactive oxygen species.	Not clear; perhaps altered membrane composition affecting signaling processes.

COX, cyclooxygenase; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; IκB, inhibitory subunit ofNFκB; IL, interleukin; LOX, lipoxygenase; NFκB, nuclear factor κB; PPAR, peroxisome proliferator-activated receptor; TNF, tumor necrosis factor.

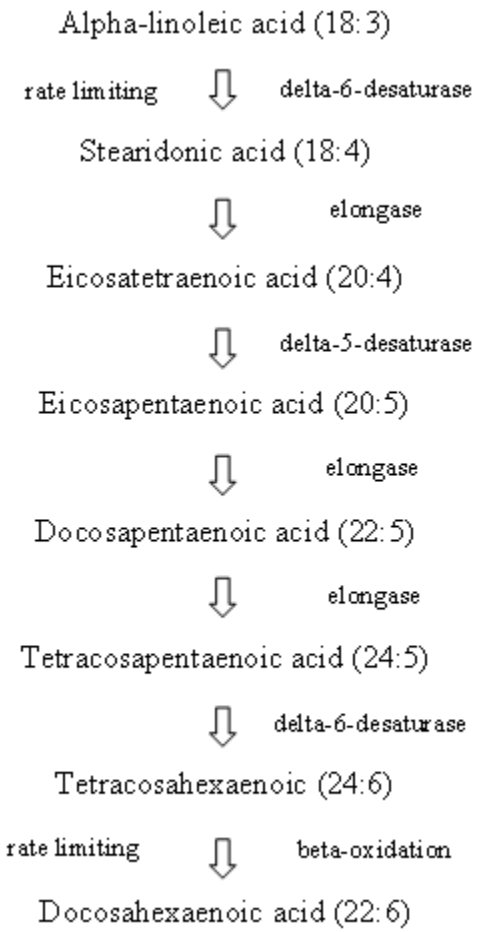


Figure 1. A series of elongation and desaturation reactions allows conversion of short-chain omega-3 fatty acids into the longer chain polyunsaturated fatty acids such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). The delta-6-desaturase catalyses the rate-limiting enzymatic reaction, leading to inefficient conversion to SDA (stearidonic acid) in humans (Burdge and Calder, 2005).

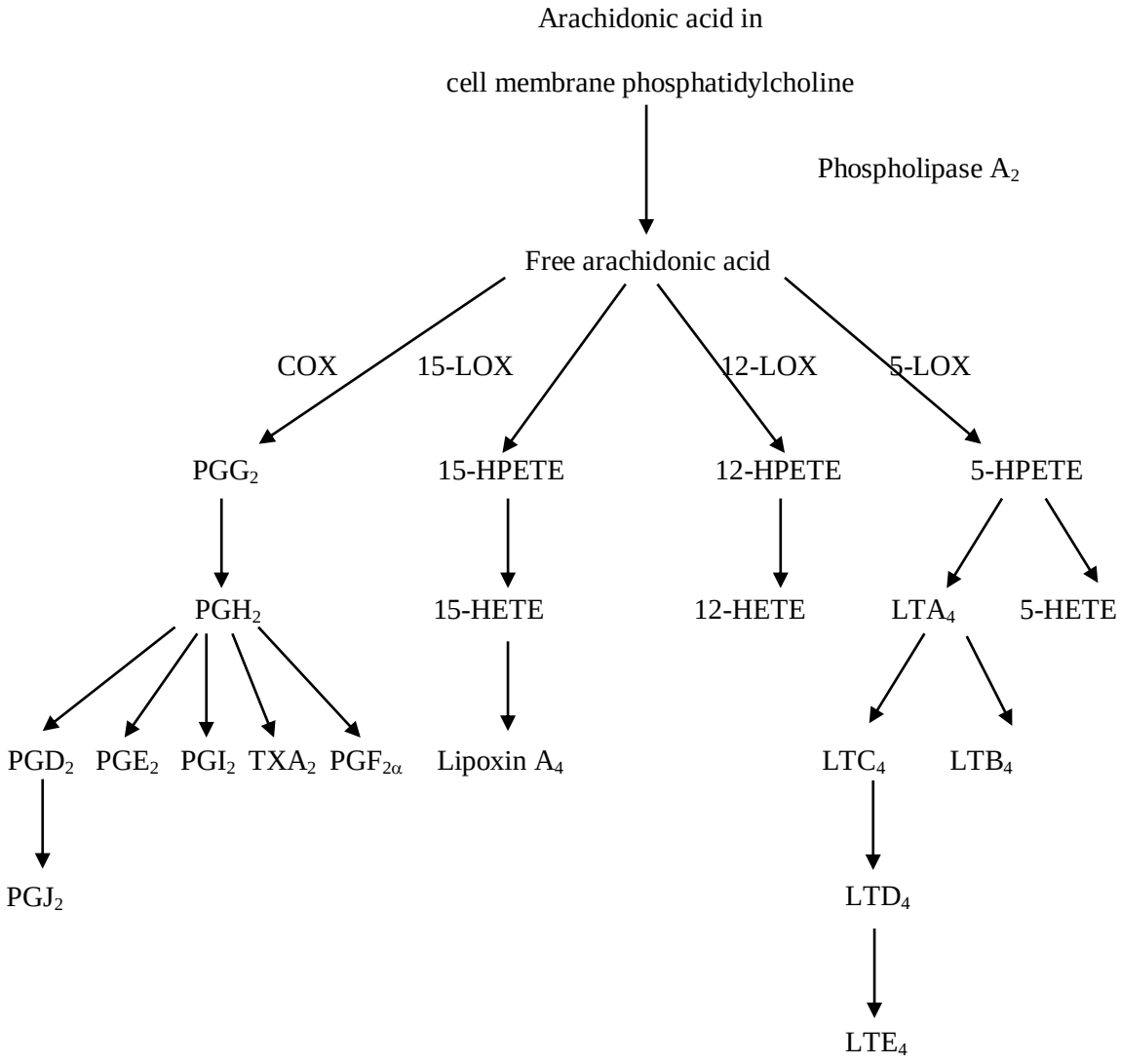


Figure 2. Generalized pathway for the conversion of arachidonic acid to eicosanoids. COX, cyclooxygenase; HETE, hydroxyeicosatetraenoic acid; HPETE, hydroperoxyeicosatetraenoic acid; LOX, lipoxygenase; LT, leukotriene; PG, prostaglandin; TX, thromboxane (Calder, 2006).

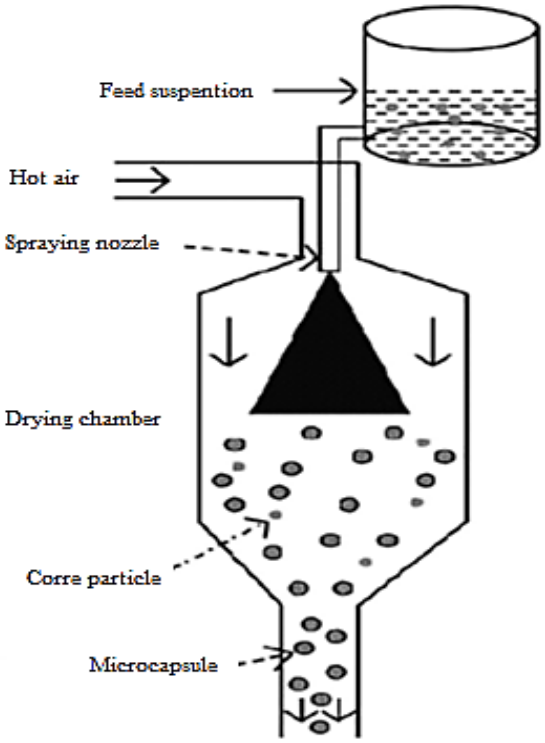


Figure 3. Schematic illustrating the process of micro-encapsulation by spray-drying (Ghosh, 2006).

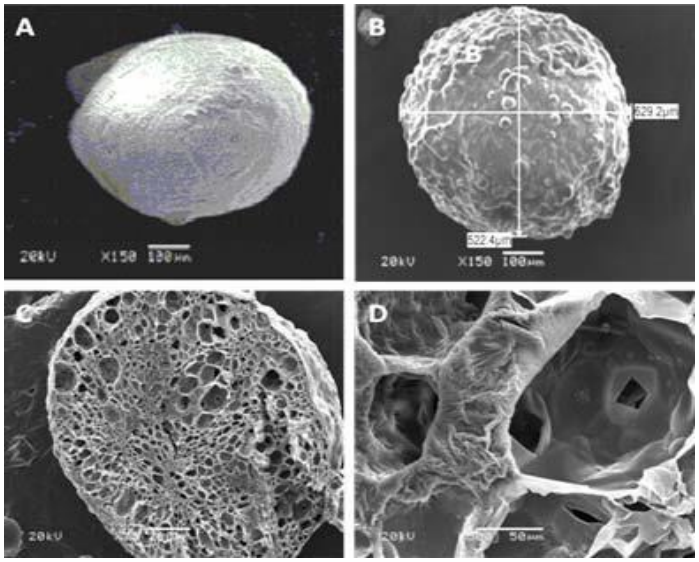
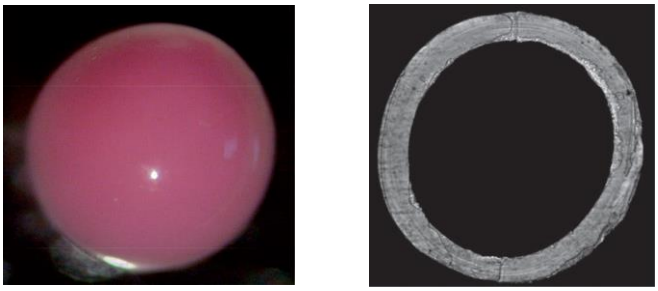


Figure 4. SEM graphs of alginate microspheres, plain (A), EO-loaded (B), cross-section of EO-loaded MS (C) and its microstructure (D). (Soliman et al., 2013).



a) b)

Figure 5. a) Wet alginate capsule produced from droplets of oil containing calcium chloride solution (water/oil emulsion). b) Confocal laser scanning microscope fluorescent images of a wet membrane layer of calcium alginate. (Abang et al., 2012).

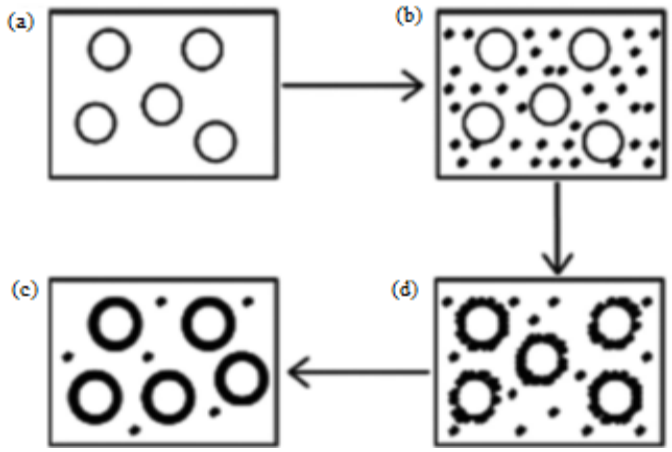


Figure 6. Schematic representation of the coacervation process. (a) Core material dispersion in solution of shell polymer; (b) separation of coacervate from solution; (c) coating of core material by microdroplets of coacervate; (d) coalescence of coacervate to form continuous shell around core particles (Ghosh, 2006).