Journal of Food Engineering 183 (2016) 16–23

Contents lists available at ScienceDirect

Journal of Food Engineering

journal homepage: www.elsevier.com/locate/jfoodeng

Mass transfer modeling of sardine oil polyunsaturated fatty acid (PUFA) concentration by low temperature crystallization

R. Morales-Medina*, G. De León, M. Munio, A. Guadix, E. Guadix

Department of Chemical Engineering, University of Granada, Granada, Spain

ARTICLE INFO

Article history:
Received 6 October 2015
Received in revised form
28 February 2016
Accepted 20 March 2016
Available online 22 March 2016

Keywords:
Solvent crystallization
PUFA concentrates
Sardine oil
Mass transfer model

ABSTRACT

Low temperature crystallization of oil is employed in the production of omega-3 which are a product of interest in the food industry. The aim of this work was to model the influence of temperature and time in the mass of the solid and liquid phases. The model developed enabled the estimation of polyunsaturated fatty acids (PUFA), eicosapentaenoic acid and docosahexaenoic acids (EPA and DHA) contents in the liquid phase. Solutions of sardine oil in hexane were crystallized at several temperatures (−55, −65, −75 and −85 °C) and times (1–24 h). The transient variation of liquid phase mass at a given temperature was modeled employing two parameters: the equilibrium mass of the phase (m_{eq}, g) and the overall mass transfer coefficient ($K_{G\cdot A}$, h^{-1}). The results showed that the highest PUFA concentrates (>80%) were produced at −85 °C and 24 h. The model proposed is a useful tool for estimating the composition and total mass of winterized oils ($r^2 > 0.83$).

1. Introduction

Fish discards are defined as the fraction of organic material in the catch which is not retained for sale but dumped back to the sea (Kelleher, 2005). Discarding presents a negative impact on the future fishing productivity and also alters the marine trophic chains causing an environmental problem. To prevent this situation, the new Common Fisheries Policy has introduced new EU fisheries regulations aimed to ban the discard practices in Europe (EU, 2013). From the 1st January 2015 on, all catches from pelagic species (mackerel, horse mackerel or sardines, among others) must be landed. In the case of the rest of the fish species this new regulation will come into force from 2017 on.

Therefore, technical solutions able to convert the fish discards into added-value products must be found. In this framework, the extraction of fish oil for the production of polyunsaturated fatty acids (PUFA) concentrates has become an interesting up-grading process. Specifically, in the Mediterranean Sea, it has been studied the oil content and composition of main pelagic discarded species. Among them, sardines presented the highest oil content (18 wt%) with a high content of polyunsaturated fatty acids (~40 wt %) (Morales-Medina et al., 2015). Hence, employing oil sardine as raw material for the production of concentrates could be an interesting up-grading proposal.

Polyunsaturated fatty acids, specially eicosapentaenoic and docosahexaenoic acids (EPA and DHA) have been described to exert a positive effect on the cardiovascular system (Jump et al., 2012). Additionally they present positive influence on the visual development of neonates (Weichselbaum et al., 2013), on the brain system functions (Bradbury, 2011), and on some types of cancer (Bougnoux et al., 2010). Consequently, refined oil products, with high concentration of polyunsaturated fatty acids, have increased their demand in the last years since they are used as food supplements and as nutraceutical products (Dillon et al., 2013; Kralovec et al., 2012; Kuratko and Salem, 2013; Lembke, 2013).

Polyunsaturated fatty acids concentration techniques can be conducted by several physical approaches such as supercritical fluid extraction, low temperature crystallization, molecular distillation or urea complexation (Rubio-Rodriguez et al., 2010). Prior to concentration, triacylglycerols are converted into methyl esters or fatty acids to enhance the yield of the concentration (Lembke, 2013). Once the PUFA has been concentrated, those esters or fatty acids must be re-esterified into triacylglycerols which are better digested and absorbed by human digestion (Small, 1991).

Winterization or fractional crystallization consists in the removal of high melting point compounds (i.e. saturated fatty acids) by cooling (Gunstone et al., 2012). For a given chain length, the melting point of the free fatty acids (FFA) decreases with the

* Corresponding author.
E-mail address: rocio_morales@ugr.es (R. Morales-Medina).

http://dx.doi.org/10.1016/j.jfoodeng.2016.03.009
0260-8774/

degree of unsaturation (Akoh, 2005; Gunstone et al., 2012). Therefore, at low temperatures, saturated fatty acids (SFA) crystallize and PUFA remain in the liquid phase (Wanasundara et al., 2005). Among the several types of winterization, solvent fractional crystallization is the most commonly employed (Cunha et al., 2009; Haraldsson, 1983). The usage of solvents promotes crystal formation (Lee and Foglia, 2001) and increases the yield and purity of the crystals (Cunha et al., 2009).

The main variables controlling solvent fractional crystallization are: (i) oil composition, (ii) temperature of crystallization, (iii) mobility of molecular species in the oil (influenced by the solvent polarity), (iv) oil:solvent ratio and (v) rate of cooling (López-Martínez et al., 2004). The oil composition refers not only to degree of unsaturation (i.e. saturated, monounsaturated of polyunsaturated fatty acids) but also the class of lipids, i.e triacylglycerols, free fatty acids or esters. The high variety of fatty acids present in fish oil results into an heterogeneous composition of triglycerides; fact which hinders the concentration process (Lembke, 2013). Hence, it is preferable to hydrolyze or esterify triacylglycerols to produce free fatty acids or fatty acids methyl/ethyl esters which can be more easily separated by physical approaches. In this point, Vázquez and Akoh (2011) described that, for the concentration of stearidonic acid (C18:4n-3) the employment of free fatty acids was more efficient than the use of esters. Regarding the temperature of crystallization, it has been widely described that: the lower the temperature, the higher the concentration of PUFA in the liquid fraction (Rubio-Rodriguez et al., 2010; Shahidi and Wanasundara, 1998; Wanasundara, 1996). Several authors (López-Martínez et al., 2004; Vázquez and Akoh, 2011; Wanasundara, 1996) have studied the influence of the organic solvent (hexane, acetone, diethyl ether or isobutanol among others), being the hexane the solvent that presented higher concentrations. Vázquez and Akoh (2011) also analyzed the employment of mixtures of hexane and acetone in several proportions; however, higher concentrations were achieved employing solely hexane. Additionally, the lower the solvent:oil ratio, the higher the purity and yield of the PUFA concentrates in the liquid phase (López-Martínez et al., 2004; Vázquez and Akoh, 2011). However, the usage of high volume of organic solvents presents an economical drawback. Hence, the selection of the oil:solvent ratio should be taken by balancing the fixed cost and the desired enrichment of PUFA.

As reviewed before, several studies have been conducted aiming to qualitatively describe the influence of several variables (i.e. temperature, time, type of solvent) on the production of PUFA concentrates. However, all those studies were focused on the composition of the liquid phase at a fixed time without considering the mass evolution of each phase. Additionally, from an industrial point of view, the knowledge of the evolution of the liquid and solid mass is fundamental for a proper design, control and optimization.

The aim of this work was to model the influence of temperature and time in the mass of the liquid phase obtained via crystallization at low temperature by following the theory of the mass transference. Moreover, the composition of the liquid phase (wt%) of polyunsaturated fatty acids, EPA and DHA was calculated employing the aforementioned model.

2. Materials and methods

2.1. Materials

Refined sardine oil with a total amount of EPA and DHA of 31.9% (21.5% of EPA and 10.4% of DHA) was provided by Industrias Afines S.L.

2.2. Free fatty acid production

Free fatty acids were released from the triacylglycerols by means of basic hydrolysis following the method described by Wanasundara and Shahidi (1999). Refined sardine oil was mixed with KOH (85%), aqueous ethanol (96%, v/v) and distilled water at 62 °C during 1 h under nitrogen atmosphere. The FFA produced were purified by subsequent extractions with distilled water (175 mL) and hexane. The remaining aqueous phase was acidified to pH 1 with 3 M HCl and the FFA were recovered by solvent extraction using hexane. Then, anhydrous sodium sulfate was added to the mixture of hexane and FFA aimed to extract the remaining water content. Finally, the mixture was filtered and the solvent was removed at 40 °C and 100 mmHg. Free fatty acids were stored at −80 °C until use.

2.3. Fatty acid profile analysis and lipid class composition

Direct transesterification was conducted to determine the fatty acid profile of samples following a method described by Rodríguez-Ruiz et al. (1998). To that end, 1 mL of the sample in hexane (1 mg/mL) was mixed with 1 mL of reagent mixture (methanol and acetyl chloride, 20:1, v/v). Then, samples were heated up to 90 °C for 1 h. As internal standard for quantitative determination nonadecanoic acid (C19:0) was employed. The fatty acids methyl esters were analyzed with an Agilent 7890A chromatograph (Agilent Technologies, S.A.) connected to a capillary column of fused silica Omegawax (0.25 mm × 30 m, 0.25 μm standard film; Supelco, Bellefonte, PA), and a flame-ionization detector (Camacho Paez et al., 2002). All measurements were done in triplicate and the results were expressed on a weight basis.

Thin layer chromatography (TLC) was employed to determine the polar lipid fraction composition of the samples. To that end, plates of silica-gel (Precoated TLC plates, SIL G-25; Macherey-Nagel, Sigma–Aldrich) were activated at 100 °C for 1 h. A mass of 1 mg of sample was spotted directly on the plate. The mobile phase was composed of chloroform/acetone/methanol (95:4.5:0.5, v:v:v) and the spots of each lipid were visualized by spraying the plate with iodine vapor in a nitrogen stream. Each fraction was scraped, methylated and analyzed as previously described.

2.4. Winterization

100 mL of a solution (5 wt%) of free fatty acids in hexane were located in amber flasks (125 mL) and stored in a Panasonic ultra-low temperature freezer MDF-U3386S (Panasonic Healthcare Co, Ltd) at several temperatures (−55, −65, −75 and −85 °C) during 1, 2, 3, 4, 6, 8 and 24 h. The selection of both, the solvent and the oil:solvent ratio, was conducted considering results described in literature. Effectively, hexane has been referred as the solvent with whom higher purity and yield were obtained (López-Martínez et al., 2004; Vázquez and Akoh, 2011; Wanasundara and Shahidi, 1999). The oil:solvent ratio was fixed to 5% (wt %) to obtain high concentrates in PUFA with an economical assumable volume of hexane (López-Martínez et al., 2004; Vázquez and Akoh, 2011).

The crystallized fatty acids obtained after each trial were recovered by filtering samples, at the same temperature as winterized, through a 25 μm filter paper using a vacuum filter pump. The solvent was removed from the liquid fraction with a Büchi Rotavapor R-210 (Büchi Labortechnik AG) during 15 min at 40 °C and 100 mmHg. The mass of the solid and the liquid fractions was determined for each trial. All crystallizations were done in duplicate. Finally, the fatty acid profile of each phase was analyzed as described before.