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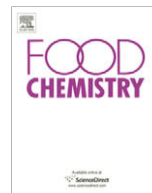

Pan-frying salmon in an eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) enriched margarine prevents EPA and DHA loss

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ABSTRACT

Salmon is a major dietary source of eicosapentaenoic acid (EPA, 20:5n-3) and docosahexaenoic acid (DHA, 22:6n-3) in North America, but the impact of pan-frying with various culinary fats on fatty acid content is not comprehensive. The fatty acid composition of Atlantic salmon after pan-frying with a novel EPA+DHA enriched margarine was examined. Pan-frying without oil, with canola oil and with stick margarine resulted in significantly lower levels of EPA+DHA (738 ± 181, 723 ± 94 and 704 ± 75 mg per 100 g salmon, respectively) as compared with raw salmon (1202 ± 191 mg per 100 g salmon). Pan-frying with EPA+DHA margarine prevented the decrease of EPA+DHA in salmon (924 ± 162 mg per 100 g salmon). Pan-frying salmon results in decreases of EPA+DHA, but a novel EPA+DHA enriched margarine can attenuate the decrease and possibly increase EPA and DHA intakes in North Americans.

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1. Introduction

The dietary intake of n-3 polyunsaturated fatty acids, particularly eicosapentaenoic acid (EPA, 20:5n-3) and docosahexaenoic acid (DHA, 22:6n-3), are associated with cardiovascular and neurological benefits (Arts, Ackman, & Holub, 2001; Freeman et al., 2006; Psota, Gebauer, & Kris-Etherton, 2006). Salmon, a fatty fish known to possess relatively high levels of EPA and DHA, is a main dietary source of n-3 polyunsaturates in the diets of North Americans (Mahaffey, Clickner, & Jeffries, 2008; Mozaffarian, Stein, Prineas, & Siscovick, 2008). Food and nutrient databases often report on the fatty acid content of raw salmon, which is not commonly consumed in western cultures, and the effect of cooking techniques and the impact of culinary fats are typically overlooked.

Pan-frying of salmon meat involves the application of heat through a surface in order to raise the temperature of the fish to a palatable level, and also to eliminate any microbial pathogens that may be present. Culinary fats (oils, margarines, butter) are often used to coat the surface of the frying pan to prevent sticking to the pan and to provide a particular flavour. It has been observed that EPA and DHA diffuse from salmon meat into the culinary fat, resulting in decreased levels of these fatty acids in the cooked salmon (Gladyshev, Sushchik, Gubanenko, Demirchieva, & Kalachova, 2006; Sioen et al., 2006). This effect has been attributed to the negligible EPA and DHA content of culinary fats studied in such investigations to date, resulting in a diffusion gradient that drives EPA and DHA into the culinary fat. The diffusion of the dominant fatty acid subclass present in the culinary fat (saturated or monounsaturated in margarine and olive oil, respectively) into the salmon meat during cooking results in higher levels of these types of fatty acids after frying (Sioen et al., 2006).

The development and commercial availability of microencapsulated fish oils has resulted in several food products that have been fortified with EPA+DHA. One such product is an EPA+DHA enriched margarine, which can be used for pan-frying in the same manner as traditional margarine. The EPA+DHA content of such an enriched margarine may prevent the diffusion and loss of EPA and DHA from salmon during pan-frying. The purpose of the present study was to examine the effects of pan-frying in hydrogenated canola/soybean oil margarine, liquid canola oil or a novel EPA+DHA margarine on the fatty acid composition of Atlantic salmon (Salmo salar) in comparison with raw salmon and salmon cooked with no culinary fat.

2. Methods

2.1. Salmon and culinary fat

Atlantic salmon and culinary fats were purchased from a supermarket store in the Kitchener-Waterloo, Ontario (Zehrs Markets, Loblaw Companies Ltd., Brampton, ON). A single, large fillet of farmed Atlantic salmon was cut into 13 pieces weighing

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73.03 ± 3.56 g. Three pieces were fried in each culinary fat, three pieces were fried with no fat, and one piece was analysed raw. Fish was not frozen prior to cooking, but was refrigerated in sealed bags for no more than six hours.

Culinary fats included hydrogenated canola/soybean oil stick margarine (No Name, Loblaw Companies Ltd., Brampton, ON), liquid canola oil (Canola oil, President's Choice, Loblaw Companies Ltd., Brampton, ON) or a novel EPA+DHA enriched margarine (Becel Omega-3 plus, Unilever Canada, Toronto, ON) and were refrigerated until used.

2.2. Cooking procedure

The pan used for cooking was made of cast iron and had a depth of 4 cm and a diameter of 23 cm. The pan surface temperature was raised to 190°C prior to cooking. The amount of culinary fat used for each sample was approximately equal to 5% of the mass of its companion fish sample as described previously (Sioen et al., 2006). The fat was then placed on the pan and spread over the entire cooking surface by tilting. The initial temperature of the fish sample was recorded using a digital meat thermometer (President's Choice, Loblaw Companies Ltd., Brampton, ON) and the fish placed on the pan. The sample was flipped every 4 min for 16 min, upon which time the final internal temperature was recorded and the sample was weighed. After the salmon sample had been removed from the pan, any cooking residue present was collected and stored at −80 °C until lipid analysis was completed.

2.3. Homogenisation and lipid extraction

A 50 g portion of each salmon sample was homogenized in a 1 l blender (Waring Laboratory & Science, Torrington, CT) chilled with ice to prevent heat generation. A total of 75 ml of deionized water was added to facilitate the homogenisation process. A 50 ml aliquot of salmon homogenate was collected and stored at −80 °C until fatty acid analysis was completed. Freezing salmon samples in this manner does not affect the presence or stability of fatty acids (Polvi, Ackman, Lall, & Saunders, 1991).

Lipids were extracted from salmon homogenates (Bligh & Dyer, 1959) in triplicate with an internal standard (22:3n-3 ethyl ester, Nu-Chek Prep, Elysian, MN). Briefly, a 1:1 (v:v) mixture of chloroform:methanol containing 50 μg/ml BHT (butylated hydroxytoluene) and 0.188 M KCl solution was added to each sample and the organic phase was collected. A second extraction was performed on the aqueous phase via the addition of chloroform and the organic phases from the two extractions were combined. For the culinary fat and cooking residue samples, 0.10 g was measured in triplicate, 25 mg of internal standard added, and lipids extracted (Folch, Lees, & Sloane Stanley, 1957). Briefly, 2:1 (v:v) chloroform:methanol was added instead of 1:1. Collected lipid extracts were percolated through Pasteur pipettes stuffed with cotton and containing approximately 2 cm of anhydrous sodium sulphate (Na_2SO_4) to remove water and any other non-lipid contaminants as described previously (Denomme, Stark, & Holub, 2005).

After percolation, an aliquot of the total lipid extract was placed in a separate 5 ml test tube with screw cap, and evaporated under nitrogen gas at room temperature (25 °C). Fatty acid methyl esters were generated by transesterification of the lipid extracts by heating to 90 °C in the presence of 14% boron triflouride in methanol (Morrison & Smith, 1964). Fatty acid methyl esters were collected for analysis by gas chromatography.

2.4. Gas chromatography analysis of fatty acid methyl esters

Fatty acid compositions were determined by gas chromatography of fatty acids similar to those described previously (Masood, Stark, & Salem, 2005). Briefly, a Varian 3900 gas chromatograph (Varian Inc, Mississauga, ON, Canada) with a DB-FFAP 15 m × 0.10 mm inner diameter × 0.10 μm film thickness capillary column (J & W Scientific, Agilent Technologies, Palo Alto, CA) was used with H_2 as the carrier gas at a flow rate of 30 ml/min. The flame ionisation detector was set at 300°C with air and nitrogen make-up gas flow rates of 300 ml/min and 25 ml/min respectively with a split ratio of 200:1 and a sampling frequency of 50 Hz. The autosampler injection volume was 1 μl with the injector temperature set at 250 °C. The temperature program was as follows: initial, 150 °C with a 0.25 min hold; ramp: 35 °C/min–200°C, then by 8 °C–225°C with a 3.2 min hold, then by 80 °C/min–245°C with a 15 min hold. Total run time for each sample was 23 min.

2.5. Statistics

Statistical analysis was performed using SPSS for windows version 15.0. Comparisons of the physical characteristics of the salmon pieces before and after cooking were examined by paired t-tests. The fatty acid composition of salmon samples and culinary fats were compared by the general linear model procedure followed by Tukey's post-hoc analysis. The changes in the fatty acid composition of the culinary fats before and after cooking were examined by paired *t*-tests.

3. Results

3.1. Effect of pan-frying on mass and temperature

The internal temperature of the salmon increased (8.4 ± 0.5 °C to 70.8 ± 2.0°C, $p < 0.0001$) and the mass of salmon samples decreased (73.0 ± 0.6 g–61.0 ± 1.8 g, $p < 0.001$) during cooking with no differences between conditions. The mass of culinary fat added was 3.7 ± 0.1 g, representing 5.0 ± 0.1 percent of the mass of the salmon pieces, and there were no significant differences in mass of culinary fat added between the groups.

3.2. Effects of pan frying on fatty acid content of salmon meat

The concentration of total fatty acids in the salmon tended to decrease with cooking but did not reach statistical significance (Table 1). There were no significant changes in the concentration of total saturates, total monounsaturates and total n-6 polyunsaturates, however there was a significant reduction in the salmon concentration of n-3 polyunsaturates with pan-frying as compared with raw samples except for salmon fried in the EPA+DHA enriched margarine. In terms of relative percent of total fatty acids, the total n-3 polyunsaturates also decreased after frying with no fat or in stick margarine but not when fried with canola or EPA+DHA margarine (Table 2). Relative percentages of n-6 polyunsaturates tended to increase when fried in canola, stick margarine and EPA+DHA margarine, while frying without fat resulted in a significant increase in the relative percentage of saturated fatty acids.

Changes in individual n-3 polyunsaturates with pan-frying were largely dependent on the content of the corresponding fatty acid in the culinary fat used during cooking. Levels of α-linolenic acid (ALA, 18:3 n-3) in salmon decreased as compared with raw samples (164 ± 61 mg/100 g) when ALA was not present as in the case of no culinary fat (104 ± 33 mg/100 g) and with a normal stick margarine (90 ± 12 mg/100 g). When appreciable levels of ALA were present in the culinary fat, ALA levels in salmon did not decrease as with cooking with canola oil (153 ± 26 mg/100 g) and EPA+DHA margarine (170 ± 25 mg/100 g). However, the relative percentage of ALA actually increased when fried in canola oil and EPA+DHA margarine (Table 2). Frying salmon with no fat, in regular margarine and in

Table 1
Fatty acid concentrations of salmon samples before and after cooking without or with culinary fats (mg/100 g of salmon).[A]

	Raw ($n = 3$)	Pan-fried			
		No fat ($n = 9$)	Canola oil ($n = 7$)	Stick margarine ($n = 8$)	EPA+DHA margarine ($n = 7$)
12:0	n.d.^{b}	11 ± 5^{a}	n.d.^{b}	8 ± 2^{a}	1 ± 3^{b}
14:0	378 ± 59^{a}	322 ± 112^{a,b}	253 ± 41^{a,b}	238 ± 36^{b}	278 ± 55^{a,b}
16:0	2090 ± 318^{a}	1691 ± 534^{a,b}	1406 ± 197^{a,b}	1381 ± 187^{b}	1616 ± 306^{a,b}
18:0	554 ± 88^{a}	426 ± 121^{a,b}	382 ± 48^{b}	377 ± 45^{b}	448 ± 85^{a,b}
20:0	n.d.	3 ± 6	n.d.	7 ± 17	n.d.
Total SFA	3023 ± 466	2476 ± 781	2057 ± 293	2033 ± 267	2347 ± 441
14:1	n.d.^{b}	15 ± 7^{a}	1 ± 4^{b}	11 ± 2^{a}	n.d.^{b}
16:1	994 ± 155^{a}	771 ± 275^{a,b}	679 ± 110^{a,b}	615 ± 96^{b}	726 ± 163^{a,b}
18:1	4706 ± 713	3611 ± 1239	3488 ± 610	3366 ± 481	3965 ± 741
20:1 n-9	405 ± 47^{a}	310 ± 116^{a,b}	315 ± 65^{a,b}	233 ± 26^{b}	293 ± 60^{a,b}
22:1 n-9	12 ± 22^{a,b}	30 ± 19^{a}	4 ± 8^{b}	28 ± 13^{a}	n.d.^{b}
Total MUFA	6117 ± 921	4738 ± 1642	4489 ± 768	4253 ± 606	4984 ± 959
18:2 n-6	1677 ± 280	1239 ± 417	1265 ± 189	1125 ± 164	1446 ± 265
18:3 n-6	n.d.^{b}	16 ± 10^{a}	n.d.^{b}	18 ± 13^{a}	n.d.^{b}
20:2 n-6	103 ± 11^{a}	66 ± 23^{b}	71 ± 11^{a,b}	58 ± 9^{b}	78 ± 16^{a,b}
20:3 n-6	8 ± 15^{a,b}	28 ± 12^{a,b}	n.d.^{b}	35 ± 29^{a}	5 ± 13^{b}
20:4 n-6	98 ± 9^{a}	65 ± 17^{b}	57 ± 14^{b}	58 ± 7^{b}	69 ± 18^{b}
Total n-6 PUFA	1887 ± 283	1413 ± 474	1393 ± 213	1293 ± 180	1598 ± 299
18:3 n-3	164 ± 61^{a}	104 ± 33^{b}	153 ± 26^{a}	90 ± 12^{b}	170 ± 25^{a}
20:5 n-3	620 ± 100^{a}	388 ± 110^{b}	383 ± 50^{b}	358 ± 44^{b}	458 ± 86^{a,b}
22:5 n-3	242 ± 91^{a}	130 ± 73^{b}	166 ± 22^{a,b}	150 ± 24^{a,b}	156 ± 60^{a,b}
22:6 n-3	581 ± 91^{a}	351 ± 72^{b}	340 ± 44^{b}	346 ± 31^{b}	466 ± 82^{a}
Total n-3 PUFA	1608 ± 269^{a}	973 ± 282^{b}	1042 ± 139^{b}	944 ± 110^{b}	1250 ± 233^{a,b}
Total PUFA	3495 ± 551^{a}	2386 ± 754^{b}	2435 ± 352^{a,b}	2237 ± 290^{b}	2848 ± 530^{a,b}
Total fatty acids	12635 ± 1928	9599 ± 3169	8980 ± 1394	8524 ± 1162	10179 ± 1922

[A] Values presented as means ± SD. Values with a different subscript within a row are significantly different by Tukey's post-hoc test ($p < 0.05$) after significant *F* value by one-way ANOVA. SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; n.d., not detected.

Table 2
Mass percent of fatty acids in salmon samples before and after cooking without or with culinary fats (% of total fatty acids).[A]

	Raw ($n = 3$)	Pan-fried			
		No fat ($n = 9$)	Canola oil ($n = 7$)	Stick margarine ($n = 8$)	EPA+DHA margarine ($n = 7$)
12:0	n.d.^{b}	0.1 ± 0.1^{a}	n.d.^{b}	0.1 ± 0.1^{a}	0.1 ± 0.1^{b}
14:0	3.0 ± 0.1^{b}	3.3 ± 0.2^{a}	2.8 ± 0.1^{b}	2.8 ± 0.5^{b}	2.7 ± 0.6^{b}
16:0	16.5 ± 0.2^{b}	17.7 ± 0.9^{a}	15.7 ± 0.4^{b}	16.2 ± 0.1^{b}	15.9 ± 0.3^{b}
18:0	4.4 ± 0.1	4.5 ± 0.3	4.4 ± 0.2	4.4 ± 0.2	4.4 ± 0.2
20:0	n.d.	0.1 ± 0.1	n.d.	0.1 ± 0.2	n.d.
Total SFA	23.9 ± 0.3^{b}	25.9 ± 1.4^{a}	23.0 ± 0.7^{b}	23.9 ± 0.3^{b}	23.1 ± 0.3^{b}
14:1	n.d.^{b}	0.14 ± 0.06^{a}	0.02 ± 0.05^{b}	0.13 ± 0.01^{a}	n.d.^{b}
16:1	7.9 ± 0.1^{a}	7.9 ± 0.4^{a}	7.6 ± 0.8^{a,b}	7.2 ± 0.2^{b}	7.1 ± 0.3^{b}
18:1	37.2 ± 0.1^{c}	37.4 ± 0.8^{b,c}	38.8 ± 1.8^{a,b,c}	39.5 ± 0.5^{a}	39.0 ± 0.5^{a,b}
20:1 n-9	3.2 ± 0.1^{a,b}	3.2 ± 0.3^{a,b,c}	3.5 ± 0.3^{a}	2.8 ± 0.2^{c}	2.9 ± 0.3^{b,c}
22:1 n-9	0.1 ± 0.2^{a,b}	0.3 ± 0.2^{a}	0.05 ± 0.11^{b}	0.3 ± 0.1^{a}	n.d.^{b}
Total MUFA	48.4 ± 0.4	49.0 ± 1.3	49.9 ± 1.5	49.9 ± 0.6	49.0 ± 0.9
18:2 n-6	13.3 ± 0.2^{b}	12.9 ± 0.2^{b}	14.1 ± 0.4^{a}	13.2 ± 0.2^{b}	14.2 ± 0.2^{a}
18:3 n-6	n.d.^{b}	0.15 ± 0.09^{a,b}	n.d.^{b}	0.21 ± 0.16^{a}	n.d.^{b}
20:2 n-6	0.82 ± 0.06^{a}	0.68 ± 0.03^{b}	0.79 ± 0.02^{a}	0.68 ± 0.02^{b}	0.77 ± 0.03^{a}
20:3 n-6	0.08 ± 0.14^{a,b}	0.3 ± 0.2^{a,b}	n.d.^{b}	0.4 ± 0.4^{a}	0.04 ± 0.11^{b}
20:4 n-6	0.78 ± 0.07	0.70 ± 0.09	0.63 ± 0.09	0.68 ± 0.02	0.68 ± 0.11
Total n-6 PUFA	14.9 ± 0.2^{b,c}	14.7 ± 0.3^{c}	15.5 ± 0.5^{a,b}	15.2 ± 0.3^{b}	15.7 ± 0.3^{a}
18:3 n-3	1.28 ± 0.32^{b}	1.09 ± 0.04^{b}	1.71 ± 0.13^{a}	1.06 ± 0.01^{b}	1.68 ± 0.10^{a}
20:5 n-3	4.9 ± 0.1^{a}	4.1 ± 0.3^{c}	4.3 ± 0.1^{b,c}	4.2 ± 0.1^{b,c}	4.5 ± 0.2^{a,b}
22:5 n-3	1.9 ± 0.1^{a,b}	1.3 ± 0.7^{b}	1.9 ± 0.1^{a}	1.8 ± 0.1^{a,b}	1.5 ± 0.4^{a,b}
22:6 n-3	4.6 ± 0.1	3.9 ± 0.8	3.8 ± 0.2	4.1 ± 0.2	4.6 ± 0.6
Total n-3 PUFA	12.7 ± 0.5^{a}	10.4 ± 1.4^{c}	11.6 ± 0.5^{a,b,c}	11.1 ± 0.2^{b,c}	12.3 ± 0.5^{a}
Total PUFA	27.7 ± 0.7^{a,b}	25.1 ± 1.5^{c}	27.2 ± 0.9^{a,b}	26.3 ± 0.3^{b,c}	28.0 ± 0.7^{a}

[A] Values are means ± SD. Values with a different subscript within a row are significantly different by Tukey's post-hoc test after significant *F* value by one-way ANOVA ($p < 0.05$). SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; n.d., not detected.

canola oil resulted in significant decreases in the concentrations of EPA+DHA in salmon whereas this reduction was not significant when fried in EPA+DHA margarine (Fig. 1).

3.3. The effects of pan-frying salmon on the fatty acid content of culinary fats

The fatty acid concentration of the culinary fats used in the pan-frying procedures changed during cooking (Table 3, Fig. 2). Prior to cooking, EPA and DHA was detected only in the EPA+DHA margarine (30.6 ± 0.3 mg/5 mg EPA+DHA), while n-3 docosapentaenoic acid (DPAn-3, 22:5n-3) was not present in any of the culinary fats. After cooking, EPA and DHA concentrations increased significantly in all three culinary fat residues. Although there was no significant difference in the EPA+DHA content in the cooking oil residues (Fig. 2), the increase in EPA+DHA was significantly less in the EPA+DHA margarine (80 ± 30 mg/5 g) as compared with the canola oil (116 ± 24 mg/5 g) and stick margarine (138 ± 22 mg/5 g)

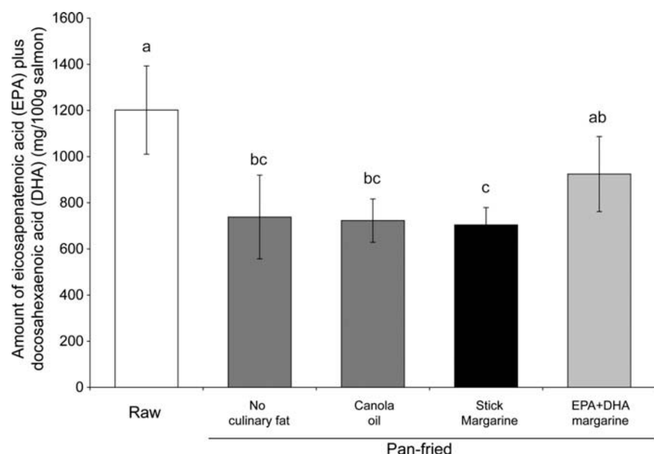

Fig. 1. The amount of EPA plus DHA in salmon samples before and after cooking without or with culinary fats. Columns that do not share an alphabetical superscript are significantly different by Tukey's post-hoc test ($p < 0.05$) after a significant *F* value by one-way ANOVA.

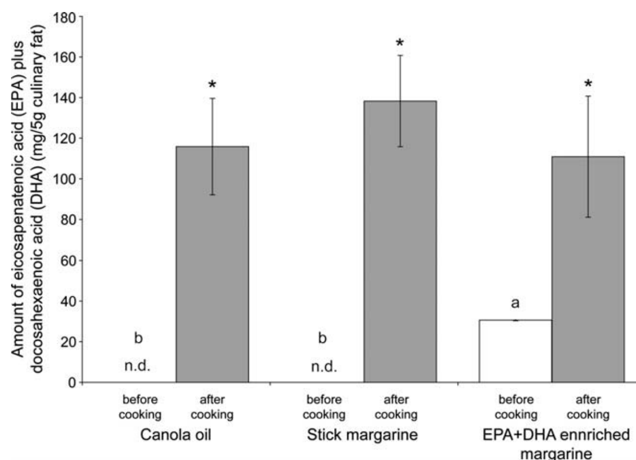

Fig. 2. The amount of EPA plus DHA in culinary fats before and after cooking. Culinary fats before cooking were examined by one-way ANOVA with different alphabetical superscripts indicating significant differences by Tukey's post-hoc test ($p < 0.05$) after a significant *F* value. Asterisks (∗) indicate a significant difference in a culinary fat after cooking as compared with before cooking by independent *t*-test ($p < 0.05$). n.d., not detected.

($p < 0.05$). DPAn-3 concentrations increased in the culinary fat residues after cooking (Table 3).

4. Discussion

In the present study, pan-frying salmon in an EPA+DHA enriched margarine prevented the significant decreases in EPA+DHA that occurred when salmon was fried with no fat or with other culinary fats. The process of frying the salmon resulted in a significant decrease in the concentration of EPA+DHA as indicated by the change from raw to salmon fried with no culinary fat. Cooking with culinary fats did not prevent losses of EPA and DHA or even ALA unless these fatty acids were present in the culinary fat in significant quantities.

Cooking results in a diffusion of fat from the salmon, with culinary fat either impeding the diffusion from the salmon, diffusing into the salmon, or contributing to the composition as a surface coating. The diffusion of polyunsaturated fatty acids from salmon may occur more readily as compared with other fatty acids by the nature of their low melting point. The relative percentage of polyunsaturates decreased significantly during cooking without fat, with no effect on monounsaturates while relative saturated fatty acids increased significantly relative to raw salmon. Furthermore, the higher degree of unsaturation and resulting lower melting point of n-3 polyunsaturates appears to make the n-3 polyunsaturates more susceptible to cooking related losses as

Table 3
Fatty acid compositions of culinary fats before and after cooking salmon (mg/5 g of culinary fat).[A]

	Canola oil		Stick margarine		EPA + DHA margarine	
	Before cooking ($n = 3$)	After cooking ($n = 9$)	Before cooking ($n = 3$)	After cooking ($n = 6$)	Before cooking ($n = 3$)	After cooking ($n = 9$)
14:0	4.3 ± 0.3[b]	53 ± 10*	4.0 ± 0.7[b]	64 ± 10*	43 ± 1[a]	69 ± 11*
16:0	223 ± 7[b]	381 ± 37*	434 ± 49[a]	641 ± 62*	390 ± 20[a]	461 ± 50*
18:0	103 ± 3[b]	127 ± 8*	393 ± 47[a]	393 ± 42	105 ± 10[b]	125 ± 17
20:0	32.8 ± 0.7[a]	19.7 ± 2.9*	15.0 ± 1.4[c]	14.2 ± 1.9	23.5 ± 3.0[b]	16.4 ± 3.4*
22:0	17.3 ± 0.7	10.7 ± 1.7*	14.6 ± 1.4	12.4 ± 1.9	13.6 ± 2.3	9.0 ± 1.9*
Total SFA	393 ± 4[c]	603 ± 51*	868 ± 103[a]	1134 ± 111*	648 ± 40[b]	739 ± 78
16:1	15.9 ± 1.4[b]	146 ± 28*	6.6 ± 0.6[c]	170 ± 27*	19.8 ± 0.8[a]	117 ± 39*
18:1	3138 ± 82[a]	2373 ± 214*	2341 ± 204[b]	2586 ± 294	2264 ± 189[b]	2021 ± 272
20:1 n-9	58.8 ± 2.0[a]	103 ± 11*	6.8 ± 1.0[c]	93 ± 13*	39.9 ± 4.3[b]	77 ± 17*
22:1n-9	0.9 ± 1.5	1.3 ± 2.4	n.d.	3.8 ± 4.2	n.d.	2.8 ± 2.4*
Total MUFA	3220 ± 84[a]	2632 ± 202*	2355 ± 206[b]	2857 ± 323*	2326 ± 193[b]	2223 ± 271
18:2 n-6	1021 ± 28[a]	791 ± 68*	366 ± 28[c]	537 ± 56*	913 ± 58[b]	803 ± 104
18:3 n-6	n.d.[b]	4.7 ± 0.5*	3.9 ± 0.4[a]	7.7 ± 1.3*	0.3 ± 0.6[b]	3.6 ± 1.1*
20:2 n-6	4.2 ± 0.1[a]	15.6 ± 2.6*	n.d.[b]	15.9 ± 2.6*	3.2 ± 0.8[a]	11.3 ± 3.7*
20:3 n-6	n.d.	6.1 ± 1.4*	n.d.	7.2 ± 1.2*	n.d.	4.1 ± 1.7*
20:4 n-6	n.d.	11.5 ± 2.4*	n.d.	13.6 ± 2.5*	n.d.	9.3 ± 3.1*
Total n-6 PUFA	1025 ± 28[a]	831 ± 65*	370 ± 28[c]	585 ± 62*	917 ± 59[b]	834 ± 102
18:3 n-3	452 ± 13[a]	273 ± 41*	24.6 ± 1.9[c]	42.2 ± 4.8*	309 ± 17[b]	230 ± 45*
20:5 n-3	n.d.[b]	65 ± 13*	n.d.[b]	78 ± 13*	16.9 ± 0.7[a]	62 ± 17*
22:5 n-3	n.d.	32.5 ± 6.7*	n.d.	39.4 ± 6.4*	n.d.	26 ± 10*
22:6 n-3	n.d.[b]	51 ± 10*	n.d.[b]	60 ± 10*	13.7 ± 0.8[a]	49 ± 13*
Total n-3 PUFA	452 ± 13[a]	422 ± 29	24.6 ± 1.9[c]	221 ± 33*	340 ± 17[b]	368 ± 35
Total PUFA	1478 ± 41[a]	1254 ± 93*	395 ± 30[c]	806 ± 100*	1257 ± 76[b]	1201 ± 133
Total fatty acids	5090 ± 129[a]	4488 ± 304*	3617 ± 336[b]	4797 ± 508*	4232 ± 309[b]	4163 ± 467

[A] Values are means ± SD. Values with a different subscript within a row are significantly different by Tukey's post-hoc test ($p < 0.05$) after significant *F* value by one-way ANOVA. SFA, total saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; n.d., not detected.

compared with n-6 polyunsaturates. In the present study, salmon cooked with no culinary fat as compared with raw salmon demonstrated a 39.5% decrease in n-3 polyunsaturates concentrations, a 25.1% decrease in n-6 polyunsaturates, a 22.5% decrease in monounsaturates and an 18.1% decrease in saturates. These results suggest that melting points of fatty acids is an important determinant of fat diffusion during pan-frying.

The use of culinary fat during the frying of salmon can influence the final fatty acid composition of the cooked salmon as higher levels of linoleic acid (18:2n-6) and oleic (18:1n-9) have been demonstrated in salmon fried in sunflower oil and olive oil, respectively (Gladyshev et al., 2006; Sioen et al., 2006). The diffusion of individual fatty acids from salmon during frying can be prevented or replaced by using a culinary fat that contains the individual fatty acids of interest. This is clearly demonstrated in the n-3 polyunsaturates. ALA concentrations did not change when ALA was present in the culinary fat (canola oil and EPA+DHA margarine), but decreased significantly when absent in the culinary fat (stick margarine and no fat). EPA and DHA in salmon decreased significantly with pan-frying except when cooked in the EPA+DHA margarine, which attenuated the loss. Decreased levels of DPAn-3 were similar after cooking with the culinary fats as none of the fats contained DPAn-3. Further support for the diffusion of n-3 polyunsaturates from the salmon is shown by examining the increases of these fatty acids in the culinary fats after cooking. EPA, DPAn-3 and DHA increased in all the culinary fats, while ALA increased in the stick margarine (that had low original levels) and decreased in the canola oil and EPA+DHA margarine.

The present study is limited as only a single type of fish was examined and a limited set of cooking conditions and culinary fats were examined. Salmon was selected for the study due to role as a main dietary source of EPA and DHA in North Americans (Mahaffey et al., 2008; Mozaffarian et al., 2008). Pan-frying was selected for examination as a cooking technique suitable for the use of a novel EPA+DHA margarine as a cooking fat. A limited set of conditions were employed so that a single salmon would serve as the source of all samples in the studies. This was done to control for considerable variability in the fatty acid composition of fish as a result of seasonal differences (Hamre, Lie, & Sandnes, 2003; Nordgarden, Torstensen, Froyland, Hansen, & Hemre, 2003; Olsson, Olsen, Carlehog, & Ofstad, 2003), as well as differences in wild versus farmed (Weaver et al., 2008) including differences in farmed fish diets (Grant et al., 2008). In the present study, uniform salmon sample sizes of approximately 70 g were used. Given that size of cooked sample, particularly the thickness and ratio of surface area to mass, may influence diffusion rates (Sioen et al., 2006), extrapolation of the present results is limited. Estimates of diffusion of fatty acids from the salmon were possible in the present study design, but attempts to estimate losses due to fatty acid oxidation during cooking were not made. Long-chain highly unsaturated fatty acids are susceptible to oxidation during cooking techniques such as pan-frying (Al Saghir et al., 2004; Echarte, Zulet, & Astiasaran, 2001). It was also not possible to discriminate between the prevention of the diffusion of fatty acids from the salmon, the diffusion of fatty acids into the salmon, or the contributions of fatty acids from culinary fat surface coatings. Further studies including surface washings, analysis of interior versus exterior cooked samples and/or the inclusion of fatty acid isotopes in the culinary fat could provide insights.

5. Conclusions

The use of an EPA+DHA enriched margarine during pan-frying of salmon prevents the significant decrease in EPA+DHA concentrations. The EPA and DHA in the margarine may prevent the diffusion of these long-chain n-3 fatty acids out of the salmon meat and into the culinary fat, or the EPA+DHA levels in the culinary fat surface coating may offset diffusion of EPA+DHA from the salmon. The cooking associated decreases in EPA+DHA were not prevented when canola oil and stick margarine were utilized as culinary fats. The use of novel EPA+DHA enriched margarine during cooking may be a strategy to maintain EPA and DHA levels of long-chain n-3 containing fish and possibly increase EPA+DHA levels of fish with low levels of n-3 polyunsaturates. An examination of the production of potentially detrimental oxidative by-products of long-chain polyunsaturated fatty acids under various cooking conditions with these novel culinary fats is required. The use of EPA+DHA margarine during cooking as a strategy to increase EPA+DHA consumption in North Americans warrants further investigation.

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