LWT - Food Science and Technology 46 (2012) 525–531

Contents lists available at SciVerse ScienceDirect

LWT - Food Science and Technology

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

Screening of macro- and bioactive microconstituents of commercial finfish and sea urchin eggs

Nick Kalogeropoulos*, Anastasia Mikellidi, Tzortzis Nomikos, Antonia Chiou

Department of Dietetics-Nutrition, Harokopio University, 70 El. Venizelou Str., 17671 Kallithea, Athens, Greece

ARTICLE INFO

Article history:
Received 7 August 2011
Received in revised form
17 November 2011
Accepted 24 November 2011

Keywords:
Fish eggs
Urchin eggs
Lipid composition
Fatty acids
Tocopherols
Carotenoids
Sterols

ABSTRACT

Crude and lipid composition, tocopherols, carotenoids, sterols and squalene content, together with fatty acids profiles of commercial marine egg products are reported, namely (a) canned eggs of lumpfish (*Cyclopteris lumpus*), Atlantic salmon (*Salmo salar*), trout (*Oncorhynchus mykiss*), smoked herring (*Clupea harengus*), and sea urchin (*Paracentrotus lividus*), (b) fresh sea urchin (*P. lividus*) eggs, and (c) beeswax covered, dried and salted mullet (*Mugil cephalus*) skeins -Greek "Avgotaracho". The protein content of marine eggs ranged from 8.7 to 33.7 g/100 g on a fresh weight (fw) basis. Among neutral lipids triacylglycerols predominated, except for mullet roe where wax and steryl esters comprised 75% of neutral lipids. Phosphatidylcholine was the major phospholipid, followed by phosphatidylethanolamine. Marine eggs contained 0.18–2.39 g/100 g fw long chain ω3 polyunsaturated fatty acids (PUFA), mainly docosahexaenoic acid (DHA) in finfish eggs and eicosapentaenoic acid (EPA) in urchin eggs. They also contained 86.2–342.0 mg cholesterol/100 g fw, low amounts of squalene and other sterols and significant amounts of tocopherols (0.45–4.4 mg/100 g) and carotenoids (0.31–11.4 mg/100 g). The commercial finfish and urchin eggs are food of high nutritive value providing significant amounts of protein, ω3 PUFA, phosphatidylcholine, carotenoids, and tocopherols, with fatty acids profiles that exhibit low atherogenic and thrombogenic potential.

1. Introduction

Eggs of aquatic animals are commonly referred to as roe, particularly when they are included in the original ovarian sac, to distinguish from caviars, which are the preserved eggs which have been separated from the supporting connective tissue. A great number of highly valued salted, smoked, boiled or canned products are made from fish eggs, and currently enjoy expanding international and domestic markets (Bledsoe, Bledsoe, & Rasco, 2003). Sea urchin roe is a luxury food product considered delicacy by consumers in several parts of the world (De la Cruz-García, López-Hernández, González-Castro, Rodríguez-Bernaldo De Quirós, & Simal-Lozano, 2000; Symonds, Kelly, Caris-Veyrat, & Young, 2007).

Edible marine eggs are considered as highly nutritive foods, having well balanced proteins with essential amino acids (De la Cruz-García et al., 2000; Iwasaki & Harada, 1985; Lu, Ma, Williams, & Chung, 1979), and providing significant amounts of long chain ω3 PUFA, mainly 20:5ω3 (EPA) and 22:6ω3 (DHA) (Lu et al., 1979), known to exert beneficial role in the prevention of cardiovascular diseases (Lee, O'Keefe, Lavie, Marchioli, & Harris, 2008). The marine egg lipids also contain significant amounts of bioactive minor compounds such as α-tocopherol, carotenoids and coenzyme Q10, which are considered critical for the normal development of aquatic embryos, and may be beneficial to human health (Moriya, Hosokawa, & Miyashita, 2007; Palace & Werner, 2006; Symonds et al., 2007).

In addition, recent literature data indicate that aquatic eggs exert significant biological actions. Mullet roe lipids were shown to exhibit a putative antithrombotic potential (Kalogeropoulos, Nomikos, Chiou, Fragopoulou, & Antonopoulou, 2008) and to inhibit *in vitro* the growth of colon cancer cells (Rosa et al., 2011), while herring roe lipids had beneficial effects on lipid and glucose metabolism in mice (Higuchi, Shirai, & Suzuki, 2006; Moriya et al., 2007). Fish roes are also source of dietary phospholipids which are thought to lower blood lipids and prevent liver steatosis (Cohn, Wat, Kamili, & Tandy, 2008). A group of sulfated polysaccharides

Abbreviations: AI, atherogenic index; BSTFA, bis(trimethylsilyl) trifluoroacetamide; CSI, cholesterol-saturated fat index; DHA, docosahexaenoic acid; DPA, docosapentaenoic acid; EPA, eicosapentaenoic acid; FAME, fatty acid methyl esters; MUFA, monounsaturated fatty acids; NL, neutral lipids; PC, phosphatidylcholine; PL, polar lipids; PUFA, polyunsaturated fatty acids; SFA, saturated fatty acids; TI, thrombogenic index; TL, total lipids; TLC, thin layer chromatography.

* Corresponding author. Tel.: +30 210 9549251; fax: +30 210 9577050.
E-mail addresses: nickal@hua.gr (N. Kalogeropoulos), amikel@hua.gr (A. Mikellidi), tnomikos@hua.gr (T. Nomikos), chiou@hua.gr (A. Chiou).

0023-6438/$ – see front matter © 2011 Elsevier Ltd. All rights reserved.
doi:10.1016/j.lwt.2011.11.014

(fucoidan), derived from the jelly coat of sea urchin eggs showed anti-adipogenic properties in rats (Kim, Chang, & Lee, 2009).

Although certain amount of data is available for the composition of raw aquatic roes (Iwasaki & Harada, 1985; Tocher & Sargent, 1984) or their commercial preparations (Kalogeropoulos et al., 2008; Rehbein, 1985; Shirai, Higuchi, & Suzuki, 2006), the majority of these works deal with crude composition, amino acid and/or lipid and fatty acid composition. The same is true for *Paracentrotus lividus,* the most widely consumed urchin species in Mediterranean; several articles report on the lipid and fatty acid composition of *P. lividus* eggs (De la Cruz-García et al., 2000; Dincer & Cakli, 2007; Gago, Luis, & Repolho, 2008) and their carotenoids composition (Symonds et al., 2007).

In the present work we report the crude composition and energy content, together with several bioactive microconstituents like tocopherols, carotenoids, sterols, and squalene, as well as the lipid and fatty acid composition of commercial finfish and urchin egg products. A nutritional evaluation of the samples was also undertaken.

To our knowledge this is the first report on bioactive microconstituents of commercial marine eggs. As these products represent highly valued foods in regional and international markets, both their composition and their health and nutritional evaluation are of significant importance.

2. Materials and methods

2.1. Reagents and chemicals

Butylated hydroxytoluene (BHT), boron trifluoride in methanol solution (14% BF_3 in MeOH), β-carotene, 5-α-cholestane, cholesterol, campesterol, β-sitosterol, stigmasterol, methyl heneicosanoate, methyl eicosapentaenoate, methyl docosahexaenoate, and squalene were purchased from Sigma Chemicals. A standard mixture of 37 fatty acid methyl esters (FAME) was purchased from Supelco. α-Tocopherol, δ-tocopherol and bis-(trimethylsilyl)-trifluoroacetamide (BSTFA) were obtained from Aldrich, and γ-tocopherol from Fluka. Hexane, chloroform, propanol-2 and acetonitrile of HPLC grade, as well as silica gel G were from Merck. All the other solvents and reagents used were of analytical grade.

2.2. Sample preparation

Commercial products of lumpfish (*Cyclopteris lumpus*), Atlantic salmon (*Salmo salar*), trout (*Oncorhynchus mykiss*), smoked herring (*Clupea harengus*), mullet (*Mugil cephalus*) (Greek "Avgotaracho") and sea urchin (*P. lividus)* eggs, were purchased from local supermarkets. In addition, as sea urchin roes are usually commercialized and consumed raw, fresh sea urchin (*P. lividus)* roes collected from Megara Bay, Greece, kept in ice and transferred daily in the local fish market, were obtained. The origin and additives of samples are given in Table 1. Three to four separate samples from every marine egg product were obtained and were pooled to prepare composite samples for the analyses. Commercial products were canned in glass jars except "Avgotaracho" which is marketed covered by several layers of beeswax, which is added melted during its preparation (Kalogeropoulos et al., 2008). According to their labels all the commercial products were provided well within their shelf lives. After arriving in the laboratory, the mullet roe was homogenized by gentle mixing by hand after removing the wax cover, and together with fresh sea urchin roe, were stored in sealed glass jars under nitrogen. All samples were subsequently stored at −40 °C.

2.3. Proximate composition and energy determination

Total lipids (TL) were isolated from 20 g portions of the aquatic egg samples, according to Folch, Lees, and Sloane Stanley (1957). After solvent removal by evaporation under vacuum (Speedvac) the extracted lipids were weighed, diluted in hexane containing 20 mg/L BHT, and stored in Teflon coated screw-capped glass vials under nitrogen at −40 °C.

Ash, protein and energy content were determined in samples of aquatic eggs which had been freeze-dried for 48 h (Heto Lyolab 3000, Heto-Holten, Allerod, Denmark), and kept at −40 °C. Freeze-drying served also for moisture determination, as the water content of freeze-dried samples was less than 3%. The ash content of freeze-dried samples was determined by a programmable muffle furnace. Total protein was calculated from the Kjeldahl nitrogen using a 6.25 conversion factor. Kjeldahl nitrogen was determined by a Buchi (Zurich, Switzerland) digesting and steam distillation apparatus. The gross energy content was determined by means of an adiabatic calorimeter (IKA C4000, Analysentechnik, Heitersheim, Germany).

2.4. Lipid analysis

Lipids were separated into polar (PL) and neutral (NL) fractions by counter current distributions (Galanos & Kapoulas, 1962). After solvent evaporation, the two fractions were weighed and the PL fraction was dissolved in BHT containing chloroform:methanol (1:1, v/v), while the NL fraction was dissolved in BHT containing petroleum ether. The lipid solutions were stored in Teflon coated screw-capped vials under nitrogen at −40 °C. The composition of NL and PL was determined by thin layer chromatography (TLC) as described by Kalogeropoulos et al. (2008).

2.5. Fatty acids analysis

Fatty acids of TL, NL and PL were determined by GC/MS, as methyl esters (FAME). FAME were prepared after hot saponification

Table 1
Proximate composition[a] and energy content of commercial fish and urchin egg products on a fresh weight basis.

Species	Origin of product	Additives	Moisture (g/100 g)	Crude protein[b] (g/100 g)	Total lipids (g/100 g)	Energy (kcal/100 g)	Ash (g/100 g)
Lumpfish	Denmark	Salt, monosodium glutamate, citric acid, colorants	69.8 ± 1.3d	16.8 ± 0.7b	4.2 ± 0.3a	131.7 ± 8.7b	6.2 ± 0.2e
Salmon	Germany	Salt	45.0 ± 2.7a	32.7 ± 0.3e	8.9 ± 0.6d	293.8 ± 11.5f	4.7 ± 0.2d
Trout	Germany	Salt, yeast extract, glucose syrup, citric acid	54.8 ± 1.5b	30.6 ± 0.4d	9.0 ± 0.4d	214.2 ± 12.4e	4.2 ± 0.3c
Smoked herring[c]	Italy	Salt, corn starch, lemon juice, squid ink, citric acid, xanthan gum, sodium benzoate	80.1 ± 1.1e	8.7 ± 0.4a	4.5 ± 0.5a	106.2 ± 11.5a	1.5 ± 0.4a
Mullet[d]	Greece	Salt, beeswax cover	48.1 ± 2.3a	33.7 ± 1.1e	17.8 ± 0.7e	302.1 ± 17.3f	2.1 ± 0.2b
Canned sea urchin	Spain	Salt	61.6 ± 3.4c	28.1 ± 1.9c	5.4 ± 0.3b	183.7 ± 10.8d	5.8 ± 0.4e
Fresh sea urchin	Greece	–	70.5 ± 3.8d	16.5 ± 1.7b	7.8 ± 0.2c	161.9 ± 13.5c	3.8 ± 0.2c

[a] Each value represents the mean ± standard deviation of 3 analyses.
[b] Protein calculated from Kjeldahl nitrogen P = N × 6.25.
[c] Contains 40% w/w of smoked herring eggs.
[d] "Avgotaracho", wax covered, salted and dried intact ovaries; means within each column not sharing lowercase letters are significantly different (Duncan's test, $p \leq 0.05$).