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ORIGINAL ARTICLE

Chemical and physical characterization of spray dried fish oil with different combination ratios of wall component

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Abstract

The combination of wall materials used in the spray drying process can be recommended to increase the stability of functional oils and to ensure the best preservation of the nutraceutical compounds. For this purpose, maltodextrin (MD) and sodium caseinate (SC) combinations were studied for the spray drying of fish oil in this study. Microencapsulation efficiency (ME) ratio was similar in all groups. However, according to SEM images, group in formulated at 1SC:2MD ratio met all necessary conditions well such as no agglomeration, homogeneous distribution and spherical shape. There was no significant difference in terms of EPA and DHA contents of the groups. The lowest peroxide value (5.46 ± 1.00 mEq/kg) and TBA value (0.27 ± 0.02 mg MA/kg) was detected in the 1SC:2MD group. It was found that 1SC:2MD ratio for wall combination was more effective than 2SC:1MD and 1SC:1MD for encapsulation and maintaining the oxidative quality of fish oil. The results obtained in this study contribute to the optimization of spray drying of fish oil, which allows for more widespread use in the food industry.

Novelty Impact Statement: In this study, it was determined the right coating material combination in the spray drying process of fish oil and recommended as an effective combination for industrial-scale production. Spray-drying with MD and SC was at the desired form physically and chemically and 1SC:2MD combination showed better oxidative stability. In addition, morphology and color properties of 1SC:2MD were better than the other groups.

1 | INTRODUCTION

Microencapsulation technique is accepted as an important tool in the production and development of "functional foods" (Ye et al., 2018). Industrial-scale spray drying is widely preferred microencapsulation technique for heat-sensitive materials due to its easy application, relatively short drying time, low cost and ability to prepare high-quality microcapsules (Zhu et al., 2011). In this process, besides the short drying time, it is thought that the applied high temperatures do not damage the product, since the wet bulb temperature (30–50°C) is not exceeded theoretically. This technology allows the conversion of liquid fish oil, which is highly sensitive to heat, into an easier-to-use and stable dry powder. The spray drying process for making microcapsules of functional oils involves three basic steps. The first of these is the selection of the best encapsulating material for the target component. The second is the preparation of an emulsion containing the core and wall materials in the right proportions, and the third is the adjustment of the device setting such as inlet and outlet temperatures, aspiration and feed rate (Minemoto et al., 2002). These steps are crucial for encapsulating nutritionally valuable sensitive ingredients, including antioxidants, bioactive phenolic and essential fatty acids.

Jafari, Assadpoor, Bhandari, and He (2008) reported that one of the significant factors affecting the encapsulation efficiency of microencapsulated oils is the type of the carrier material. Maltodextrin (MD) is a hydrolyzed starch that is widely used as a wall material in the microencapsulation of food ingredients (Gharsallaoui et al., 2007). The use of MD as a coating material provides many advantages

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such as low cost, neutral taste and aroma, low viscosity at high solid concentrations, and good preservation against oxidation. However, its low emulsifying capacity is the biggest disadvantage of this wall material. Therefore, some researchers suggested the combination of MD with other surface-active biopolymers such as gum arabic, modified starches and proteins to provide effective microencapsulation by spray drying (Bae & Lee, 2008; Bule et al., 2010; Fernandes et al., 2008; Hogan et al., 2003; Soottitantawat et al., 2003). The selection of coating material combinations was reported to affect also powder properties such as morphology, particle size, surface oil (SO), and oxidative stability (Jafari, Assadpoor, He, & Bhandari, 2008). Therefore, in this study, it was aimed to determine the combination of the wall materials commonly used during spray drying to improve the stability of fish oil and to ensure the best protection of the valuable fatty acids in its structure. For this purpose, sodium caseinate (SC), defined as a milk protein, and MD combined in different ratio and they were emulsified with fish oil for the microencapsulation by spray drying. The ME, oxidative stability and microencapsule morphology of the obtained microencapsules were investigated for the determining of the most appropriate coating ratio.

2 | MATERIAL AND METHODS

2.1 | Material

Fish oil (anchovy oil, *Engraulis encrasicolus*) was supplied from a local company (Kardez Su Urn.). MD (DE: 18–20) and SC were purchased from a commercial company (Kimbiotek Chemical Company Ltd, Istanbul). All chemicals used were of analytical grade and obtained from Merck.

2.2 | Emulsion preparation

As core material, 10% fish oil was used in all groups. In oil/water emulsions, oil and coating material ratio was 1:2. Total solid content of the emulsion was maintained at 30% (Kwamman & Klinkesorn, 2015). In the first group, SC and MD were mixed in a ratio of 2:1 (2SC:1MD), respectively, then in the second and third groups, they were combined in ratios of 1:1 (1SC:1MD) and 1:2 (1SC:2MD). The wall materials were dissolved in distilled water under magnetic agitation, 1 day before emulsification and kept at 4°C to complete the hydration of the ingredients. The coating solutions were mixed and kept in bain-marie (50°C) for 1 hour prior to emulsification. After cooling, fish oil (10%) was added and homogenized at 14000 rpm for 10 min by an ultra-turrax (IKA T-25).

2.3 | Microencapsulation by spray drying

The laboratory-scale spray dryer (Buchi Mini Spray Dryer, B-290) with a 0.7 mm diameter spray nozzle was used for the drying of the emulsions. The emulsion was fed into the main chamber through a peristaltic pump, and feed flow rate was 20 ml/min, and aspiration rate was about 30 m^3/h. The inlet temperature was set to 160°C, the outlet temperature was recorded as 90 ± 5°C. The microencapsulated fish oils were collected from a cyclone separator and stored in plastic bottles with a lid at 4°C for further analysis.

2.4 | Microencapsulation efficiency

Total oil was measured by Bligh and Dyer method (Bligh & Dyer, 1959), and SO was measured by washing 2 g of microencapsulated fish oil with 15 ml of hexane followed by 2 min vortex at room temperature. The solvent mixture was filtered (Whatman no. 1 filter paper) followed by rinsing the papers three times with 20 ml of hexane (Bae & Lee, 2008). Evaporation of the solvent was carried out using a rotary evaporator (Hei-VAP-Value, Heidolph Instruments GmbH, Germany). The resulted SO was calculated based on the weight differences between the initial clean flask and that containing the extracted oil residue. The ME was calculated using the following equation:

$$\text{Microencapsulation efficiency} = \left[(\text{Total oil} - \text{Surface oil}) / \text{Total oil}\right] \times 100.$$

2.5 | Morphology of microcapsules

Particle morphology of microencapsulated fish oil was evaluated by scanning electron microscopy (SEM) at Central Research Laboratory of Cukurova University (CUMERLAB). Powders were coated with gold for 30 s to a thickness of 2 nm using a Q150R ES Coater (Quorum Technologies). SEM (Quanta FEG 650) was operated at voltage of 10 kV with a magnification of 1000 and 3000.

2.6 | Color analysis (L^*, a^*, and b^* values)

Color parameters were measured using a portable chromameter (CR-400, Konica Minolta, Osaka, Japan). The results were expressed in terms of L^* value (lightness) ranging from 0 (black) to 100 (white). The redness (a^* value) assigns positive values for the red and negative values for green. With respect to yellowness (b^* value), positive values represents yellow and negative ones to blue. Before measurements, the colorimeter was calibrated with a white standard plate. The whiteness index (WI) was calculated with the following formula (Park, 1994).

$$\text{Whiteness index (WI)} = 100 - \left[(100 - L)^2 + \left(a^2 + b^2\right)\right]^{0.5}.$$

2.7 | Fatty acid analysis

Fatty acid methyl esters (FAMEs) from the extracted oil of the powders and raw fish oil were prepared with 2 M methanolic KOH

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according to methods of Ichihara et al. (1996). The mixture was centrifuged at 2594 g for 10 min and then n-heptane layer was taken for GC analyses. Agilent GC–MS/MS (7890B GC-7010B MS) with an autosampler (Gerstel) equipped with a flame ionization detector and a capillary Agilent J&W DB-WAX column (60 m × 0.25 μm × 0.25 μm) were used for the analyses. The oven temperature was held at 50°C for 1 min, increased to 200°C at a rate of 25°C/min held for 10 min and then to 230°C at a rate of 3°C/min kept this temperature for 18 min. The injector (250°C) and detector temperatures (300°C) were set. The sample size was 1 μl, and the flow rate of carrier gas was 1 ml/min. The split was 1:40. Fatty acids were defined by comparing the retention times of FAME with the standard 37-component FAME mixture (Supelco-37 Comp. FAME Mix).

2.8 | Analysis of lipid quality indicators

Peroxide values (PVs) of the powders were measured according to the Cd8-53 method (AOCS, 1994). 20 ml of chloroform was added to 1 g of sample, then 50 ml of acetic acid: chloroform (60:40, v/v) solution was added to the mixture and shaken until the lipid was completely dissolved. 1 ml of saturated aqueous potassium iodide solution (KI, ACS grade) was added and stirred for 1 min. The samples were kept in a dark place for 30 min. Then, 100 ml of distilled water and a starch indicator solution were added to the mixture and titrated with 0.002 M sodium thiosulfate until a clear color appeared. All these applications were repeated in the blind group without sample. The results were expressed in miliequivalents of active oxygen per kilogram of oil. PV was calculated according to the following formula

$$\mathrm{PV} = \left[(S - B) \times N \times 1000\right] / \text{weight of sample.}$$

The AOCS official method Cd 19–90 (1998) was used to measure thiobarbituric acid reactive substance (TBAR) values of fish oil microcapsules. The oil sample (50–200 mg) was solubilized in 1- butanol, mixed with TBA reagent (0.02% in 1-butanol), heated in a 95°C water bath for 2 h, and rapidly cooled under running topwater. Finally, the absorbance was measured at 532 nm against blank and the results are expressed as malonaldehyde equivalents in μg/g fish oil.

The free fatty acid (FFA) contents of the samples were measured titrimetrically according to the official method of AOCS (Ca 5a-40, 1994). The weighed sample was neutralized in dietielether:ethanol (25:25 ml ratio). The solution was titrated with 0.1 M sodium hydroxide in the presence of 1% phenolphthalein indicator until a permanent pink color was obtained. The results were expressed as oleic acid equivalents (oleic acid %) and calculated with the following formula:

$$\text{FFA, as oleic acid}\% = \left[\left(\text{alkali volume}\ [\text{ml}] \times \text{alkali normality} \times 28.2\right) / \text{sample weight(g)}\right].$$

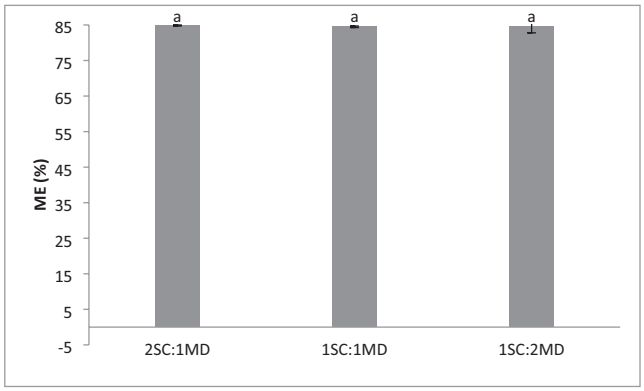

FIGURE 1 Microencapsulation efficiency of microencapsulated fish oils. Bars with the different letters indicate significant differences ($p < 0.05$).

2.9 | Statistical analyses

All data were subjected to analysis of variance (one-way ANOVA), at 5% confidence level using the Duncan multiple-range test.

3 | RESULTS AND DISCUSSION

3.1 | Microencapsulation efficiency

ME was calculated based on the ratio of SO to total oil content, and the results are presented in Figure 1. For all combinations of wall materials, there was quite high encapsulation efficiency (84.49%–84.85%). Pourashouri et al. (2014) reported that the ME values of fish oils with different wall material composition were in the range of 67–88%. Carneiro et al. (2013) investigated the potential of MD combination with different wall materials in the microencapsulation of flaxseed oil by spray drying, to maximize encapsulation efficiency and minimize lipid oxidation. ME values were reported to be between 62.3% and 95.7%, and the lowest encapsulation efficiency was obtained for the combination of MD and whey protein. It was reported that carbohydrate and protein combination could influence on the physical properties of coating material and improve the encapsulation process (O'Regan & Mulvihill, 2010). However, Lai et al. (2021) recorded that the low protein and high carbohydrate combination showed higher encapsulation efficiency. In addition to high ME values in all groups in this study, no significant difference was found among the groups at different ratio MD and SC ($p > 0.05$).

3.2 | Morphology of microcapsules

The final utilization of microcapsules in the food products depends on appearance, quality and flowability of the produced powder. These microcapsule properties are directly dependent on the particle size of the powder (Mahdi et al., 2020). Size also plays a role in gastrointestinal performance: microparticles below 800 μm can pass through

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the pylorus without the effect of gastric emptying, thus eliminating interpersonal and intrapersonal (nutritional) differences (Lengyel et al., 2019). The typical shape of microparticles is spherical in the size range of 1–1000 μm, and particles between 1–100 μm can be produced by spray drying (Sahil et al., 2011). Powders added to foods are expected to have sizes in these ranges. The particle size of microencapsulated fish oil produced with different wall combination in this study was within the desired range, with value of 17.00–32.03 μm for 2SC:1MD group, 2.82–12.95 μm for 1SC:1MD group and 3.40–8.99 μm for 1SC:2MD (Figure 2). Small particle size is preferred as less effect on food texture is expected. The microcapsule with 1SC:2MD as a wall material showed considerably smaller powder particle size in comparison with others. The largest diameter particles were observed in the 2SC:1MD group. It is essential for the microcapsules to be uniform and round particles to protect the core material from oxygen and the release of oil droplets to the surface of the particles. The morphology of the particles showed these characteristics in all groups without any cracks and pores although in some places surface dents resembling craters were observed. It can be observed from the Figure 2 that agglomeration increased with increase in SC ratio in the coating formulation. To obtain a uniform distribution and non-agglomerated capsules, it is necessary to find the optimal SC/MD ratio. Formulated in 1SC:2MD ratio, microcapsules met all necessary conditions such as no agglomeration, homogeneous distribution and spherical shape. It can be concluded that the microcapsules formulated in 1SC:2MD ratio provided all the necessary conditions such as no agglomeration, homogeneous distribution and spherical shape.

3.3 | Color characteristics of microcapsules

Color data of obtained microcapsules are presented in Table 1. There was no significant difference in all groups in terms of L^* value representing the light–dark spectrum (0–100). It was determined that when the protein ratio was increased (2SC:1MD group), although the lightness of the microencapsules did not change, the yellowness increased (11.04 ± 0.09) greenness (−0.03 ± 0.02) decreased ($p < 0.05$). However, the rising in b^* values can be associated with the potential oxidation in foods (Ceylan et al., 2018; Mousavipour et al., 2021). According to the whiteness value calculated from the L^*, a^* and b^* value, the highest whiteness value (80.27 ± 0.28) was observed in 1SC:2MD group ($P < 0.05$). It was observed that the color values of all groups were in accordance with the color values obtained in previous studies (Özyurt et al., 2020; Uçar et al., 2020).

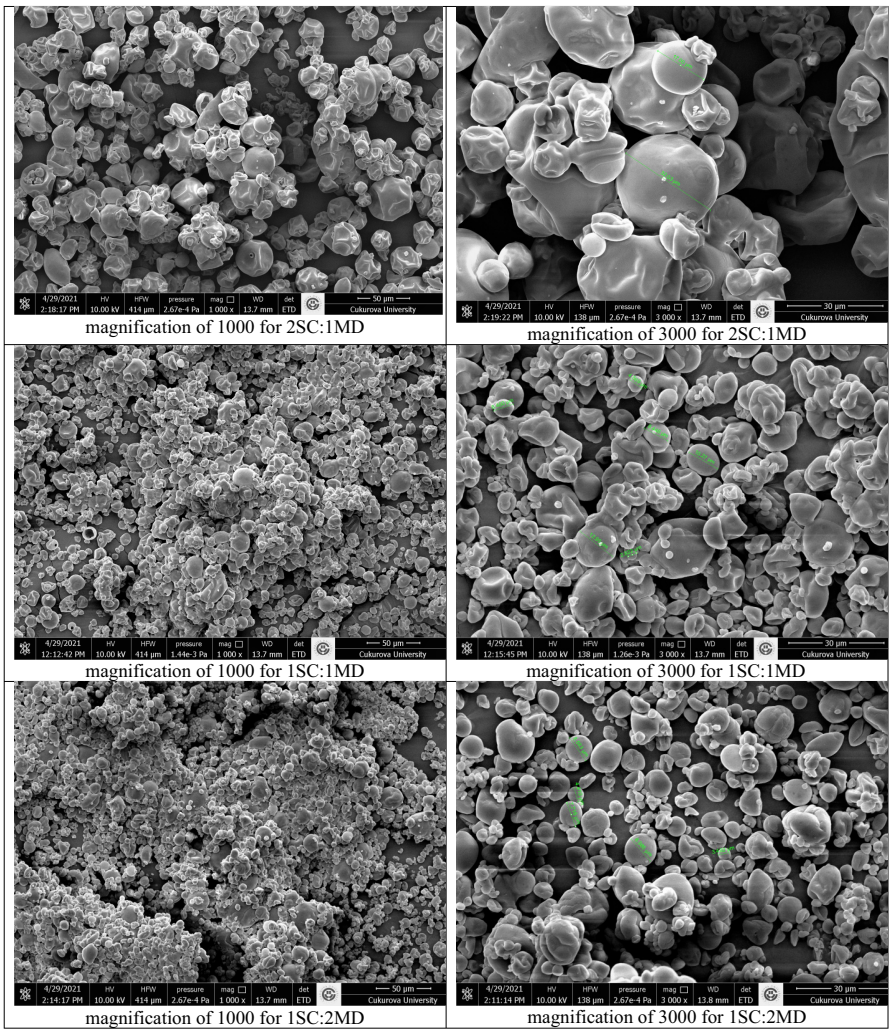

FIGURE 2 Scanning electron microscopy images of the microencapsulated fish oils. (a) Free fatty acid value of microencapsulated fish oils. (b) Peroxide value of microencapsulated fish oils. (c) Thiobarbituric acid reactive substance value of microencapsulated fish oils.

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3.4 | Fatty acid profiles of microcapsules

Fatty acid profiles of fish oils with different wall component ratio after microencapsulation process are shown in Tables 2. Similar fatty acid profiles were found in both raw fish oil and microencapsulated oils. The main fatty acids in all groups were myristic acid, palmitic acid, stearic acid, palmitoleic acid, oleic acid, eicosapentaenoic acid (EPA, C20:5ω3) and docosahexaenoic acid (DHA, C22:6ω3). The total saturated fatty acid ratio in raw fish oil was 34.29%, and after microencapsulation, it considerably increased to 37.08%–37.55%. On the other hand, there were similar rate decreases at total PUFA ratio in all microcapsule groups. However, the least decrease in PUFA content was observed in the 1SC:2MD group (31.17%) compared with the raw fish oil (33.49%). There was no significant difference in terms of EPA and DHA contents of the microencapsulated groups ($p > 0.05$). The EPA ratio of raw fish oil was found to be 9.70%, and it was observed in the range of 8.85%–9.21% in microencapsulated groups. While the rate of DHA was determined as 17.57% before microencapsulation, it was determined as 15.71%–16.12% in microcapsulated groups. These results showed that homogenization of the emulsion and/or microencapsulation by spray drying affected the fatty acid composition of fish oil. However, reduction in PUFA in this study was found to be at low levels compared with most previous studies (Annamalai et al., 2020; Botrel et al., 2017; Jeyakumari et al., 2017; Łozińska et al., 2020; Yeşilsu & Özyurt, 2019).

TABLE 1 Color evaluations (L^*, a^*, b^*, and whiteness value) of microencapsulated fish oils

	2SC:1MD	1SC:1MD	1SC:2MD
L^*	82.85 ± 0.12^a	82.47 ± 0.26^a	82.77 ± 0.34^a
a^*	-0.03 ± 0.02^c	-0.31 ± 0.02^a	-0.19 ± 0.02^b
b^*	11.04 ± 0.09^c	10.63 ± 0.25^b	9.6 ± 0.05^a
Whiteness	79.61 ± 0.14^a	79.49 ± 0.11^a	80.27 ± 0.28^b

Note: $n = 3$, ±SD. Letters (a–c) in the same line indicate statistical difference between groups ($p < 0.05$).

3.5 | Lipid quality of microcapsules

FFA, peroxide, and TBAR values of microencapsulated fish oils are presented in Figure 3a–c. The accumulation of FFA can increase lipid oxidation products, which can negatively affect the flavor of the edible oil and also develop rancidity. Thus, the evaluation of the FFA content is an important parameter for the quality evaluation of fish oils which are highly susceptible for oxidation. The FFA value of raw anchovy oil was 2.30±0.12% oleic acid, and there was no significant difference in FFA levels in all groups (4.26%–5.14% oleic acid) after microencapsulation procedures (Figure 3a). The FFA values of all groups in this study

TABLE 2 Fatty acid compositions of microencapsulated fish oils (%)

	Raw fish oil	2SC:1MD	1SC:1MD	1SC:2MD
C12:0	0.08 ± 0.00^a	0.10 ± 0.00^b	0.13 ± 0.01^c	0.12 ± 0.00^c
C14:0	6.35 ± 0.20^a	7.01 ± 0.07^b	6.90 ± 0.37^{ab}	6.89 ± 0.10^{ab}
C15:0	1.05 ± 0.03^a	1.15 ± 0.00^b	1.13 ± 0.02^b	1.12 ± 0.01^b
C16:0	20.33 ± 0.28^a	22.91 ± 0.16^b	21.98 ± 0.57^b	22.72 ± 0.09^b
C17:0	0.84 ± 0.05^a	0.92 ± 0.01^a	0.89 ± 0.02^a	0.92 ± 0.02^a
C18:0	5.19 ± 0.06^a	5.04 ± 0.09^a	5.63 ± 0.06^b	5.16 ± 0.10^a
C20:0	0.46 ± 0.02^a	0.44 ± 0.02^a	0.44 ± 0.03^a	0.42 ± 0.04^a
∑SFA	34.29	37.55	37.08	37.33
C16:1	5.43 ± 0.12^a	5.13 ± 0.45^a	5.35 ± 0.13^a	5.35 ± 0.13^a
C18:1	17.57 ± 0.21^a	17.68 ± 0.18^a	17.39 ± 0.04^a	17.40 ± 0.08^a
C18:1ω7	2.07 ± 0.02^c	1.88 ± 0.04^b	1.83 ± 0.03^{ab}	1.78 ± 0.01^a
C20:1	0.49 ± 0.01^a	0.52 ± 0.04^a	0.47 ± 0.07^a	0.48 ± 0.03^a
∑MUFA	25.54	25.35	25.17	25.24
C18:2ω6	2.33 ± 0.06^a	2.30 ± 0.05^a	2.22 ± 0.04^a	2.31 ± 0.06^a
C18:3ω6	1.03 ± 0.05^b	0.13 ± 0.03^a	1.03 ± 0.01^b	0.97 ± 0.04^b
C18:3ω3	1.74 ± 0.05^a	1.68 ± 0.03^a	1.62 ± 0.01^a	1.62 ± 0.04^a
C20:2	0.16 ± 0.08^a	0.22 ± 0.01^a	0.22 ± 0.01^a	0.23 ± 0.03^a
C20:3ω3	0.97 ± 0.03^b	0.81 ± 0.08^a	0.93 ± 0.06^{ab}	0.83 ± 0.03^{ab}
C20:5ω3	9.70 ± 0.01^b	9.21 ± 0.08^{ab}	8.85 ± 0.08^a	9.12 ± 0.08^{ab}
C22:6ω3	17.57 ± 0.21^a	16.12 ± 0.28^b	15.70 ± 0.65^b	16.11 ± 0.57^b
∑PUFA	33.49	30.46	30.56	31.17

Note: $n = 3$, ±SD. Letters (a–c) in the same line indicate statistical difference between groups ($p < 0.05$).

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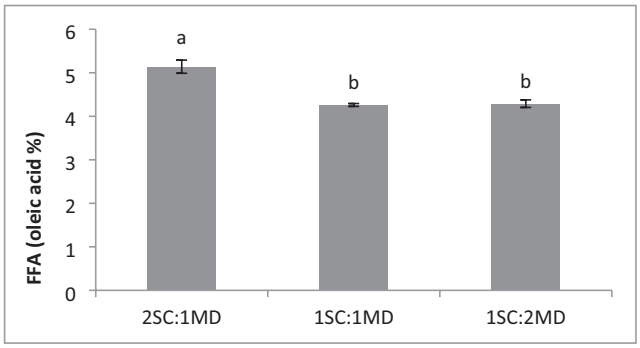
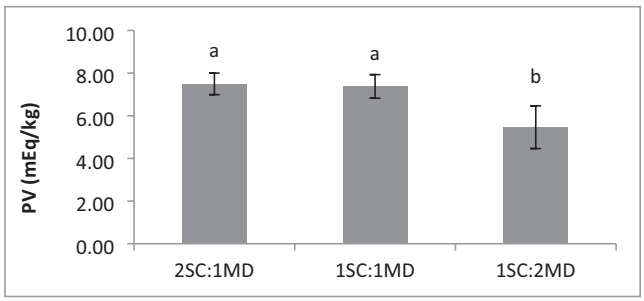
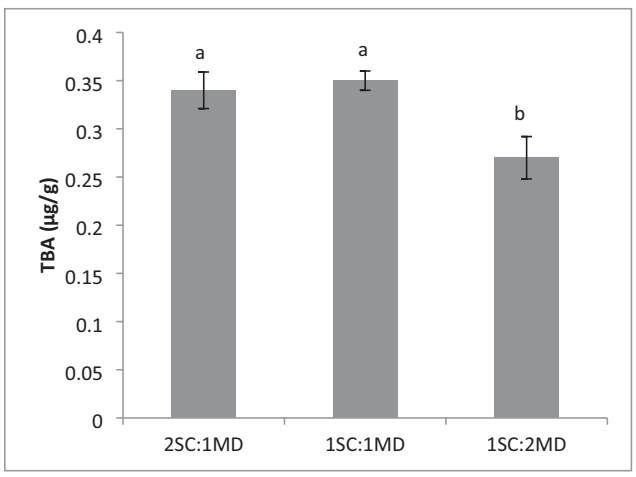


(a) FFA value of microencapsulated fish oils



(b) PV value of microencapsulated fish oils



(c) TBARs value of microencapsulated fish oils

FIGURE 3 (a–c) Peroxide, thiobarbituric acid reactive substance, and free fatty acid values of microencapsulated fish oils. Bars with the different letters (a–c) indicate significant differences of the parameter with respect to kind of microencapsulated fish oil ($p < 0.05$).

were within the allowable limit (1%–7% oleic acid) according to the International Association of Fish Meal and Oil Manufacturers (1981).

PVs of raw fish oil, 2SC:1MD, 1SC:1MD and 1SC:2MD groups were found as 3.23 ± 0.11, 7.49 ± 0.51, 7.38 ± 0.55, and 5.46 ± 1.00 mEq/kg, respectively (Figure 3b). Aghbashlo et al. (2013) reported that the PV of raw fish oil was 5.00 mEq/kg and increased in microcapsulated oils with different wall materials spray-dried at 160°C (6.85–8.50 mEq/kg). The researchers noted that the oxidative stability of the microcapsules was greatly affected by the wall material combination. The highest lipid protection against oxidation was found to be MD and whey protein combinations compared with gum arabic-MD and single MD combinations. Kagami et al. (2003) recorded that the microcapsules of the protein-only wall matrix resulted lower oxidative stability. Similarly, in this study, when the protein ratio in the wall combination increased, the PV increased and it was observed that increasing the MD ratio in the wall component improved the oxidative stability of the microcapsules. Results on secondary lipid oxidation (TBARs) in fish oil microcapsules are presented in Figures 3c. TBARs value of raw fish oil was determined as 0.24 ± 0.01 mg MA/kg, and it was found that microencapsulated groups had TBARs values of less than 1 mg MA/kg. The lowest TBAR value (0.27 ± 0.02 mg MA/kg) was detected in the 1SC:2MD group in parallel with PV results. However, TBARs values in all groups were found below the limit of threshold for the oxidation acceptability (2.5 mg MD/kg) (Zhang et al., 2019).

It is known that hydrolysed starches such as MD are generally used to increase the oxidative stability of encapsulated oils by reducing oxygen permeability of wall matrix (Kagami et al., 2003). However, crystallization of carbohydrates can cause disruption of structural integrity of the wall matrix and induces agglomeration or caking of powders, resulting in the release of some encapsulated oil and induction of lipid oxidation oxidation (Drusch et al., 2006). Our results showed that 1SC:2MD ratio for wall combination was more effective than 2SC:1MD and 1SC:1MD for encapsulation and maintaining the oxidative quality of fish oil.

4 | CONCLUSION

Spray drying is a convenient way of preserving functional oils in the form of micro and nanoparticles and is a rapidly expanding technology for the food and many other industries. Although the use of MD as a carrier agent in this technique seems advantageous, its combination with proteins may give better results for microcapsule production. In this study, it was found that fish oil microcapsules produced by spray drying with MD and SC was at the desired form physically and chemically. However, 1SC:2MD combination showed better lipid quality, coupled with desirable morphology and color characteristics. A 1:2 mixing of SC and MD, respectively, can be recommended as an effective combination for industrial-scale fish oil microencapsulation.

AUTHOR CONTRIBUTIONS

Gülsün Özyurt: Conceptualization; Project administration; Investigation; Methodology; Writing—original draft. Yetkin Sakarya: Formal analysis; Writing—review & editing. Mustafa Durmuş: Investigation; Methodology; Formal analysis; Writing—review & editing.

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CONFLICT OF INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

DATA AVAILABILITY STATEMENT

The data sets are available from the corresponding author on reasonable request.

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