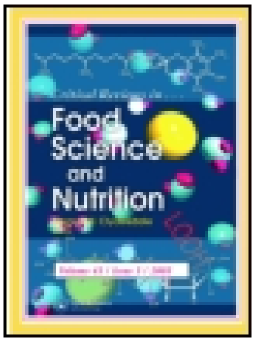

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A systematic review and meta-analysis of fish oil encapsulation within different micro/nanocarriers

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ABSTRACT
Fish oil is one of the most important sources of omega 3 polyunsaturated fatty acids (PUFAs), especially eicosapentaenoic acid and docosahexaenoic acid which are the most important PUFAs with several health benefits. However, PUFAs are prone to oxidation and have a poor water solubility which limits the use of fish oils into food formulations. Encapsulation techniques can be applied to overcome these challenges. There is a large number of published micro/nanoencapsulation papers, where each of them contains a limited number of wall materials, feed formulation, encapsulation technique, and storage conditions. Therefore, without systematic evaluation of the data extracted from available studies, the design of functional foods containing fish oil would not be very successful. The objective of this systematic review is a meta-analysis of published researches on the nano/microencapsulation of fish oil. A comprehensive literature search was performed between 1 October and 31 December 2019 with encapsulation, fish oil, and oxidative stability keywords. Overall, 39 qualified articles were selected for the statistical analysis. Based on the technique used for encapsulation, the fish oil-loaded carriers were classified into four main groups: (a) spray-dried particles; (b) freeze-dried particles; (c) electrospun fibers and electrosprayed capsules; and (d) other carriers prepared by supercritical antisolvent, gelation, liposomes, spray–freeze drying, and transglutaminase catalyzed cross-linking. The three most frequent methods applied for fish oil encapsulation were spray drying (42.86%), freeze drying (21.43%), and electrohydrodynamic (19.04%) methods, respectively. Averagely, the best encapsulation efficiency was obtained for electrohydrodynamic processes. Also, the combination of polysaccharide–protein based wall materials provided the best performance in terms of fish oil encapsulation efficiency.

KEYWORDS
Carriers; encapsulation efficiency; fish oil; oxidative stability; wall materials

Introduction

Fatty acids as the basic and simplest unit of lipids are classified into saturated and unsaturated fatty acids. Omega-3 fatty acids, that is, docosahexaenoic acid (C22:6n-3, DHA), eicosapentaenoic acid (C20:5n-3, EPA), and α-linolenic acid (ALA) are the most important polyunsaturated fatty acids (PUFAs) with several health benefits, including anti-cardiovascular, anti-inflammatory, anti-hypertensive, anticancer, anti-depression, anti-arthritis, and antioxidant effects besides their crucial roles in the growth and development of heart, brain, and eye retina (Manerba et al. 2010; Marventano et al. 2015; Widomska, Zareba, and Subczynski 2016). The lack of specific desaturase enzymes in mammals causes no production of ω-3 fatty acids in the human body and so, they must be obtained by the diet (Surette 2008). As a result, the food industry exhibits a growing interest in developing ω-3 enriched foods. Fish oil is one of the most prevalent natural sources of ω-3 PUFAs, especially EPA and DHA. Therefore, foods containing fish oil can be very important in this regard. However, PUFAs are very unstable and very sensitive to oxidation under normal conditions (Abbasi et al. 2019; Khoshnoudi-Nia and Moosavi-Nasab 2019).

Decomposition of fatty acids into volatile molecules during oxidation not only has a significant role in destroying the taste and aroma of the oils (known as rancidity), but also it leads to the production of the various compounds with toxic effects (e.g. free radicals) on human health (Pham-Huy, He, and Pham-Huy 2008; Ghorbanzade et al. 2017). On the other hand, unpleasant and typical fishy flavors of fish oils is another drawback limiting their application (Serfert, Drusch, and Schwarz 2010). In general, the major challenge of producing ω-3 enriched foods is to suppress or retard the oxidation of fatty acids during processing and storage until consumption and consequently, provide stable products with an acceptable taste (Drusch et al. 2006). Micro/nanoencapsulation is one of the common techniques which can be offered to produce such products. Encapsulation can form a matrix or polymeric cover over the fish oil and subsequently, protects food against the effects of various environmental factors (e.g. temperature, moisture, pH, oxygen, etc.) and provides the controlled

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release of the ω-3 fatty acids under certain conditions (Kolanowski, Laufenberg, and Kunz 2004; Hambleton et al. 2008).

Several methods have been used to encapsulate fish oil for application in food products. The choice of the best encapsulation technique basically depends on the features of the final product (Dias et al. 2017). These methods are based on different physical and chemical processes such as spray drying, freeze drying, spray–freeze drying, electrospraying, liposomal entrapment, supercritical antisolvent process (SCAP), and so on, which have been applied successfully to encapsulate fish oil (Assadpour and Jafari 2019a, 2019b; Rezaei, Fathi, and Jafari 2019). The encapsulation efficiency and the storage stability of encapsulated fish oil depend on the type of wall materials, the encapsulation process (and the properties of final product. Various biopolymers have been investigated to encapsulate fish oil. Polysaccharides such as gum arabic (GA) (Binsi, Natasha, et al. 2017), pullulan (García-Moreno et al. 2016), chitosan (Chang et al. 2020), cyclodextrins (Choi et al. 2010), pectin (Vaucher et al. 2019), xanthan (Bahramizadeh and Rahmanifarah 2017), maize starch (Botrel et al. 2017), maltodextrin (Unnikrishnan et al. 2019), and starch (Botrel et al. 2017), as well as proteins including gelatin (Yu et al. 2017), whey protein concentrate (Wang et al. 2016), soy protein isolate (Di Giorgio, Salgado, and Mauri 2019), caseinate (Binsi, Nayak, et al. 2017), and zein (Miguel et al. 2019) are some examples.

There is a large number of published micro/nanoencapsulation papers, where each of them contains only a limited number of wall materials (type and concentration), feed formulation, encapsulation technique, and storage conditions. Therefore, without systematic evaluation of the data extracted from available studies, the design of functional foods containing fish oil will be less successful than it should be. According to the above-mentioned explanations, the current study aimed to investigate systematically the studies focused on the encapsulation of fish oil based on mentioned techniques and carriers and in particular, highlighting their effects on the encapsulation efficiency, particle size, oxidative stability, and sensory properties. The meta-analysis of the experimental and literature data was performed using one-way analysis of variance (ANOVA) and descriptive analysis.

Methods

Searching procedure

A comprehensive literature search was performed between 1st of October, 2019 and 31st of December, 2019 on Google Scholar databases. Data were collected from scientific articles ranging from 2016 to 2020. The several keywords and Boolean operators were used in the searching process such as, encapsulation and "fish oil," "oxidative stability," "fish oil and spray drying," "fish oil and freeze drying," and "fish oil and electrohydrodynamic." This search was limited to English articles after 2016. There was no restriction in terms of the type of publication, study design, and outcome measurement method.

Qualification

The articles were considered eligible if they satisfied the following criteria: (i) experimental study design; (ii) application in the food industry; (iii) since 2016; (iv) measurement of at least two specified parameters (size, encapsulation efficiency, oxidative stability, and sensory score); and (v) published in English language. In contrast, studies were excluded if they were letters, conference abstracts, patents, review articles, and abstracts without details. Our main goal was to focus on the encapsulation efficiency of various methods in order to show the future directions on the encapsulation of fish oil. Thus, 39 articles (42 case studies: some articles had compared the two or three encapsulation methods) were selected.

Data collection, extraction, and classification

Initially, the researched articles were screened according to the title, abstract, and keywords. Afterward, the relevant full-text articles were evaluated for the qualification based on the inclusion and exclusion criteria, mentioned in the previous section. Disagreements were resolved through discussion. The data extraction was executed *via* a data extraction form (Tables 1–5) based on the carrier material, solvent, processing conditions, particle size, encapsulation efficiency, oxidative stability, and sensory properties.

Calculation of the oxidative stability

One of the first problems during analyzing the literature data was lack of consistency or a standard in the presentation of oxidative stability. In order to facilitate the comparison process, coefficients of variation in both primary and secondary oxidation products were measured to evaluate the oxidative stability. For this purpose, the difference between the initial (preferably the oxidation level of crude oil before encapsulation at the beginning of the process) and the maximum amounts of primary and secondary oxidation products was calculated for each treatment and expressed as a percentage. Although this method simplified the comparison between treatments of a specific study, the major drawback was that many researchers had not reported the standardized periods and storage conditions (time, access of light and oxygen, temperature, humidity). This makes it difficult to compare the oxidative stability of various studies to obtain conclusive results.

Statistical analyses

The meta-analysis was carried out based on the descriptive (frequency of carrier agents and encapsulation techniques) and comparative analysis (the effect of encapsulation method and wall material on the encapsulation efficiency of fish oil). Encapsulation efficiency values were subjected to one-way ANOVA; Fisher *post hoc* test was used to investigate the differences between the means ($p < 0.05$). Statistical analysis was performed using the Minitab 17.1.0 software (Minitab Inc. PA, USA).

Table 1. Overview of recent studies (2016–2020) on spray drying encapsulation of fish oil.

						Oxidative stability		
Source	Polymers	Solvent	Processing conditions	Encapsulation efficiency (%)	Size	POP (%)	SOP (%)	Ref.
Fish oil (10% w/w)	β-LG (0.5% w/w) TM β-LG (0.5% w/w) Ch (0.5% w/w) MD: 15% w/w	Water	TMβ–LG +Ch+ MD = TBCM β-LG (pH = 2) + MD = BM TMβ–LG + MD = TBM T: 160, 170, & 180 °C	TBCM 160; 170; 180 °C 95.53; 89.4; 90.8 BM 160; 170; 180 °C 94.9; 94.1; 92.9 TBM 160; 170; 180 °C 79.2; 69.6; 71.6	µm TBCM 160 °C: 0.96 170 °C: 1.15 180 °C: 1.25 β-LG (pH = 2) 160 °C: 0.23 170 °C: 0.28 180 °C: 0.31	a_w < 0.4 → stability toward oxidation	–	Chang et al. (2020)
Fish oil (10% w/w)	SPI (0.35 & 0.4 w/w) MD (DE:21): 7% w/w	Water	T: 160 °C Pressure [MPa]: 0.5–25 SPI: Oil = 0.02–0.22	SPI: Oil = 0.13 Pres (MPa): 0.5; 2; 7.5; 12.5; 25 80.7; 88.5; 91.5; 93.6; 94.1 Pres (MPa): 12.5 SPI: Oil 0.02; 0.04; 0.07; 0.13; 0.22 53.8; 71.6; 90.1; 93.4; 92.1	SPI: Oil = 0.13 Pres (MPa): 0.5; 2; 7.5; 12.5; 25 169; 163; 126; 90; 74 Pres (MPa): 12.5 SPI: Oil 0.02; 0.04; 0.07; 0.13; 0.22 116; 87; 114; 107; 86	6 weeks–27 °C–Dark (based on hydroperoxides) *Pres (MPa): 0.5; 2; 7.5; 12.5; 25 * Total oil: 14,800; 11,200; 5900; 4800; 5900 * Non-encapsulated oil 7300; 4800; 1900; 1900; 1800 * Encapsulated oil: 8100; 7300; 4300; 4300; 4800 *SPI: Oil 0.02; 0.04; 0.07; 0.13; 0.22 * Total oil: 5700; 4100; 4100; 4100; 4100 * Non-encapsulated oil 3650; 2200; 1700; 1200; 1100 * Encapsulated oil: 2900; 2500;2500; 3900; 4500		Linke, Weiss, and Kohlus (2020)
Fish oil (1.5 g/100 mL)	SC (3 g/100 mL): GA (3 g/100 mL) SC:GA:Oil 2:2:1 Sage Ex (1% w/w oil)	Water	Saga Ex + GA + SC = GA + Ex GA + SC = GA T: 160 °C (Solid total: 7.5%) SC: emulsifier	GA + Ex = 73 GA = 69	µm	7 days; 60 °C F. oil: 1160 (3 days) F. oil + Ex: 810 (2 days) GA: 1500 (5 days) GA + Ex: 900 (5 days)	7 days; 60 °C FO: 3400 (7 days) FO + Ex: 2800 (7 days) GA: 1500 (6 days) GA + Ex: 900 (6 days)	(Binsi, Nayak, et al. 2017)
Fish oil	MS GA CG (15 g/100 mL)	Water	T = 160–220 °C	MS: 75.8 GA: 59.8 CG: 75.8	µm MS: 26 GA: 22 CG: 63	(56 days; 25 °C) Fish oil: 2630 (50 days) MS: 1230 (35 days) GA: 2120 (35 days) CG: 720 (30 days)	(56 days; 25 °C) Fish oil: 2200 (50 days) MS: 1700 (35 days) GA: 2600 (50 days) CG: 600 (30 days)	Botrel et al. (2017)
Fish oil	SPI (5% w/v)	Water	SPI: Oil 1:1; 2:1; 3:1; & 4:1 Prepare emulsion by: UT UT + US Spray drying: Pressure: 6–8 bar T: 180 °C	UT 1:1 = 57.8 2:1 = 69.2 3:1 = 69.1 4:1 = 65.2 UT + US 1:1 = 66.9 2:1 = 57.7 3:1 = 69.8 4:1 = 88.7	15–20 µm	UT 1:1 = 1400 2:1 = 2200 3:1 = 3000 4:1 = 4400 UT + US 1:1 = 50 2:1 = 100 3:1 = 200 4:1 = 400	UT 1:1 = 400 2:1 = 1200 3:1 = 31; 1300 4:1 = 1400 UT + US 1:1 = 10 2:1 = 250 3:1 = 600 4:1 = 650	Di Giorgio, Salgado, and Mauri (2019)
Roes of rohu fish (*Labeo rohita*) oil	GA (5% w/w)	Water	T: 160 °C RC: spray-dried oil RGA: GA–oil	RC = 72.36 RGA = 6.95	µm RC = 76–216 RGA = 89–134	(7 days; 60 °C) RC = 700 (5 days) RGA = 300 (7 days)	(7 days; 60 °C) RC = 500 RGA = 150	Binsi, Natasha, et al. (2017)
Fish oil (2.5 g)	HPC (1.7–10.8 g/ 87–96 g solvent)	Water Ethanol Methanol Acetone	Water (T): 200 °C Ethanol: 135 °C Methanol: 79 °C Acetone: 78 °C Oil:solvent = 1:4	F–W: 71.1 F–M: 75 F–E: 80.4 F–A: 92	µm F–W: 2.43 F–M: 1.79 F–E: 0.53 F–A: 1.77	–		Encina et al. (2018)
Fish oil (25 g)	HPMC 15 cps = H HPMC 5 cps = h	Water (448.7–467.5 g)	H + h + W (g) CF1: 9.375 + 9.375 + 455 CF2: 6.25 + 6.25 + 461.25 CF3: 3.125 + 3.125 + 467.5 DF1: 0 + 25 + 448.75 DF2: 6.25 + 18.75 + 448.75 DF3: 12.5 + 12.5 + 448.75 DF4: 18.75 + 6.25 + 448.75 DF5:25 + 0 + 448.75 T = 180 °C	CF1: 70.9 CF2: 69.3 CF3: 67.3 DF1: 74.7 DF2: 73.7 DF3: 72.3 DF4: 70.4 DF5:69.2	µm CF1: 29.56 CF2: 37.2 CF3: 18.3 DF1: 54.7 DF2: 34.3 DF3: 31.8 DF4: 30.0 DF5:27.84	PV28–PV0* 28 days, 25 °C CF1: 84 CF2: 38 CF3: 47 DF1: 252 DF2: 34 DF3: 60 DF4: 47 DF5:60	–	Karim et al. (2016)

(*continued*)

Table 1. Continued.

Source	Polymers	Solvent	Processing conditions	Encapsulation efficiency (%)	Size	Oxidative stability POP (%)	Oxidative stability SOP (%)	Ref.
Sardine oil	MD GA SC PH	Water	Fish oil:wall material 1:5 $T = 160\,°C$	SO: 73.9 SPO:78.3 PO:76.6 SP: 78.7	μm	28 days, 25 °C SO = 830 SPO = 820 PO = 1500 SP = 500 Fish oil = 820	28 days, 25 °C SO = 333 SPO = 333 PO = 1200 SP = 333 Fish oil = 1400	Unnikrishnan et al. (2019)
Fish oil	MD + SC (DE: 11, 19, 25)	Water	Feed solution: 24 wt.% = fish oil 1.8 wt%: SC (emulsifier) 34.2 wt% MD 40 wt.% water T: 140 °C	DE11: 74 DE19:89 DE25:90	μm DE11: 45 DE19: 38 DE25: 41	(5 days; 60 °C) Surface oil; en oil; total oil DE11: 7850; 800; 1150 DE19: 7900; 320; 710 DE25: 7800; 250; 620	–	Abd Ghani et al. (2017)
Crude fish oil	GA MD	Water	T: 170 °C Antioxidant: 0.58% w/v GA + MD + FO + ALT (%) GA1: 15 + 15 + 15 + 0 GA2: 15 + 15 + 15 + 0.58 GA3: 10 + 20 + 10 + 0.58	GA1 = 51.2 GA2 = 56.8 GA3 = 54.7	μm GA1 = 57.4 GA2 = 58.7 GA3 = 29.3	(40 °C, 28 day) FO; FO + ALT; 875; 980 GA1 = 815 GA2 = 777.5 GA3 = 581.2 (21 day)	(40 °C, 28 day) FO; FO + ALT; 360; 375 GA1 = 100 GA2 = 120 GA3 = 160	Vaucher et al. (2019)
Fish oil	CP MD	Water (40 mL)	Complex coacervation followed by *spray drying* T: 170 °C CP + MD + FO + ALT CP1: 7.5 + 7.5 + 7.5 + 0 CP2: 7.5 + 7.5 + 7.5 + 0.58 CP3: 7.5 + 15 + 7.5 + 0.58	CP1 = 67.9 CP 2 = 65.3 CP 3 = 64.7	μm CP 1 = 22.45 CP 2 = 18.6 CP 3 = 37.9	(40 °C, 28 day) FO; FO + ALT; 875; 980 CP 1 = 137 CP 2 = 137 CP 3 = 112.5	40 °C, 28 day) FO; FO + ALT; 875; 980 CP 1 = 62 CP 2 = 62 CP 3 = 34	Vaucher et al. (2019)
Fish oil (1, 5, & 10)	WPI (1 & 2)	Water	T: 170 °C Oil: wall (solid content) T (10.1): 10:1 (10%) NT (10.1) = 10:1 (10%) T (5.1) = 5: 1 (10%) NT(1.1_{25}) = 1:1 (25%) NT (1.1_{10}) = 1:1 (10%) NT (1.2) = 1:2 (25%)	T (10:1) = 25 NT (10:1) = 15 T (5:1) = 44 NT($1{:}1_{25}$) = 72 NT ($1{:}1_{10}$) = 70 NT (1:2) = 91	μm	(40 °C; 50 days) FO = 2500 (50 d) T (10:1) = 1100 (50 d) NT (10:1) 3429 (50 d) T (5:1) = 1166 (30 d) NT($1{:}1_{25}$) = 1433 (30 d) NT ($1{:}1_{10}$) = 1900 (30 d) NT (1:2) = 1100 (20 d)		Wang et al. (2016)
Fish oil	Gel:GA (1:1 w/w)	Water	$T = 190\,°C$ *Regular spray drying (RSM) Solid content: 30%; fish: wall: 2:1 *Complex coacervation → spray drying (CCM) transglutaminase (0.2%/g gelatin) *Double-encapsulated microcapsules (DEM) Complex coacervation + 100 mL Gel/GA solution → spray drying	RSM = 86.9 CCM = 89.8 DEM = 93.7	μm RSM = 2.14 CCM = 4.5 DEM = 6.3	–		Yu et al. (2017)
Fish oil (15% OF Emul)	WPI	Water	$T = 100$–$160\,°C$	100:79.2 120:78.5 140:77.9 160:80.31	3.89 μm	100:82 120:90.1 140:84.9 160:81.25	–	Lavanya et al. (2020)
Fish oil	SA AG (3:1)	Water	$T = 180\,°C$ Wall:fish oil = 4:1 Feed flow rate: 1×10^3 mL/h Air flow rate: 35 m^3/h	49.7	μm	–	Based on propanal 436.4	Pang et al. (2017)
Fish oil (10%, 20%, & 30 %)	CSM/Ch (0.2%, 0.4%, and 0.6% w/v) CSM: Ch = 1:4; 1:1; 4:1	Water 1% acetic acid solution	$T = 120\,°C$ CSM:Ch; FO; polymer concentration (A) 1:1; 0.2; 30 (B) 1:1; 0.2; 10 (C) 4:1; 0.5; 30 (D) 1:1; 0.4; 20 Opt: 48: 52%; 0.23; 33	A: 80 B: 83 C: 90 D: 94.25 Opt: 90	μm A: 1.949 B:1.737 C: 1.586 D: 2.487 Opt: 2.025	40 °C; 14 days NFO = 554.5 EFO = 100	–	Kavousi, Fathi, and Goli (2017)

Fish oil (10%, 20%, & 30 %)	Cas SPI Gel HPW (0.5%, 1%, & 1.5% w/v)	Water	Emulsion preparation: By glass microfluidic device + addition of fungal protease $T = 170\,°C$ Gel: FO + 1.5 Gel GH: FO + 1 Gel + 1 HWP CG: FO + 0.5 Cas + 0.5 Gel	–	μm 70–90	Gel:1000 Gel-HPW:150 Cas-Gel: 25 FO: 150	–	Comunian et al. (2018)

AG, acacia gum; Alg, alginate; ALT, antioxidant combination of 8.6% (w/v) ascorbic acid, 5.2% (w/v) lecithin and 86.20% (w/v) tocopherol; β-LG, β-lactoglobulin fibril; Cas, casein; CCM, complex coacervation microcapsules; CG, cashew tree gum; Ch, chitosan; CP, casein–pectin (% (w/w)); CSM, cress seed mucilage; DE, dextrose equivalent; DEM, double-encapsulated microcapsules; EE, encapsulation efficiency; Ex, extract; en oil, encapsulated fish oil; F–A, fish oil–acetone; F–E, fish oil–ethanol; F–M, fish oil–methanol; FO, fish oil; FO + ALT, fish oil + ALT (antioxidant); F–W, fish oil–water; GA, gum arabic; Gel, gelatin; GG, guar gum; HPC, hydroxypropyl cellulose; HPH, high-pressure homogenization; HPMC, hydroxypropyl methylcellulose; HWP, hydrolyzed whey protein; MD, maltodextrin; MS, modified starch; Opt, optimum treatment; PC, hydroxypropyl cellulose; PH, protein hydrolysate; PO, MD + GA + PH (2:2:1); SP: PH; POP, primary oxidation products; RH, relative humidity; RSM, Regular spray-dried microcapsules; SA, sodium alginate; SC, sodium caseinate; SO, MD + GA + SC (2:2:1); SOP, secondary oxidation products; SPI, soy protein isolate; SPO, MD + GA + SC + PH (2:2:0.5:0.5); T, inlet temperature; TMβ–LG, thiol-modified β-lactoglobulin fibrils; US, ultrasound; UT, ultraturrax; WPI, whey protein isolate.

Results and discussion

Although, there are many papers dealing with the effect of encapsulation process on the stability and quality of the fish oil, it is difficult to provide conclusive results because different wall materials (proteins, carbohydrates, and their combination with various concentrations), encapsulation methods (spray drying, freeze drying, electrohydrodynamic processes, etc.), and storage conditions during experiments (time, access of light and oxygen, temperature, humidity, and the use or not use of antioxidants) are considered in published studies. Therefore, this critical review is an attempt to provide a general conclusion about fish oil encapsulation by relying on the results and data of previous research activities.

In this work, the techniques used to produce fish oil-loaded micro/nanocarriers were investigated in the following five groups: (a) spray-dried particles (method: spray drying); (b) freeze-dried particles (method: freeze drying); (c) electrospun fibers/electrosprayed particles (method: electrohydrodynamic processes); (d) supercritical antisolvent particles (SCAP); and (e) other techniques/carriers (methods: nanoliposomes, gelation, spray–freeze drying, and transglutaminase catalyzed cross-linking). As shown in Figure 1, the three most frequent methods for fish oil encapsulation were spray drying (42.86%; 18 out of 42 case studies), freeze drying (21.43%), and electrohydrodynamic (19.04%) methods, respectively.

For general comparison of these methods, encapsulation efficiency (EE) was used. EE is one of the most important indices in nano/microencapsulation which can be correlated with the oxidative stability of fish oil-loaded carriers. Meanwhile, the calculation method of EE is the same in different studies and unlike oxidative stability indices, there is not much fluctuation in EE measurements. Therefore, this parameter, as a criterion of encapsulation performance, was investigated in current work. Evaluation of the effect of encapsulation method on EE revealed that the best performance was obtained for electrohydrodynamic processes (Figure 2). However, the differences among this method and spray drying, freeze drying and "other" techniques was not significant ($p > 0.05$).

It should be noted that many factors such as the type and concentration of carrier agent, the ratio of fish oil to wall material, the feed formulation and the process conditions can affect the EE. In this regard, investigating the effect of the type of wall material (based on three groups: proteins, carbohydrates, and their combination) on EE did not show any significant differences ($p > 0.05$). However, the combination of polysaccharides–proteins and single protein-based particles provided the best and the worst performance in terms of EE, respectively (Figure 3). Overall, the interaction of different factors on EE of fish oil-loaded carriers makes it difficult to interpret the results and conclude on which method and/or wall material is more suitable for encapsulation of fish oil. A comparison between the different encapsulation methods and the effect of process parameters on the encapsulation performance of fish oil will be discussed as follow.

Table 2. Selected recent studies (2016–2020) on freeze-drying encapsulation of fish oil.

Source	Polymers	Solvent	Processing conditions	Encapsulation efficiency (%)	Size	Oxidative stability		Sensory evaluation	Ref.
						POP	SOP		
Kilka fish oil	Ch (0.5% w/v) MS (9.5% w/v)	Water Acetic acid	F1: Ch:MS = 0.5:9.5%w/v F2: Ch:MS = 1:9%w/v F3: Ch:MS = 1.5:8.5%w/v FO:wall solution = 1:4 w/w Freeze drying: −70 °C; 72 H	F1: 79.4 F2: 88 F3: 82	μm F1: 4.7 F2: 2.3 F3: 4.2	–	–	Bread with 0%, 1%, 2.5%, and 5% fish capsule B_0: 8.4 B_1: 7.4 $B_{2.5}$: 5 B_5: 4.6	Hasani et al. (2019)
Fish oil	designed N-lauroyl chitosan	Acetic acid (60 mL, 1.0%, v/v) + methanol (40 mL)	Fish oil:wall material (6:1 w/w)	62.6	μm	–	–	–	Chatterjee and Judeh (2016)
Kilka fish oil (EPA and DHA concentrate)	WPI + SC (4:1)	Water	−42 °C WUS: without ultrasound 180w–1 min 180w–3 min 380w–1 min	WUS: 68.2 180w–1 min: 88 180w–3 min: 91 380w–1 min:92.8 380w–3 min: 94.1	μm	Conjugated dienoic Oil–N_2 = 12.5 Oil–air = 887 WUS = 712 180 w–1 min–en = 562 180 w–3 min–en = 412 380 w–1 min–en = 387 380 w–3 min–en = 362	Trienoic acids Oil–N2 = 25 Oil–air = 3300 WUS = 3100 180w–1 min–en = 2800 180w–3 min–en = 2100 380w–1 min–en = 1600 380w–3 min–en = 1300	–	Hosseini et al. (2019)
Fish oil	GA (20% & 25% w/w)	Water	US treatment on emulsion (24 kHz–2 min) Freeze drying: −20 °C–24 h * Emulsifier-wall material 2–20; 2–25; 4–20; 4–25	2–20:81.77 2–25:85.29 4–20:81.79 4–25:87.17	–	Fish oil = 700 2–20 = 2400 2–25 = 1540 4–20 = 1000 4–25 = 1300	–	Overall acceptance of LMF (1st day → 14thday) Fish oil = 4 → 4 2–20 = 3.2 → 3 2–25 = 3.5 → 3.2 4–20 = 3.4 → 3 4–25 = 4.2 → 3.8	Moghadam et al. (2019)
FPH + fish oil	Fuc Inu WPC	Water	W_1/O & W_1/O/W_2 W_1/O: FPH (4 g) + NaCl (0.5 g) water (25.5 g) + PGPR (6.28 g) + fish oil (63.72 g) −80 °C 24 h *IW: In-WPC * IFW: In-Fuc-WPC * FW: Fuc-WPC	IW: 81.2 IFW:78.5 FW: 86.3	nm 536–842	Hydroperoxide (4 °C; 8 weeks) IW: 103 IFW:226.3 (6 weeks) FW: 198	Anisidine IW: 566 (5 weeks) IFW: 400 (8 weeks) FW: 257 (7 weeks)	Fishery taste of yoghurt Control: 3 IW: 6 IFW: 6.5 FW: 7	Jamshidi et al. (2018)
Fish liver oil (*Gadus morrhua*)	PG + Ch (2:1; 1:1; 1:2 (w/w)		Fish oil:GEO 75:25% Core:wall = 10:2 (w/w)) Feed formulation: E-LBLD electrostatic layer-by-layer deposition method HS: 4000 & 1000 rpm Frozen at −70 °C, 12 h Freeze dryer: −51 °C, 12 Pa; 72 h	PG:$Ch_{(2:1)}$ = 79 PG:$Ch_{(1:1)}$ = 86 PG:$Ch_{(1:2)}$ = 63	nm PG:$Ch_{(2:1)}$ = 96 PG:$Ch_{(1:1)}$ = 64 PG:$Ch_{(1:2)}$ = 271				Raeisi et al. (2019)
Fish oil (ω_3) (10–30%)	F-Gel (33–66%) GA F-Gel: GA 1: 2; 2:1; 1:1	Water	Complex coacervation: HS: 10,000–20,000 rpm), wall materials: 30% w/v cross linking agent 1% $CaCl_2$ freeze drying: −85 °C	Gel:ω_3:HS 33:10:10,000 = 69.7 66:30:10,000 = 82.7 66:10:10,000 = 82.8 49:10:15,000 = 98.41 33:30:20,000 = 98.38 33:10:20,000 = 98.25	Gel: ω_3: HS (nm)	66:10:10,000 = 27 66:20:15,000 = 298 66:30:20,000 = 278 33:10:10,000 = 114 33:20:15,000 = 1520 33:30:20,000 = 617 49.5:10:10,000 = 42 49.5:20:15,000 = 114 49.5:30:20,000 = 420	–	–	–

									Esfahani et al. (2019)
Fish oil (0.5%, 1%, 2%)	B-Gel: GA (1:1) (1–2%)	Water	Complex coacervation: HS: 9000 rpm freeze drying: −60 °C; 24 h	Wall–core (%) 1–1 = 62.6 1–0.5 = 55 1–2 = 63 1.5–1 = 65 1.5–2 = 77 1.5–0.5 = 53 2–1 = 61 2–0.5 = 53 2–2 = 63	>0.5 μm	–	–	Pomegranate juice control: 7.9 +0.04% capsule: 6.2 +0.07% capsule: 5.5 +0.1% capsule: 5	Habibi et al. (2017)
Fish oil	SA + AG (3:1)	Water	Wall:fish oil = 4:1 (1) −25 °C; 24 h; 20 Pa and (2) −50 °C; 36 h	72.6	μm		Based on propanal 472.7	7.13 (from 10)	Pang et al. (2017)

AG, acacia gum; B-gel, bovine-gelatin; Ch, chitosan; EE, encapsulation efficiency; E-LBLD, electrostatic layer-by-layer deposition method; F-gel, Fish gelatin; FPH, fish protein hydrolysate; Fuc, fucoidan; GA, gum arabic; GEO, garlic (*Allium sativum* L.) essential oil; HS, homogenization speed; Inu, inulin; LMF, low-fat probiotic fermented milk + encapsulated/non-encapsulated fish oil; MS, modified starch; PG, Persian gum; POP, primary oxidation products; SA, sodium alginate; SC, sodium caseinate; SOP, secondary oxidation products; WPC, whey protein concentrate; WPI, whey protein isolate.

Spray drying encapsulation of fish oil

Among the different encapsulation methods, spray drying process of oil-in-water (O/W) emulsions, as a low cost, reproducible, simple, and easy to scale-up procedure, is a common method to encapsulate fish oil (Jafari, Assadpoor, Bhandari, et al. 2008; Jafari, Assadpoor, He, et al. 2008). In this method, fish oil emulsion is sprayed in drying chamber and water content is rapidity evaporated based on the differences between the inlet and outlet air temperature of drying chamber (Figure 4(A)). The results of the current study showed that 42.86% (18 out of 42 case studies; Figure 2) of the investigated works were established based on this technique. Fish oil can be encapsulated in a relative wide range of biopolymers, feed formulations (emulsion or coacervation solution; different viscosities, solid concentrations, etc.), and operational conditions (inlet and outlet temperatures, feed flow rates, drying gas flow rates, and humidity, besides atomizer pressures and speeds). Optimization of these parameters has an important effect on a successful spray drying encapsulation of fish oil (Gharsallaoui et al. 2007; Jafari, Assadpoor, Bhandari, et al. 2008; Jafari, Assadpoor, He, et al. 2008; Nayak and Rastogi 2010; Botrel et al. 2017).

As shown in Table 1 and Figure 5, various biopolymers have been used as wall materials for the encapsulation of fish oil via spray drying. Carbohydrates are more popular than proteins (27.3% and 19.7%, respectively) for this purpose, due to poor heat stability of proteins which leads to droplet aggregation and disruption of the emulsion stability as well as sensitivity of proteins to deterioration by organic solvents (Sliwinski et al. 2003). Maltodextrin (six out of 18 case studies) and GA (six out of 18 studies), either alone or in combination with other biopolymers, were chiefly used as a wall material. Relatively low cost, good solubility, film forming properties, and providing a low viscosity at high solid concentrations were some of the reasons for selecting the maltodextrin (MD) as a suitable wall material for encapsulation of bioactive compounds such as fish oil (Nayak and Rastogi 2010; Premi and Sharma 2017). However, the poor emulsifying properties, low oil retention, and weak emulsion stability of MD is limiting the use of this biopolymer as a single wall material for encapsulation of fish oil (Kagami et al. 2003).

On the other hand, GA exhibits a good emulsifying capacity and even in relatively high concentrations (5–20% w/v), provides an acceptable viscosity for atomization and subsequently, results in an adequate thickness of microparticle wall. Therefore, this biopolymer has attracted the attention of many researchers. But, this gum has a high price and shows almost high permeability to oxygen (Charve and Reineccius 2009). In this regard, Vaucher et al. (2019) used the combination of MD and GA to encapsulate fish oil. Depending on the ratio of these biopolymers, EE was reported in the range of 51.2–56.8% (Table 1). Moreover, the oxidation of samples with a higher MD content were lower than other samples which can be related to capability of MD to control lipid oxidation by its film forming properties (Vaucher et al. 2019). Proteins also show good emulsifying properties. Therefore, the combination of carbohydrates,

Table 3. Overview of recent studies (2016–2020) on encapsulation of fish oil based on electrohydrodynamic processes.

Source	Polymers	Solvent	Processing conditions	Encapsulation efficiency (%)	Size	Oxidative stability POP (%)	Ref.
Fish oil (30% w/w of zein)	Core: zein (25% w/v) Shell: PVP 27–31% w/v Zein:FO = 2:1	Ethanol:water 80:20–100:0 w/w	Coaxial			electrospinning 0.2 mL/h 16 kV 14 cm Best conditions: PVP: 31% & 100% ethanol	96.9
nm (fiber) 250–830 (mean: 560)	45 °C–25 days: anaerobic condition Fish oil: 252 Single & coaxial fiber: 184 & 168 45 °C–5 days: aerobic condition Fish oil: 18,400 Single & coaxial fiber: 2040 & 2000	Yang et al. (2017)					
Highly DHA-enriched fish oil (85 wt.%)	Zein (4.5 wt.%) Zein: FO (2:1 wt.)	Ethanol	EAPG: 10 L/min 20 kV	84	1.4 μm (capsule)	*RH: 0%; *T*: 5 °C; air; darkness; 40 days Fish oil: 2800 (18 days) Capsules: 1400 (18 days) *RH: 0%; *T*: 23 °C; air; light; 18 days Fish oil: 3000 (8 days) Capsules: 1266 (8 days) *RH: 65%; *T*: 23 °C; vacuum; light; 18 days Fish oil: 1200 (2 days) Capsules: 133 (20 days)	Busolo et al. (2019)
Cod liver oil	Zein (5% wt.) Zein: FO = (4:1 wt)	Ethanol:water 85:15	EAPG: 1.4 mL/min 10 kV	83	2–3 μm (capsule)	20 °C–35 days Capsule: 3400 (3 days) Fish oil: 1240 (21 days)	Miguel et al. (2019)
Cod liver oil (5%, 7.5%, & 10% wt. of emulsion)	PVA* (10.5%wt)	Water	* Emulsion: pH: 2 (0.1 N HCl) Emulsifier: WPI (1% wt); FPH (2% wt) *			Electrospinning: 0.2 mL/h, 20 kV, 10 cm	92.4
nm (fiber) PVA + Oil (wt %)	10.5 + 1.5 = 163 9 + 2 = 120 7.5 + 2.5 = 120	PV (%):				40 °C–14 days–darkness FO: 4575 (9 days) FO, N_2: 4425 (14 days) Fiber = 98,425 (4 days) Fiber, N_2 = 76,000 (4 days)	García-Moreno et al. (2016)
Fish oil (20 wt.% of biopolymer)	Pul + Dex_{70} (15 + 25 wt.%) WPC (10–30% wt.) = control	Water	Stabilizer: 1 wt.% WPI-pH 7. 0.01 mL/min 20 kV 15 cm	* Pul: EFO: 69 NFO: 90 * Dex: EFO: 68 NFO: 76	* Dextran capsules (nm) EFO: 300–600 NFO: 600–900 Control: 300–600	* Pul: E-FO:1240 NFO: 800 * Dex: EFO:43115 NFO: 1060	García-Moreno et al. (2017)

					* Pullulan fiber EFO: 700–800 NFO: 900–1000 Control: 500–600		
Fish oil (20 wt.% of biopolymer)	Pul (1–5%) Dex (15% or 20%) Glu (15%) WPC (1% wt.)	–				WPC + Pul + Dex = D WPC + Pul + Glu = G *Pilot plant scale 1.5–1.8 mL/min 10–15 kV; 15 cm *HPH: 9000 psi *RSE	D-HPH: 86.9 D-RSE: 91.7 G-HPH:78.1 G-RSE:85.7
μm (capsule) D-HPH: 1.76 D-RSE: 2.55 G-HPH: 3.01 G-RSE: 3.96	D-HPH: 430 D-RSE: 250 G-HPH:230 G-RSE:200	García-Moreno et al. (2018)					
Fish oil	WPI: 10% MD: 2%	Water	* HPH: 1000 & 2000 bar 1–8 passes * Electrospray: 0.02 mL/min; 18 KV; 13 cm	1000: 60–80 2000:70–98	nm (capsule) 1000/2: 974 1000/4:501 2000/2:635 2000/4:198	–	Paximada, Howarth, and Dubey (2018)
Fish oil (20% wt.)	WPC + Pul + Glu (0.5 + 4 + 15 %wt.) Pul + dextran (1 + 15)	Water	HPP = 9000 psi			0.003–0.010 mL/min 15–20 kV	–
μm (capsule) $D = 1.26$ $G = 1.39$	Peak-to peak (%) $D = 7000$ $G = 14{,}800$ –	Boerekamp et al. (2019)					

a: (0) no difference against reference; (1) little difference against reference; (3) clear difference against reference; (5) big difference against reference; Dex, dextran; EAPG, electrospraying assisted by pressurized gas; EE, encapsulation efficiency; EFO, emulsified fish oil; FO, fish oil; FPH, fish protein hydrolysate; Glu, glucose syrup; HPH, high-pressure homogenization; NFO, neat fish oil; Pul, Pullulan; PVA, poly(vinyl alcohol); PVP, polyvinylpyrrolidone; RSE, rotor-stator emulsification; WPC, whey protein concentrate; WPI, whey protein isolate

Table 4. Some studies performed on encapsulation of fish oil based on supercritical antisolvent (CO_2) process.

Source	Polymer	Solvent	Processing conditions	Encapsulation efficiency (%)	Size	Oxidative stability: POP	Oxidative stability: SOP	Ref.
Fish oil (20–30% omega-3)	HPMC	Water	Optimum condition: Feed rate: 1.36 mL/min; 150 bar; 60 °C; AF1 = solid content: 10.2; AF2 = solid content: 9; AF3 = solid content: 7.75; AF4 = solid content: 6.5	AF1 = 81.7; AF2 = 75.3; AF3 = 71.2; AF1 = 69.5	μm; AF1 = 3.75; AF2 = 7.24; AF3 = 12.7; AF1 = 13.7	FO: 111 (PV0: 18); AF1 = 150 (PV0: 2); AF2 = 160; AF3 = 275; AF1 = 500		Karim et al. (2017)
Fish oil	PCL	Acetone; Water	+ α-tocopherol; Liquid-cooled CO_2 at 263 K; 0.8 MPa; R: PCL + Sol + Tur; I: PCL + Sol + Gly; O: PCL + Sol + Xn + Tur	R: 43; I: 38; O: 12	nm; R: 73; I: 8; O: 6	–	–	Prieto and Calvo (2017)
Fish oil (10–50% w/w)	Liquid FHSO	–	Fish oil-loaded hollow solid lipid micro/nanoparticles; 200 bar; 57 °C	$FO_{(10\%)}$: 80.2; $FO_{(20\%)}$: 93.2; $FO_{(30\%)}$: 85; $FO_{(40\%)}$: 92; $FO_{(50\%)}$: 97.5	μm; 5–18	$FO_{(Free)}$: 1661 (10 d); $FO_{(10\%)}$:2523 (23 d); $FO_{(20\%)}$:2703 (17 d); $FO_{(30\%)}$:3070 (13 d); $FO_{(40\%)}$: 2873 (8 d); $FO_{(50\%)}$:2508(6 d)	$FO_{(Free)}$: 1158 (12 d); $FO_{(10\%)}$:1668 (23 d); $FO_{(20\%)}$:1668 (15 d); $FO_{(30\%)}$: 2740 (15 d); $FO_{(40\%)}$: 1600 (12 d); $FO_{(50\%)}$: 1450 (9 d)	Yang and Ciftci (2017)

FHSO, fully hydrogenated soybean oil; Gly, glycerol; HPMC, hydroxypropyl methyl cellulose; PCL, polycaprolactone; PG, prosopis alba exudate gum; Sol, solvent; Tur, turbidity; Xn, xanthan; Y, control sample (yogurt); Y + FO, yogurt enriched with free form of fish oil; Y + NFO, yogurt enriched with nano-liposomal encapsulated fish oil.

especially MD with low price and good functional properties, with various proteins have been reported in several previous studies (Charve and Reineccius 2009; Abd Ghani et al. 2017; Binsi, Nayak, et al. 2017; Chang et al. 2020; Linke, Weiss, and Kohlus 2020).

Moreover, dextrose equivalence (DE) of MD can influence on the EE of fish oil. Abd Ghani et al. (2017) investigated the effect of various DEs (11, 19, and 25) of MD on powder structure, surface–oil ratio, and oxidative stability of fish oil. The results showed that by increasing DE value, the EE and oxidative stability of fish oil are increased. They also found that particles made by a higher DE (19 and 25) had spherical shapes and smooth surfaces. MD with a high DE may have a plasticizer role and subsequently, inhibit the shrinkage of particles (Gharsallaoui et al. 2007; Abd Ghani et al. 2017). Also, the large vacuole diameter and dents observed in powders prepared with MD of lower DE resulted in the higher surface–oil ratio for the powders and a lower EE.

Due to the important role of the wall materials on the encapsulation performance, the effort to find a new source of carrier agent is still interesting for researchers. Botrel et al. (2017) compared the effect of cashew tree gum, as an innovative wall material, with conventional materials. They found that this new carrier agent has a good potential to encapsulate fish oil (Botrel et al. 2017). Kavousi, Fathi, and Goli (2017) used cress seed mucilage and hydrogels to fabricate fish oil microparticles. Their results revealed that developed carriers show a great potential for encapsulation of fish oil to protect it against oxidation (Kavousi, Fathi, and Goli 2017).

Another parameter which may affect the encapsulation performance of fish oil is the rate of fish oil to the wall material. For example, Linke, Weiss, and Kohlus (2020) confirmed that by increasing the ratio of wall material (soy protein isolate) to fish oil, the EE and oxidation stability was increased (Linke, Weiss, and Kohlus 2020). The solids content also has an influence on the encapsulation performance. Wang et al. (2016) reported that when the solids content reached from 10% to 25%, the EE increased from 70% to 72%. Moreover, when they increased the ratio of wall: fish oil from 1:1 to 2:1 and solids content was 25%, the EE reached to 91%. Non-encapsulated (surface) oil content of micro/nanoparticles is directly correlated with the oxidation and results in unpleasant flavors; because the surface oil has a more sensitivity to environmental oxygen and external conditions. Therefore, by increasing EE, the oxidative stability improves (Wang et al. 2016). Although, a high concentration of solids can decline migration of fish oil onto the surface of the microparticles. However, the concentration employed for encapsulation of fish oil should not cause a high viscosity to avoid clogging of the atomization nozzle during spray drying (Vaucher et al. 2019).

As previously mentioned, besides the type and concentration of wall material, formulation and preparation conditions of the feed solution have an important effect on the encapsulation performance of fish oil via spray drying. Vaucher et al. (2019) investigated the effect of homogenization pressure on

Table 5. Selected studies performed on encapsulation of fish oil based on other techniques.

Source	Polymers	Solvent	Processing conditions	Encapsulation efficiency (%)	Size	Oxidative stability		Sensory evaluation	Ref.
						POP	SOP		
Spray- freeze drying									
Fish oil	SA + AG (3:1)	Water	Spray–freeze drying Flow rate: 15 mL/min Vacuum pressure: 20 bar Atomization pressure: 5 bar Freeze drying: 20 bar; −65 °C	90.8	μm	–	Based on propanal 336.4	9.5 (from 10)	Pang et al. (2017)
Ionic Gelation									
Fish oil (10%)	Alg + Ch (1% + 2%) Alg + Ch + PG (1% + 2% + 2%)	Water	$CaCl_2$: cross-linking agent Vacuum drying	Alg + Ch: 89.95 Alg + Ch + PG: 98.63	mm Alg + Ch: 1.14 Alg + Ch + PG: 1.49	–	–	–	Vasile, Judis, and Mazzobre (2017)
Nanoliposomes									
Fish oil	Lecithin, sunflower oil	Water	Homogenizing for 10 min by a rotor–stator Homogenizer Sonication: 7 min: 20 kHz (1 s: On and 1 s: Off)	92.22	nm 300–500	4 °C; 21 days Y + FO: 77.8 Y + NFO: 7.7	–	Yogurt Y: 4.72 (from 5) Y + FO: 2.65 Y + NFO: 4.23	Ghorbanzade et al. (2017)
TG									
Fish oil (20%)	SPI: Inu (0.5, 0.6, 1, 1.4)	Water	Complex coacervation Turbidity Transglutaminase (10 U/g protein	Inulin: SPI; Fish oil 1.4; 71.2 = 36.13 0.6; 28.8 = 64.07 0.6; 71.2 = 69.3 0.4; 20 = 94%	μm <100	–	• –	–	Rios-Mera et al. (2019)

AG, acacia gum; Alg, alginate; Ch, chitosan; Inu, inulin; PG, prosopis alba exudate gum; SA, sodium alginate; SPI, soy protein isolate;

the oxidative stability of encapsulated fish oil. They reported that by increasing the pressure from 0.5 to 12.5 MPa, the oxidation rate decreased. However, at pressures >12.5 MPa, a higher oxidation was observed. The oxidative stability of particles is directly related to EE and by increasing pressure, this factor increases. However, Di Giorgio, Salgado, and Mauri (2019) did not recognize any clear relationship between ratio of fish oil to wall material and EE (Di Giorgio, Salgado, and Mauri 2019).

In encapsulation of fish oil, feed solution is usually in the form of O/W (wall materials solution) emulsions. However, in aqueous medium, hydrocolloid(s), may be unstable. Therefore, in recent years some researchers produced the spray-dried fish oil microparticles using “simple/complex coacervation,” which is considered as a simple, scalable, low-cost, reproducible, and one of the most effective methods to encapsulate hydrophobic bioactive compounds and produce delivery systems based on the nature of liquids (Patrickab et al. 2013; Ghasemi et al. 2017; Rajabi et al. 2019). Coacervation encapsulation (simple and complex) is the phase separation of one or many biopolymers from the initial solution due to opposite charges to form coacervates phase deposited around the active agent as a core (Gouin 2004). In this regard, Yu et al. (2017) investigated the physical properties and thermal stability of fish oil-loaded microcapsules developed by various encapsulation techniques as followed: directly spray-dried microcapsules, complex coacervation microcapsules, and double-encapsulated microcapsules based on gelatin/GA. The results showed that the EE of complex coacervation microcapsules was more than emulsion-based microparticles (Table 1). However, the best result was obtained for double-encapsulated microcapsules (93.7%), due to the second covering layer of the surface oil. So, the highest moisture content and largest particle size was observed in these particles. However, the average size of all microcapsules was 2.14–6.31 μm in diameter. The directly spray-dried microcapsules exhibited spherical shapes with smooth surfaces and lots of pores. However, the surfaces of most coacervate microcapsules were collapsed (with wrinkles). The double-encapsulated microspheres had spherical shapes with no wrinkles, due to the coating by second layer. Moreover, the SEM images showed that the shell of directly spray-dried microspheres consisted air holes or oil droplets structure and the droplet of fish oil adhered to the shell of microparticles which result in multiple-core (matrix or monolithic) microcapsules, while complex coacervation formed single-core (reservoir) that was included an outer shell surrounding the (bio)active compound (Yu et al. 2017).

Moreover, Vaucher et al. (2019) showed that microcapsules obtained by complex coacervation between a protein (casein) and two polysaccharides (MD and pectin) followed by spray drying provided a higher EE and oxidative stability as compared to those prepared with simple emulsification followed by spray drying. Complex coacervation produced a thicker outer shell than the emulsion-based particles which decreased the surface oil and consequently, the oxidation of fish oil. They introduced this process as a more cost-effective method than emulsion-based particles (Vaucher et al.

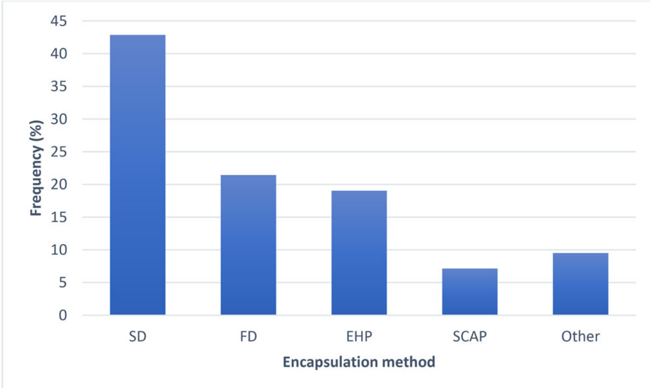

Figure 1. Frequency of fish oil encapsulation methods. SD, spray drying; FD, freeze drying; EHP, electrohydrodynamic processes; SCAP, supercritical antisolvent process.

2019). Unlike Yu et al. (2017), these authors showed that complex coacervation resulted in particles with smaller size than directly spray-dried microcapsules. They attributed this phenomenon to the higher concentration of polymers in directly spray-dried microcapsules.

Outlet and inlet air temperature is another important parameter during encapsulation of fish oil via spray drying that should be taken into consideration (Arpagaus et al. 2018; Assadpour and Jafari 2019a, 2019b). The previous studies have reported that the raise of the inlet air temperature can negatively affect the oxidative stability of encapsulated fish oil (Jafari, Assadpoor, Bhandari, et al. 2008). The results of several studies have shown that oil droplets underwent certain extent of oxidation during spray drying because of relatively high inlet temperatures (Binsi, Nayak, et al. 2017; Linke, Weiss, and Kohlus 2020). Due to the importance of process temperature on the encapsulation performance, optimization of this factor is still one of the most frequent research fields. In this regard, Chang et al. (2020) estimated the effect of inlet air temperatures (160–180 °C) on the fish oil microencapsulation process (Chang et al. 2020). The EE decreased at higher inlet air temperatures. Moreover, oxidation of fish oil was increased in high temperatures. As a result, encapsulation of fish oil has usually been suggested in 160–170 °C (11 out of 18 studies: 61.1%). Higher wall material concentration can protect fish oil against the high temperature during the spray drying. Also, high molecular packing inhibits the diffusion of oxygen into the spray-dried particles (García-Tejeda et al. 2016). Encina et al. (2018) changed the solvent of encapsulation solution to decrease the inlet temperature during spray drying; the use of acetone decreased the inlet temperature to 78 °C and the highest EE was obtained in this condition (Encina et al. 2018). Binsi, Nayak, et al. (2017) investigated the synergistic effect of sage extract and GA to protect fish oil against heat and oxygen induced deterioration. Encapsulation of fish oil with GA and sage extract showed a higher EE and oxidative stability than capsules containing GA alone (Binsi, Nayak, et al. 2017).

It is worth mentioning that the particle size significantly affects the application and performance of micro/nanocarriers. The normal spray drying technique is not capable to produce nanoparticles (Table 1); however, a new developed device known as nano spray drier can result in bioactive-loaded nanoparticles (Arpagaus et al. 2018, Assadpour and Jafari 2019a, 2019b). The control of size is an important challenge for developing particles by spray dryer. The carrier size can be affected by different factors such as atomizing pressure, nozzle size and position, viscosity, and flow rate of the feed solution (Mahdavi et al. 2014).

Freeze drying encapsulation of fish oil

The spray-dried microcapsules show a poor solubility and consequently, the application of them is limited in dry food products (Jensen et al. 2010). On the other hand, the oxidative stability of these particles is usually lower than neat fish oil. In addition, drying at high temperature air (100–240 °C) can lead to initial oxidation of the fish oil (Nielsen and Jacobsen 2009). So, freeze drying process can be suitable for the encapsulation of heat-sensitive bioactive compounds (Jafari, Mahdavi-Khazaei, and Hemmati-Kakhki 2016). As shown in Figure 4(B), encapsulation of fish oil by freeze drying is usually achieved by emulsifying this core in appropriated wall material matrix systems and then co-lyophilizing and sublimation of ice crystals (Fang and Bhandari 2012). Freeze drying results in formation of pores and enhancement of the rehydration feature of capsules. On the other hand, it is believed that oxygen can diffuse faster through these pores and they are capable of scavenging the oxygen. Hence, freeze-drying process may not provide a very effective protection against oxidation (Anandharamakrishnan, Rielly, and Stapley 2010).

For the last reason, despite the advantages of freeze-drying method, the use of this technique has not received much attention for encapsulation of oxygen sensitive compounds such as fish oil. However, the point to note is that in most of the studies, the oxidative stability of freeze-dried microparticles was not investigated.

The results of the current review showed that 21.43% (the 2nd most frequent method for fish encapsulation; Figure 2) of studies were established based on freeze drying method, in which, the effect of the type and concentration of wall composition, the ratio of shell: core and feed preparation conditions on the encapsulation performance of freeze-dried fish oil particles were investigated. In this method, carbohydrates were also the most common wall material (70.6%). The results showed that the wall material of freeze-dried microparticles can be made from a carbohydrate material (22.2%; two out of nine studies), the combination of two or more carbohydrates (33.3%: three out of nine studies), the combination of two proteins (11.1%: one out of nine studies), and the combination of protein and carbohydrate materials (33.3%: three out of nine studies). The most common wall martials were gums (29.4%: 17.65% GA and 11.76% other gums), chitosan (17.65%), milk proteins (17.65%), gelatin (11.76%), and other biopolymers (23.53%: fucoidan, inulin, modified starch and sodium alginate). Suitable solubility, low viscosity at high concentrations, emulsifying properties, and the high holding capacity of oil droplets besides low price have caused gums especially GA to be widely noticed for nano/microencapsulation of fish oil by freeze drying (Shiga et al. 2001, 2004; Esfahani et al. 2019; Raeisi et al. 2019). Proteins are also considered as great encapsulating agents. Milk proteins are natural amphiphilic biopolymers which can be used as hydrophilic emulsifiers.

Several studies revealed that the combination of polysaccharides and proteins can offer the best encapsulation performance. For instance, Jamshidi et al. (2018) applied double emulsions followed by freeze drying to encapsulate fish protein hydrolysate (FPH) and fish oil based on three different combination of wall materials; whey protein concentrate (WPC) with inulin (Inu) and fucoidan (Fuc). The results showed that the highest EE and solubility were obtained in Fuc–WPC microcapsules. Furthermore, the combination of Fuc–WPC could delay the formation of the secondary oxidation products and increase the oxidative stability of encapsulated fish oil. The Inu–WPC samples exhibited a gradual increase up to five weeks followed by a decrease trend. Moreover, a slight increase in *p*-anisidin values were observed after 7 weeks storage in the samples containing fucoidan (Inu + Fuc–WPC and Fuc–WPC samples). The antioxidant attributes of Fuc besides the difference of surface and encapsulated oil between various microcapsules are considered as two possible reasons for such trends. Moreover, the combination of Inu–Fuc in Inu + Fuc–WPC samples resulted in higher negative charges of microcapsules which increased the chance of ionic metals absorption (with positive charges) and caused fatty acid degradation (Alemán et al. 2015; Jamshidi et al. 2018). The combination of gelatin and GA also showed a good potential for encapsulation of fish oil depending on the concentration of fish oil and each biopolymer besides homogenization speed used for formation of emulsions (Habibi et al. 2017; Esfahani et al. 2019).

Moghadam et al. (2019) investigated the effect of the concentration of wall material and emulsifier on the encapsulation performance and sensory properties of low-fat milk enriched with fish oil. The results exhibited that encapsulated fish with 25% GA and 4% emulsifier had a higher EE than samples with 20% wall material and 2% emulsifier (Table 2). By increasing the viscosity of continuous phase, the mobility of the droplets are decreased which provides the required time for adsorption of the emulsifier onto the surface of the oil droplets and so, increases the EE of the capsules (Makri and Doxastakis 2006). Moreover, the use of nanoencapsulated fish oil, especially the samples prepared with 25% GA and 4% emulsifier, did not have any adverse effects on the sensory acceptance of fermented low-fat milk (Moghadam et al. 2019).

Investigation of the feed preparation conditions by Hosseini et al. (2019) revealed that sonication treatment by decreasing the presence of oil at the surface of particles and creating finer pores may control the oxidation of encapsulated oil. These authors encapsulated fish oil by a mixture of whey powder and sodium caseinate (4:1) *via* freeze drying. The emulsification was carried out by ultrasound treatment (power: 180–380 W; time: 1–3 min). Sonication treatment resulted in finer and higher pores as compared to control

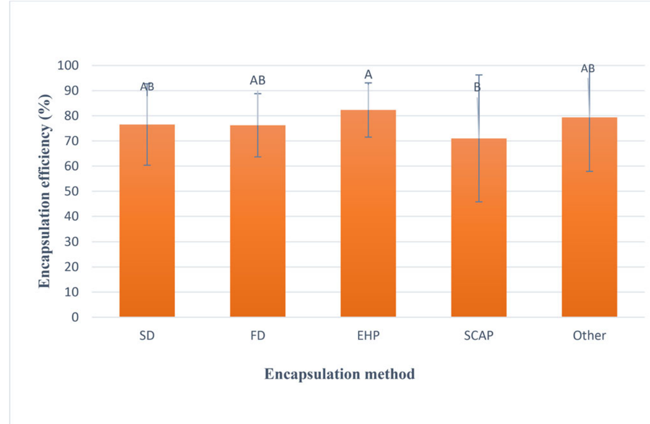

Figure 2. The effect of encapsulation method on fish oil encapsulation efficiency. SD, spray drying; FD, freeze drying; EHP, electrohydrodynamic processes; SCAP, supercritical antisolvent process.

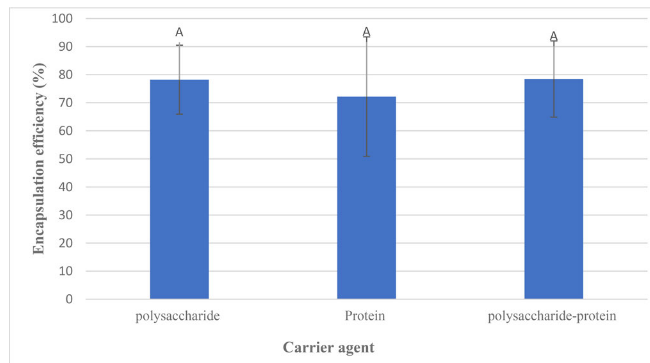

Figure 3. The effect of carrier type on fish oil encapsulation efficiency.

samples (microcapsules prepared without sonication). However, such pores were merged into larger pores or destroyed during storage (40 °C for 12 days). This phenomenon may be related to the reaction of vitamins, amino acids, and proteins with oxidation products (peroxides and free radicals) during storage time which could affect the texture of the particles (Hosseini et al. 2019; Khoshnoudi-Nia and Sedaghat 2019). In this regard, Karthik and Anandharamakrishnan (2013) also reported that the matrix changing from glassy to rubbery state by decreasing the moisture content results in the reduction of pore size in encapsulated oil (Karthik and Anandharamakrishnan 2013). The sonication (380 W; 3 min) had also a positive effect on EE; 94.1% vs. 68% for control sample, as shown in Table 2. In this study, the highest oxidation was observed for unencapsulated fish oil stored in air atmosphere followed by the control samples and the powders from sonication treatment, respectively; as well as the lowest one obtained for unencapsulated product storage under N_2 (Hosseini et al. 2019). Therefore, sometimes, the storage condition of fish oil may be more effective than the encapsulation process to control the oxidation. Furthermore, Esfahani et al. (2019) used complex coacervation followed by freeze drying to encapsulate fish oil. The homogenization speed had a significant effect on EE of fish oil. In fact, by increasing mechanical energy, particle size will increase and surface area of the system decreases due to recoalescence of emulsion droplets.

Therefore, more oil droplets are entrapped within fish gelatin–GA capsules. The mechanical stress, also leads to rupture of the capsule membranes and increasing aggregation, which causes hydrophobic interactions and combination of near capsules (Liu, Low, and Nickerson 2010). This method resulted in a high EE (98.4%) in optimized conditions as fish gelatin: GA (33: 67%), fish oil: 30%, and homogenization speed: 20,000 rpm (Esfahani et al. 2019).

The morphology of freeze-dried powders is non-uniform in terms of particle size which originates from the grinding or agglomeration of the material after drying, thus there is no particle size control (Ozkan et al. 2019). Additionally, the presence of porosity in all samples is possible due to sublimation of ice crystals, which are formed in the freezing step. The typical morphology of freeze-dried particles is of thin sheets with a porous and rough continuous surface (Machado et al. 2014; da Fonseca Machado et al. 2018).

The morphology of freeze-dried particles was usually smooth, non-uniform, and fragile with a porous structure. Non-uniformity and inhomogeneous structures are due to the grinding, sieving, and/or agglomeration of the particles after drying, thus there is no particle size control (Table 2). Additionally, the porosity is probably due to sublimation of ice crystals, which are formed during freezing steps. The typical morphology of freeze-dried particles is of thin sheets with a porous and rough continuous surface (Chatterjee and Judeh 2016; Jamshidi et al. 2018; Hasani et al. 2019; Raeisi et al. 2019)

The sensory properties of products containing fish oil capsules are very important; however, most studies have not addressed this feature. Hasani et al. (2019) developed a functional bread enriched with fish oil, ω-3 fatty acids. They microencapsulated fish oil with chitosan and modified starch in various ratios (Table 2) *via* freeze drying. The combination of chitosan:modified starch (1%:9%) introduced as the best formulation of wall materials for encapsulation of fish oil and the EE of this sample was significantly higher than other samples. This microcapsule was added to bread at the concentration of 0–5% w/w. Results showed that enriched bread with 5% encapsulated fish oil, had the highest value in crust, color, and firmness and the sensory properties of this bread was significantly lower than control sample and breads containing 1% fish oil (Hasani et al. 2019).

In general, spray drying is considered as a low cost and relatively fast method to produce microparticles. Nevertheless, due to high drying temperatures, this method is not suitable for encapsulation of heat-sensitive products. On the other hand, freeze drying can remove the water by sublimation under vacuum. However, it is a time consuming and expensive method. Spray–freeze drying method can combine the advantages of both methods to produce microparticles without heat damage and protect powders against agglomeration (Semyonov et al. 2010). In this regard, Pang et al. (2017) compared the performance of spray drying, freeze drying, and spray–freeze drying to encapsulate fish oil. The results revealed that the lowest and highest yields and EE were obtained for spray drying (20.9% and 49.7%, respectively) and spray–freeze drying (95.07% and 90.8%) methods (Tables 2 and 4). The low drying temperature during spray–freeze drying and freeze-drying techniques effectively avoid exhausting and evaporation of products and consequently, causing a high yield. In addition, the freeze-dried particles exhibited the lowest oxidative stability while the highest level was obtained in spray–freeze-dried microparticles. Therefore, the sensory score of freeze-dried capsules was lower than other particles. Irregular and porous structures besides higher surface oil content of freeze-dried particles are the possible reasons for this phenomenon (Pang et al. 2017). Therefore, spray–freeze drying may be proposed as a suitable and cost-effective method to produce fish oil-loaded microparticles. However, further studies are needed to optimize the feed formulation and operational conditions.

Electrohydrodynamic processes for encapsulation of fish oil

Although, spray drying is a simple and relatively inexpensive method and consequently, common to produce fish oil-loaded nano/microparticles, the application of these particles is limited in dry food products, because of their poor solubility (Drusch and Berg 2008). Moreover, the spray drying method is carried out under air at high temperatures (160–240 °C), which intensifies the oxidation of fish oil; then, drying under inert atmosphere is needed (Eratte et al. 2016). The deposition of produced powders in outlet pipe and chamber wall is another challenge in spray drying method. On the other hand, freeze drying may produce the particles with a better solubility. However, porous and irregular structure of freeze-dried particles may increase the extent of oxidative reactions which is a limiting factor in the application of this method for producing encapsulated fish oil particles (Anandharamakrishnan, Rielly, and Stapley 2010). Moreover, this technique has a low yield and it is costly and time consuming. Therefore, the development of alternative delivery systems for fish oil are required. In this context, electrohydrodynamic processing (EHDP), including both electrospinning (product: fibers) and electrospraying (product: capsules) techniques, is an innovative and straightforward encapsulation technique which can be applied particularly to produce nano–micro carriers containing sensitive bioactive compounds such as fish oil (García-Moreno et al. 2016; Jacobsen et al. 2018; Busolo et al. 2019).

As shown in Figure 4(C), in this technique, a high-voltage electrostatic field (as a power source) is used to charge the surface of a feed droplet formed at the end of a syringe needle. These ultrathin droplets are drawn from the spinneret tip to a grounded collector by an external electric field to produce nano/micro capsules/fibers after solidification (Rostami et al. 2019; Sharif, Khoshnoudi-Nia, and Jafari 2020, Soleimanifar, Jafari, and Assadpour 2020). Mutual charge repulsion results in a force directly opposite to the surface tension and gives a conical shape to droplet (i.e. Taylor cone). By overcoming electrical forces on surface tension, an electrically charged jet is stretched out because of several instabilities (such as bending motions and/or whipping) which helps to evaporate the solvent and produces

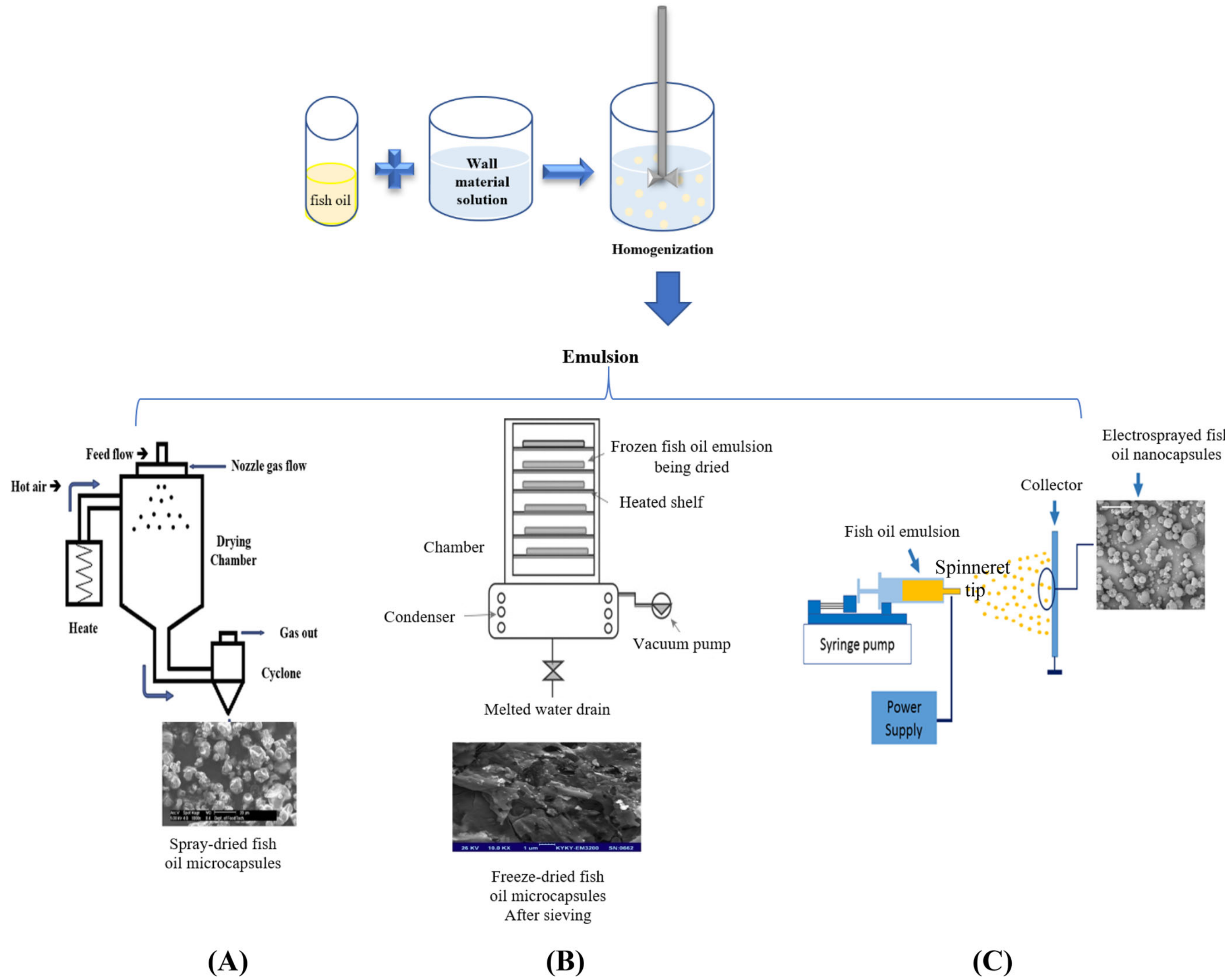

Figure 4. Schematic of fish oil encapsulation by: (A) spray drying; (B) freeze drying, and (C) electrospraying method.

dried fibers/capsules (Frenot and Chronakis 2003; Köse, Başpınar, and Bayraktar 2019).

Electrohydrodynamic process provides several advantages including (Jia et al. 2002; Khoshnoudi-Nia, Sharif, and Jafari 2020): (i) it is a non-thermal method, thus avoiding deterioration of the thermo-sensitive bioactive compounds; and (ii) the size of the carriers obtained by electrohydrodynamic method is small (as compared to spray-dried and freeze-dried particles with 10–100 μm) which allows their incorporation into enriched food systems without negative effects on the organoleptic properties of the final product. In recent years, encapsulation of fish oil via electrohydrodynamic techniques is a field which has attracted the attention of researchers (19.04% of studies were based electrohydrodynamic method, Figure 2).

The effects of wall material composition (type and concentration of biopolymers), electrohydrodynamic process, and feed preparation conditions are the most important factors investigated recently. In contrast to the previous methods (spray/freeze drying), proteins are frequently used to encapsulate fish oil by using electrohydrodynamic methods; proteins: 37.5%; carbohydrates: 12.5%; combination of proteins and carbohydrates: 37.5%; synthetic polymers (such as polyvinyl alcohol, PVA): 12.5% (Figure 5). Based on Table 3, the common proteins applied for fish oil encapsulation include dairy proteins (25%: 4 out of 16 cases) and zein (18.75%: 3 out of 16 cases). Several polysaccharides, such as pullulan (18.75%: 3 out of 16 carriers), dextran (18.75%: 3 out of 16 carriers), glucose syrup (12.5%: 2 out of 16 carriers); and MD (6.25%: 1 out of 16 carriers) have also been used as wall materials for the production of fish oil delivery systems by electrohydrodynamic processes. Zein, generally recognized as safe (GRAS), can provide several advantages including biocompatibility, high thermal resistance, hydrophobicity (low moisture absorption), film-forming, oxygen barrier, and antioxidant properties (Gezer et al. 2015), which makes it as an attractive wall material for encapsulation of bioactive compounds *via* electrohydrodynamic processes. Various carrier systems (e.g. nano/microcapsules or fibers, films, hydrogels, etc.) based on zein have exhibited good potentials in terms of delivery efficiency, stability, protection, and release properties (Zhang et al. 2015; Yang et al. 2017; Busolo et al. 2019; Miguel et al. 2019). However, the food applications of zein is restricted due to the high cost of this biopolymer. The lack of solubility in water and yellowish color are some other limitations of this compound. Moreover, the use of low-cost hydrophilic polymers can eliminate the need for organic solvents (Lawton 2002).

Therefore, García-Moreno et al. (2016, 2017, 2018) studied the development of carbohydrate-based carriers containing fish oil *via* electrohydrodynamic processes (García-Moreno et al. 2016, 2017, 2018). Two neutral polysaccharides of microbial origin (i.e. pullulan and dextran with linear and non-linear structures, respectively), and glucose syrup were evaluated as wall materials. All of these carbohydrates are food grade, biodegradable, biocompatible, and water soluble (Park and Khan 2009). When García-Moreno et al. (2016) developed the fish oil delivery systems by emulsion-based electrospinning using PVA as polymer, they reported that despite the high EE (92.4 ± 2.3%), their fibers was unable to protect fish oil against oxidation. Even the highest amount of oxidation was estimated for encapsulated samples. Due to the content of trace metals in PVA, this polymer had a negative influence on the oxidative stability of encapsulated fish oil (García-Moreno et al. 2016; Jacobsen et al. 2018). Several strategies can be applied to enhance the oxidation resistance of nanofibers, such as performing electrospinning process under partial nitrogen atmosphere, increasing the concentration of wall materials to enhance the fiber diameter, and consequently improving the entrapment of fish oil, and adding various antioxidant agents (e.g. EDTA, essential oils) into emulsions (Moomand and Lim 2014). García-Moreno et al. (2016) showed that the best oxidation resistance was obtained when EDTA added into emuslsions before blending with the PVA solution (Table 3). This effect was related to the metal chelating properties of antioxidants (García-Moreno et al. 2016). Overall, by evaluating the results presented in Table 3, a higher protective effect of zein as a hydrophobic wall material was clearly observed as compared to the use of hydrophilic wall materials (glucose syrup, pullulan, or dextran) to encapsulate fish oil; the mean of EE was 87.8% vs. 80.4%, as shown in Table 3. Therefore, the great potential and versatility of zein is a promising result to continue investigating the use of this biopolymer as wall material.

Solution (emulsion) properties such as viscosity, surface tension, and conductivity, are important for the successful EHDP. These properties are depending on the solvent type, polymer molecular weight, and concentration. García-Moreno et al. (2017) reported that the viscosity of dextran 500 solution was considerably higher than that obtained for dextran 70 solutions at the same concentration, due to the higher molecular weight of dextran 500. Therefore, the use of dextran 700 was not considered suitable for preparation of fibers or capsules via electrohydrodynamic processes (García-Moreno et al. 2017). Moreover, García-Moreno et al. (2017) showed that the method of oil addition into the electrospinning solution (emulsified or neat) had a great influence on the oxidative status, particle size and EE of the fibers. The fibers containing neat fish oil showed more oxidative stability than fibers with emulsified oil. It correlated well with the high EE obtained for fibers based on neat oil,

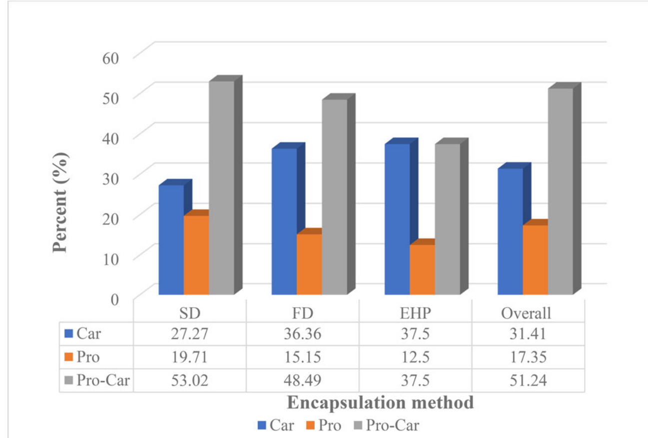

Figure 5. Frequency of carrier agents based on various encapsulation methods used to produce fish oil particles; Car, carbohydrate-based carrier; Pro, protein-based carrier; Car-Pro, carbohydrate-protein-based carrier. SD, spray drying; FD, freeze drying; EHP, electrohydrodynamic processes; SCAP, supercritical antisolvent process.

due to a lower surface oil content. This might be because of the lipid oxidation during emulsion preparation (due to increase in the surface area and oxygen inclusion) and encapsulation process (due to the exposure of the surface oil to the air) (Drusch et al. 2006; García-Moreno et al. 2017).

Paximada, Howarth, and Dubey (2018) investigated the effect of emulsion homogenization pressure and passes (1, 2, 4, & 8) on the size and EE of fish oil capsules. All the particles produced at higher homogenization pressure and a greater number of passes showed smaller droplet sizes and a higher EE. Emulsions treated by 1 pass, were not stable, while the emulsions homogenized by 8 passes could not produce any powder (probably due to their high viscosity). Therefore, the emulsions prepared by 2 and 4 passes were considered suitable for encapsulation *via* electrospraying method (Paximada, Howarth, and Dubey 2018). García-Moreno et al. (2018) applied whey protein and carbohydrates (pullulan and dextran or glucose syrup) mixtures as wall materials to encapsulate fish oil. The process used to emulsify the fish oil, including high-pressure homogenization or rotor-stator emulsification was studied (García-Moreno et al. 2018). When the oil was emulsified by a rotor–stator, the fibers exhibited higher EE and consequently higher oxidative stability than capsules prepared based on high-pressure homogenization. In general, optimization of the feed formulation has a positive effect on the encapsulation performance via electrohydrodynamic processes.

Yang et al. (2017) also studied the effect of solvent and polymer concentration on the morphology of nanofibers. Their results showed that the formation of distinct nanofibers of polyvinylpyrrolidone (PVP) at a high ethanol concentration (100%) was easier than that produced at low ethanol concentration. The fast evaporation rate of ethanol can be a possible reason for this phenomenon. Moreover, by increasing the chain entanglement in high concentrations of PVP, the stability of the polymer jet also increased (Vega-Lugo and Lim 2012). So, higher concentration of PVP leads to more smooth nanofibers. Therefore, the best PVP solution was selected as 31% in pure ethanol.

The electrohydrodynamic technique itself can also affect the encapsulation performance of fish oil. In this regard, Yang et al. (2017) compared coaxial and single electrospinning to encapsulate fish oil in zein nanofibers. The loading capacity and EE of fish oil in the coaxial electrospun nanofibers were reported as 14.5% and 96.9%, respectively. These results indicated a higher efficiency compared to some other studies (García-Moreno et al. 2016; Busolo et al. 2019). Also, FTIR data demonstrated that fish oil was successfully entrapped into the coaxial electrospun zein nanofibers. The oxidative stability of encapsulated fish oil (coaxial nanofibers or single nanofibers) was significantly higher than non-encapsulated fish oil and the coaxial nanofibers provided the highest protection against oxidation. High EE can minimize the exposure of fish oil to oxygen (Torres-Giner et al. 2010).

Busolo et al. (2019) and Miguel et al. (2019) selected zein as a carrier for fish oil via an innovative process termed as electrospraying assisted by pressurized gas (EAPG). In this technique, a pneumatic injector, by compressed gas, is used to atomize the polymer solution which nebulization is carried out within a high electric field. The results showed a suitable EE (83–84%) but the performance of coaxial electrospinning was better (96%). This result can be related to this fact that the fiber forming carrier is more efficient than capsules (García-Moreno et al. 2016; Yang et al. 2017). However, the EE of the zein microcapsules produced by EAPG was comparable with the electrosprayed fish oil capsules made by water soluble carriers such as whey protein isolate, dextran, and pullulan (Miguel et al. 2019), as summarized in Table 3. In addition, Yang et al. (2017) produced nanoparticles *via* coaxial electrospraying, while microparticles were obtained in the studies of Busolo et al. (2018) and Miguel et al. (2019). This difference is related to the process parameters and the ratio of zein: fish oil. For example, the ratio of zein: fish oil was 2:1 in the study of Busolo et al. (2019), while Miguel et al. (2019) decreased the fish oil load of the capsules (zein: fish oil of 4:1); by decreasing the fish oil load, the particle size increases. The capsule diameter is mainly affected by the oil load, electrospraying conditions, type, and concentration of the shell material. Overall, a size range of 1–3 μm was reported for the electrosprayed particles (Table 3).

Investigation of the effect of various storage conditions (RH: 0% and 65%, temperature: 5 and 23 °C, and environment: air and vacuum as well as light and darkness) on the zein/DHA-enriched fish oil microcapsules showed that DHA oxidation was limited by the continuous bubbling of nitrogen to the zein solution during the process as well as the frequent withdrawal of the product from the collector and subsequent storage under vacuum. The coefficient of variation for peroxide value of free fish oil was significantly higher than that observed for the encapsulated zein microcapsules and the lowest one was reported for the samples stored under vacuum. In addition, due to their high EE, and oxidation stability, the fortified milk samples with the zein/DHA-enriched fish oil microcapsules showed a little difference with the non-fortified reconstituted milk (Busolo et al. 2019). Miguel et al. (2019) reported that the encapsulated fish oil was oxidized during electrospraying process. They related this phenomenon to the higher specific surface area of the capsules and the exposure of surface oil to atmospheric oxygen (Miguel et al. 2019). Thus, further research is still required to control the lipid oxidation during electrohydrodynamic processes.

Overall, the results showed that electrospinning is an efficient encapsulation technique to protect fish oil against oxidation and the increasing chance of oil entrapment within the matrix (e.g. by increasing the emulsion homogenization pressure and/or optimization of the type and concentration of wall material and ratio of fish oil to wall material); the use of new techniques such as coaxial processes are some of the efficient solutions to improve the performance of this method, which should be further investigated.

Supercritical antisolvent encapsulation of fish oil

SCAP can be considered as a continuous process which is important for the industry and large-scale production (Karim et al. 2017). SCAP is based on dissolving the solutes into a solvent by a supercritical fluid such as CO_2 which saturates the solvent and leads to the precipitation of solute using an antisolvent effect. In this process, the optimization of experimental parameters such as temperature, pressure, and feed flow rate are very significant in terms of encapsulation performance. In this regard, Karim et al. (2017) applied SCAP to encapsulate fish oil by hydroxypropyl methyl cellulose (HPMC) as a wall material and optimized temperature, pressure, and feed emulsion rate by response surface methodology (RSM). The temperature at 60 °C, pressure at 150 bar, and feed rate at 1.36 mL/min were selected as the optimum conditions. They analyzed the effect of HPMC content on EE and oxidative stability of fish oil particles. The results showed that EE of fish oil increased at higher polymer contents, probably due to its influence on the emulsion droplet size, which decreased with increasing solids content. It is worth noting that the EE has a positive correlation with oxidative stability; therefore the highest oxidative stability obtained for particles with the highest solids content (Karim et al. 2017). Overall, the results of current study showed that lowest EE was observed for the SCAP, as shown in Figure 2. Prieto and Calvo (2017) reported also a relatively poor EE (<45%) for fish oil-loaded particles produced *via* SCAP. For this reason, along with the limited number of studies evaluating this method, the average EE obtained for SCAP was low. Although, Prieto and Calvo (2017) did not achieve a high EE (<45%); this performance was similar to that generated by conventional solvent evaporation. However, a high control on the particle size was observed in particles obtained during SCAP (Prieto and Calvo 2017).

Yang and Ciftci (2017) used fully hydrogenated soybean oil as an inexpensive solid lipid to fabricate hollow solid lipid micro/nanoparticles via atomization of supercritical CO_2 (SC-CO2)-expanded lipid. This process is a single step "green" method to encapsulate fish oil which does not require emulsion preparation or any organic solvent for dissolving the lipid phase (e.g. fish oil). In this method, the lipid phase (including wall material: a solid lipid, and core material: liquid fish oil) is expanded using SC-CO2 (Yang and Ciftci 2017). The great EE of 97.5% w/w was obtained at 50%, w/w initial fish oil concentration. The mean particle size of particles increased at higher initial fish oil concentrations (5–18 μm), due to agglomeration of the particles with increasing surface oil. The control of the particle size in a wide range (microparticles to nanoparticles) is one of the most important advantages of SCAP. Oxidative stability of the loaded fish oil was significantly increased compared to the free fish oil. However, the coefficient of variation for encapsulated fish oil was more than free fish oil (Yang and Ciftci 2017), as shown in Table 4.

Although, SCAP is a green and promising method to encapsulate fish oil, the disadvantages of SCAP include; a longer washing period (because of aggregation and agglomeration of the particles in the nozzle), and a high capital cost which limits the application of this method for encapsulation of fish oil in an industrial scale (Sanguansri and Augustin 2006; Karim et al. 2017). Therefore, further investigation is needed for industrial applications.

Other techniques for encapsulation of fish oil

Nanoliposomes

To increase the bioavailability and oxidative stability of fish oil, encapsulation by nanoliposomes technology can be suggested. Liposomes are spherical and vesicular particles which consist of one or more bilayer membranes surrounding an aqueous core (Ostro and Cullis 1989; Tavakoli et al. 2018; Sarabandi and Jafari 2020). Ghorbanzade et al. (2017) produced fish oil-loaded nanoliposomes (300–500 nm) and utilized them for the fortification of yoghurt (Table 5). It was found that yoghurt fortified with nanoencapsulated fish oil showed a high oxidative stability during 21 days storage at 4 °C as compared to yoghurt fortified with neat fish oil. Moreover, the EE of the nanoparticles was very good (92.4%). The use of encapsulated fish oil did not have any significant effect on the sensorial properties of yoghurt (Ghorbanzade et al. 2017). Further studies should be carried out to evaluate the efficiency of this encapsulation method.

Transglutaminase catalyzed cross-linking

The use of transglutaminase (TGase) enzyme, as an efficient protein cross-linker, can help to prepare a stable protein-based microcapsule containing bioactive compounds (such as fish oil) for food applications. Rios-Mera et al. (2019) microencapsulated fish oil based on the complex coacervation of inulin and soy protein isolate as wall materials. They solved the instability problem of complex coacervates with additional TGase enzyme, as a protein cross-linking. The particles were spherical (<100 μm) with smooth surfaces and some wrinkles. An excellent EE (94%) was obtained at optimized conditions (e.g. inulin: soy protein isolate = 0.4 and fish oil: 20%). Overall, a good thermal and oxidative stability for fish oil-loaded microparticles was reported in the presence of TGase (Rios-Mera et al. 2019).

Ionic gelation

Ion gelation method is another mild process to encapsulate fish oil. The complexation of two differently surface charged biopolymers or solutions is the base of this method (Ko et al. 2019). The nano/microcapsules are commonly prepared by extruding a wall material solution through a needle/nozzle into a gelation solution containing specified ions (Kuo and Ma 2001). For example, Vasile, Judis, and Mazzobre (2017) used this method to encapsulate fish oil in calcium–alginate–chitosan beads. They evaluated the effect of *Prosopis alba* exudate gum as a novel excipient. Alginate and this gum had a synergistic effect on the emulsion stability properties and resulted in the highest EE (98.63%) and yield (89%) (Vasile, Judis, and Mazzobre 2017).

Conclusion

In this meta-analysis and systematic review, the various encapsulation techniques used to encapsulate fish oil were studied. The results showed that, in spite of the negative effects of high temperature on the quality of encapsulated fish oil, spray drying method is still considered as the most common technique for producing fish oil-loaded particles because of its simplicity, low cost, and short drying times (5–30 s). The development and scale-up of milder approaches is one of the most interesting topics for future research. Freeze drying process, as a milder procedure, although may enhance the rehydration feature of capsules, oxygen can diffuse faster through these pores and the pores enabled the scavenging of oxygen. Electrohydrodynamic method is one of the most promising techniques for encapsulation of fish oil. Evaluation of the effect of encapsulation method on the encapsulation efficiency revealed that this technique provides the best performance for encapsulation of fish oil. Nevertheless, high investments costs and relatively low yields are the major barriers for application of this techniques in the food industry. Therefore, further researches are required to decline the final cost of the electrospun fibers and electrosprayed capsules and control the lipid oxidation during electrohydrodynamic processes. Spray–freeze drying is another mild technique which combines the advantages of freeze drying and spray drying to produce microparticles without heat damage and protects powders against agglomeration. Other techniques such as supercritical antisolvent process, ionic gelation, nanoliposomes, and transglutaminase catalyzed cross-linking have also been reported for encapsulating fish oil. But these methods need more study to optimize their performance and making them appropriate for industrial application to encapsulate fish oil.

Disclosure statement

No potential conflict of interest was reported by the authors.

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