

Review

Review on retention of long-chain omega-3 polyunsaturated fatty acids (EPA and DHA) in fish as affected by cooking methods

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Abstract

Fatty acids are essential building blocks for the structural components of the cells, tissues, organs, and certain biologically active substances synthesis. Omega-3 polyunsaturated fatty acids are long-chain fatty acids essential for several biological functions including oxidative stress reduction and cardiovascular safety. Diet rich in omega-3 fatty acids is well acknowledged as beneficial to one's health and well-being. For the development of balanced diets, the nutrient content of raw and cooked foods is crucial. However, cooking method, animal age, and carcass characteristics might affect nutrient retention during cooking, and these factors are often unique to specific countries. This review thus provides a general overview of several cooking effects on long-chain omega-3 retention in fish. It can be concluded that the DHA and EPA's true retention values are correlated to the impact of different cooking treatments.

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Introduction

Fatty acid (FA) is an essential part of the human body as an energy source crucial for a healthy metabolism, and acts as essential cell membrane components and eicosanoid hormone precursor (Ng, 2006; Pereira *et al.*, 2013; Kaur *et al.*, 2014; Sokoła-Wysoczańska *et al.*, 2018). The FA chain length may consist of two and up to more than 30 double bonds (Calder, 2008). Fatty acids with one double bond are called monounsaturated FA (MUFA). In contrast, FA with more than one double bond are categorised as polyunsaturated FA (PUFA) (Ratnayake and Galli, 2009).

The critical roles of PUFA and MUFA in preventing diseases might be a great tool that helps decrease the prevalence of chronic diseases worldwide. For example, omega-3 and omega-6 polyunsaturated are long-chain PUFA that are crucial due to their multiple biological roles such as reducing

oxidative stress and cardiovascular protection. According to WHO (2017), an estimated 17.7 million people or 31% of all global deaths in 2015 were due to cardiovascular diseases (CVD). By 2030, approximately 23.6 million people are expected to die from CVD, primarily heart diseases and stroke, accounting for 29% of global mortality (Bhardwaj *et al.*, 2016; WHO, 2017).

Adequate consumption of particular FA, especially omega-3 PUFA, can relieve ulcerative colitis, menstrual pain, and joint pain; and prevent several chronic diseases and illnesses such as CVD (Delgado-Lista *et al.*, 2012; Williams *et al.*, 2013; Barbalho *et al.*, 2016). On the other hand, inadequate fat intake is related to certain disorders such as abnormality in the liver, impaired growth rate, immune function, depression, and skin dryness. Therefore, the Food and Agriculture Organization (FAO) recommends that adults should consume below 10% of diets from saturated FA (SFA), and

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between 20 - 35% of total fat energy consistently, in compliance with the latest expert consultation guidelines (WHO and FAO, 2003). In addition, the recommendations are to ensure that the total energy intake, essential FA, and fat-soluble vitamins consumption for most individuals is sufficient, to take cholesterol less than 300 mg/day, and to consume trans FA as low as possible (Russo, 2009; FAO, 2010).

Nevertheless, PUFA must be acquired from the diet as the human body cannot produce them. Marine foods are excellent dietary sources of SFA, MUFA, and PUFA. In addition to shellfish, fish is one of the most abundant marine foods with PUFA, minerals, and vitamins (Russo, 2009; Marimuthu *et al.*, 2012). However, the composition of PUFA differs by fish species, and the significant presence of long-chain PUFA in fish and marine lipids is highly vulnerable to oxidation (Hsieh and Kinsella, 1989; Choo *et al.*, 2018). In addition, PUFA are thermosensitive compounds that can be altered by heat during cooking (Choo *et al.*, 2018), besides the food colour, taste, and flavour enhancement that could attract the human.

Omega-3 polyunsaturated fatty acids (n-3 PUFA)

There are three primary *n-3* PUFA, namely alpha-linolenic acid (ALA, 18:3*n-3*), eicosapentaenoic acid (EPA, 20:5*n-3*), and docosahexaenoic acid (DHA, 22:6*n-3*) that are consumed in foods, and utilised by the body (Ng, 2006). The long-chain PUFA, DHA and EPA, can be produced from ALA, the parent omega-3 FA (Grosso *et al.*, 2014). Most of the beneficial effects of omega-3 PUFA consumption are related to positive effects on human health. Omega-3 FA have been demonstrated to lower the risk of chronic diseases including heart diseases and cancers, lower inflammation, and protect against neurodegeneration (Calder, 2008; Asmah *et al.*, 2014). In the human diet, the EPA and DHA are primarily found in marine products such as fish, shellfish, and aquatic freshwater products (Choo *et al.*, 2018). In contrast, ALA can be obtained mainly from vegetable oils, walnut, and cereals (Russo, 2009). Salmon, mackerel, sardine, anchovy, trout, and tuna are among the fish rich in omega-3 consumed by consumers in the United States (Jin, 2016). In a study by Abd Aziz *et al.* (2013), the EPA, DHA, and ALA levels were significantly greater in longtail shad, yellow stripe scad, and moon trout, respectively.

Sufficient intake of omega-3 FA contributes to decreasing the risk of coronary heart diseases (CHD), blood pressure, plasma triglycerides, and optimal cognitive output as they are highly concentrated in the brain, which help to improve cognitive control and visual sharpness early on (Mozaffarian and Wu, 2011; Muldoon *et al.*, 2014). Therefore, the omega-6/omega-3 FA balance is critical for good health. The American Heart Association suggested that adults should consume 500 mg DHA+EPA equivalent (provided by two servings weekly of oily fish) for primary prevention of CHD, 1,000 mg daily as secondary prevention for those with documented CHD, and 2 - 4 g daily for those with high triglycerides (to lower by 20 to 40% triglyceride) (Kris-Etherton *et al.*, 2003). It is also recommended for children under the age of 10 years, healthy adults, and pregnant women to take 100 to 250, 250, and 300 mg, respectively, of DHA+EPA daily (FAO, 2010). The *n-3:n-6* close to 1 is suggested for optimal health (Simopoulos, 2008).

Fish consumption in Malaysia

The Malaysian Adults Nutrition Survey (2002 - 2003) showed the prevalence of consumption of marine fish among rural and urban Malaysian populations is relatively high, accounting for 51% and 34%, respectively (Norimah *et al.*, 2008). The high rate of fish consumption in Malaysia shows that the awareness of the nutritional content of fish, particularly PUFA, among most Malaysians is very high (Choo *et al.*, 2018). Meanwhile, increased fish consumption is proposed for omega-3 (*n-3*) FA to lower the CVD risk factor (Raatz *et al.*, 2013). In line with this, the Dietary Guidelines for Americans in 2010 (USDA, 2010) recommend replacing 10% of SFA with MUFA and PUFA. The guideline also suggested an average intake of EPA and DHA by 1,750 mg weekly (250 mg daily), via 8 ounces or more seafood consumption weekly (USDA, 2010; Aranceta and Pérez-Rodrigo, 2012). Therefore, eating more unsaturated FA, especially fish high in omega-3 FA, offer a better way to combat diseases, and should be suggested to be incorporated into a daily diet.

Apart from the high content of PUFA, fish and seafood also contain high alpha-tocopherol concentration, making them highly recommended for regular consumption (Azrina *et al.*, 2015). An investigation by Azrina *et al.* (2015) also found that

almost all fish in the study had low cholesterol levels, except for several fish like sixbar grouper (*Epinephelus fasciatus*), long-tailed butterfly ray (*Gymnura* sp.), yellowstripe scad (*Selaroides leptolepis*), cuttlefish (*Sepia officinalis*), large-scale tongue sole (*Cynoglossus arel*), and longtail shad (*Hilsa macrura*). The nutritional value of fish is possible to be calculated by the quantification of the protein and fat content (rich in PUFAs), and by the determination of the concentration of minerals such as calcium (Ca) and iron (Fe), as well as zinc (Zn) (Toppe *et al.*, 2007). In a study by Nurnadia *et al.* (2013), the microminerals amount in all samples other than oyster were below the allowable levels, with copper marginally greater than the FAO/WHO (1984) limit, but below the Malaysian Food Regulations (1985) limit; and zinc content marginally greater than the Malaysian Food Regulations (1985) limit, but below the FAO/WHO limit (1984).

Nutrient retention in food

Food composition databases (FCDB) present detailed information on foods' nutritional composition, and are typically country specific (Finglas *et al.*, 2014; Marconi *et al.*, 2018). These databases are available in various formats such as paper-based tables, and often referred to as "Food Composition Tables" or as nutrient databases or databanks, in an electronic-based form (Uusitalo *et al.*, 2011). The Malaysian Food Composition table, for example, provided nutrient information primarily based on raw food. However, limited data is available for cooked food, and none for omega-3 FA are available. Therefore, the database might not give explicit information on nutrients as it does not include the composition data for cooked food.

According to the technical report from National and Health Morbidity Survey in 2019 (MOH, 2019), 1.7 million Malaysians currently live with three major risk factors for cardiovascular: high cholesterol (16.6%), hypertension (9.3%), and diabetes (4.15%) (MOH, 2019). Therefore, a precise understanding of foods' nutrient content is also crucial for assessing communities' nutrient intakes (Murphy *et al.*, 1975) since this will allow the community to select a balanced diet. The comprehensive food composition databases should be based on empirical evidence to get accurate and reliable information on nutrient intake and health (Leclercq *et al.*, 2001). Therefore, the data from cooked food must be included in the database instead

of the available raw food information in most nutrient compositions. However, there are limitations in getting the data of all cooked foods as the analysis is laborious and time-consuming. Besides, the chemical analysis is costly and impossible to be carried out daily. Consequently, there are insufficient nutrient data for cooked/prepared foods, especially in developing countries including Malaysia.

Furthermore, FCDB have two flaws: they still differ by country, and have insufficient nutrient and bioactive compounds coverage, thus resulting in missing data. To overcome the country-specific limitations, in 2007, the EuroFIR (2005) proposal to harmonise recipe calculation procedures was finalised (EuroFIR, 2007). At the recipe level, weight yields should be applied, whereas, at the ingredient level, the nutrient retention factors should be considered (EuroFIR, 2007). Missing data can affect the FCDB integrity. Therefore, according to the FCDB principle, missing values in FCDB must never be assigned with zero value since the nutrient value may be zero (Westrich *et al.*, 1994; Greenfield and Southgate, 2003). The commonly used approach to resolve the missing data in FCDB is extracting data from another nation's FCDB, where a reference back to the source may or may not be feasible (Finglas *et al.*, 2014; Ispirova *et al.*, 2019). The proportion remaining in the cooked food as compared to the original nutrients present in the raw food is known as true retention (Murphy *et al.*, 1975). However, advances in food composition studies have allowed the nutrient composition of cooked/prepared food and dishes from raw food to be measured, if nutrient retention factors are available (USDA, 2007). This condition implies that retention variables can be used to calculate the nutrient values in the absence of analytical data for cooked foods.

Nutrient retention and cooking yield are two factors that need to be considered when looking into nutrient availability for cooked food. It is needed for formulating a balanced and healthy diet (Van Heerden and Strydom, 2017). The use of factors for weight yield (fat/water and alcohol) and nutrient retention is directly related to the composite food recipe measurement procedures. In this way, it is possible to measure packaged foods' nutrient content from their ingredients for publication in FCDB labelling and special diets.

To date, the USDA nutrient retention table is the primary reference for the nutrient retention database for the US and international food

composition (USDA, 2007). It consists of a retention factor of around 290 foods for 16 vitamins, eight minerals, and alcohol (USDA, 2007). However, there is no retention factor for omega-3 FA currently available. The available retention data for other individual food constituents, particularly niacin, biotin, folate, pantothenic acid, vitamins (E, K, and B₁₂), FA, amino acids, and cholesterol are incomplete, and should be considered as crude estimates. The retention factor of 1 is preliminarily recommended for all other constituents, for which there are no data available at present (EuroFIR, 2006). Hence, the nutrient retention of the prepared foods or dishes will also vary, always country-specific, as noted in the literature (Van Heerden and Strydom, 2017).

Malaysia, unlike the US, has not yet developed its table of nutrient retention. Therefore, a correct interpretation of findings is critical to avoid false interpretations, over- or underestimation, and unwarranted conclusions; and only can be achieved by having reliable analytical data (Peters *et al.*, 2007). Hence, it is vital to establish a validated method of FA analysis, especially for the most sensitive FA like omega-3, as initial steps to obtain the detailed analytical data of omega-3 composition and its retention in the future. There are many techniques used to analyse FA such as high-performance liquid chromatography (HPLC), high-performance liquid chromatography-mass spectrometry (HPLC-MS), gas chromatography-mass spectrometry (GC-MS), and gas chromatography flame ionisation detector (GC-FID) (Juárez *et al.*, 2010). Of all the techniques, GC-FID is the most often used technique as a quantification method for FA (Godswill *et al.*, 2014; Zhang *et al.*, 2015).

Long-chain omega-3 fatty acids (EPA and DHA) as affected by cooking methods and their retention

Cooking produces physical and chemical benefits to foods that are compatible with observed human dietary adaptations such as chewing, increased digestibility, and increased net energy content of daily human consumption of plant and animal foods (Carmody and Wrangham, 2009). Foods, including meat and fish, become nutritious and digestible upon cooking (Uran and Gokoglu, 2014). Cooking methods are always country-preferable; poaching, microwaving, steaming, broiling, and oven-baking are the standard cooking methods for fish cooking in western countries. In Malaysia, deep-fried fish,

followed by fish cooked in thick and/or thin chilli gravy, fish curry, and fish cooked in coconut gravy are Malaysians' most preferred cooking styles (Ahmad *et al.*, 2016). Japanese typically eat raw fish (*e.g.*, sashimi and sushi), whereas broiled fish and fried fish are commonly eaten in the Western hemisphere (Nanri *et al.*, 2020).

Besides the various cooking methods available worldwide, reducing nutrient loss and ensuring that microbial pathogens are destroyed are the aim of cooking, as well as to produce food with the desired sensory qualities like colour, texture, aroma, and taste (Uran and Gokoglu, 2014; Abraha *et al.*, 2018; Gök *et al.*, 2019). Recent studies have shown several ways to increase the supply of healthy nutrients by thoroughly choosing the cooking method. One of the most common methods for achieving microbial stability in various meat products is heat treatment (Gök *et al.*, 2019). Cooking methods also influence meat quality characteristics such as yield, tenderness, juiciness, taste, and palatability by causing protein, starch, lipid, and other minor components to alter physically and biochemically (Pathare and Roskilli, 2016).

Different cooking methods could lead to food weight changes such as loss or gain of solids, liquids, or both (Murphy *et al.*, 1975). In addition, some nutritional values in foods might be affected; for example, thermo-sensitive compounds in foods such as EPA, DHA, and fat-soluble vitamins can influence the heat applied during cooking treatments (Choo *et al.*, 2018). Previous studies have shown that long-chain PUFAs are vulnerable to oxidation during heating and other culinary treatments. The heat treatment results in undesirable modifications, such as the depletion in food nutritional values, owing to lipid oxidations and specific components of the protein fraction alterations (Uran and Gokoglu, 2014). Our previous study found that the DHA and EPA were retained highly in yellow stripe scads and Japanese threadfin bream fillet when using steaming and baking in foil methods as compared to raw fillet (Choo *et al.*, 2018).

Various factors might affect the retention of nutrients in food such as cooking, animal age, and carcass characteristics, and they are unique to specific countries (Van Heerden and Strydom, 2017). Methods and temperatures in cooking play a vital role in nutrient retention, and might be applied differently using dry- and moist-cooking methods. The process of decomposition, polymerisation, and redox

reactions of FA might occur in food samples like fish during cooking. Essential FA, DHA, and EPA, are easily oxidised and diminished upon preparation and further processing for cooking. Cooking might also change the nutritional composition of fish species, and might determine the quality of essential FA in the consumed product (Gladyshev *et al.*, 2007). Processing is a term that refers to the treatment of food from the time it is collected/harvested until it is consumed (Aberoumand, 2014). Traditional domestic processing such as washing, peeling, cutting, chopping, soaking, sun-drying, refrigerating, and freezing before cooking might affect the nutrient content in food, in addition to cooking methods. Refrigeration and freezing are two common approaches for preserving fish at home (Ruiz-Rodriguez *et al.*, 2008). A study by Yamamoto and Imose (1989) found that the lipid in the dark meat of sardine (*Sardinops melanosticta*) is fragile and prone to oxidation after a week of storage in the refrigerator (Yamamoto and Imose, 1989). The processing

methods also result in chemical composition changes. The changes can primarily be reflected in denaturation, coagulation, reduced digestibility of proteins, oxidation, and vitamin loss (Abraha *et al.*, 2018).

In fish, the proteins, fats, minerals, vitamins, and sensory features such as taste, colour, appearance, and texture are the most compositional components influenced by processing methods. Thermo-sensitive compounds such as fat-soluble vitamins and PUFA in fish fillets can be affected by cooking (Abraha *et al.*, 2018; Choo *et al.*, 2018). Furthermore, during preparation, the temperature, surface touch, fish size, and initial fat content of the fish samples might also influence the FA composition of the fish fillet (Choo *et al.*, 2018).

Few studies have been carried out on the cooking effect and FA retention in fish. The effect of cooking methods by dry- and moist-heat on FA retention, including DHA and EPA, in fish is shown in Table 1.

Table 1. Effects of moist- and dry-heat cooking methods on fatty acid retention including omega-3 PUFA in fish.

No.	Cooking method/sample	Finding	Conclusion	Reference
1.	Boiling, deep-frying, steaming, grilling: Farmed rainbow trout	(i) PUFAs were preserved in steamed fish. (ii) <i>n</i> -3 and <i>n</i> -6 FA increased in grilled sample. (iii) Frying decreased the EPA, DHA, and linoleic acid significantly. (iv) Boiling decreased the EPA significantly. (v) α -linolenic acid (C18:3n3) significantly decreased in all cooking processes except for grilled samples. (vi) The fried and boiled samples did not yield any linoleic acid (C18:3n6).	Preservation of omega-3 FA was better in grilled and steamed samples as compared to the others.	Cano-Estrada <i>et al.</i> (2018)
2.	Deep frying, grilling, baking in foil, steaming: Salmon, yellow stripe scad, and Japanese threadfin bream	(i) The steaming method retained DHA+EPA (mg/100 g) the highest as compared to raw, baked, baked-in-foil fillet, grilled, and deep-fried samples. (ii) Total PUFA in steamed and BIF fish was higher than grilled and fried fish. (iii) DHA and EPA decreased significantly in deep-fried and grilled fish. (iv) The saturated fatty acids (SFA) were higher in deep-fried samples (955 mg/100 g) as compared to the others (499 - 612 mg/100 g). (v) MUFA retention decreased significantly in the baking-in-foiled sample.	Steaming and baking in foil were the best cooking methods for retention of DHA and EPA in yellow stripe scad fillet.	Choo <i>et al.</i> (2018)
3.	Steaming, oven, canned, microwaving:	(i) Omega-3 increased significantly in steamed (21%) and canned (18.97%) salmon fish.	All cooking treatments applied showed no significant changes in	Bastías <i>et al.</i> (2017)

	Salmon and Chilean jack mackerel	(ii) Omega-3 increased significantly in canned Chilean fish (17.84%), microwaved (19.96%), and steamed samples (18.34%). (iii) No significant difference in PUFA changes in cooked salmon as compared to the raw sample. (iv) PUFA increased significantly in cooked Chilean jack as compared to the raw sample.	fatty acids in salmon and Chilean jack mackerel.	
4.	Steaming: Bluefish and rainbow trout	(i) Total PUFA and MUFA increased significantly ($p < 0.05$) but decreased in SFA ($p > 0.05$) in steamed rainbow trout as compared to bluefish. (ii) Steaming did not alter total PUFA, SFA, and MUFA in bluefish.	Steaming was suggested as a mild and less aggressive cooking method.	Stancheva <i>et al.</i> (2014)
5.	Broiling, baking, frying: Chinook salmon, common carp, white sucker, and walleye fish	(i) Cooking treatments had no significant effect on EPA+DHA content in fish ($p > 0.05$). (ii) Higher $n-6$ and MUFA in fried samples were due to the cooking oil used (canola).	Broiling or baking was considered the healthiest method for PUFA retention.	Neff <i>et al.</i> (2014)
6.	Pan frying, oven-baking, boiling: Salmon	(i) No significant effects on omega-3 and omega-6 in pan-fried, boiled, and oven-baked fish ($p > 0.05$). (ii) The peroxide concentrations increased in pan-fried and baked salmon samples. (iii) The MDA concentration increased in pan-frying method	Frying and baking reduced the PUFA in cooked fish.	Leung <i>et al.</i> (2018)
7.	Poaching, steaming, microwaving, pan-frying (no oil-added), oven-baking, deep frying (in sunflower oil): New Zealand king salmon	(i) All cooking treatments except deep frying did not alter the omega-3 in fish significantly. (ii) Deep-fried decreased the DHA, EPA and DPA significantly, and increased the omega-6 content significantly ($p < 0.05$). (iii) No significant difference between SFA in cooked and raw fish samples. (iv) Deep-fried salmon showed a substantial decrease in SFA linoleic acid used as frying oil.	No significant changes in omega-3 (DHA, EPA, and DPA) in all cooked fish except deep-fried fish.	Larsen <i>et al.</i> (2010)
8.	Baking, boiling, frying, grilling: Striped snakehead fish	(i) The fried sample showed significantly higher fat, followed by grilling, baked, boiled, and raw fish fillets ($p < 0.05$). (ii) Total fat content in boiled, baked, and grilled fish did not differ significantly from raw fish fillet ($p > 0.05$).	Grilling was the best for healthy eating.	Marimuthu <i>et al.</i> (2012)
9.	Grilling, boiling, frying, microwaving: Freshwater mud eel	(i) All cooking processes decreased the omega-3 PUFA content in the studied sample. (ii) Boiling decreased the omega-3 PUFA significantly ($p < 0.05$). (iii) SFA increased slightly in all cooked sample.	Boiling decreased the PUFA significantly as compared to the other cooking methods.	Islam <i>et al.</i> (2020)
10.	Pressurise:	(i) The omega-3 content of pressurised fish decreased slightly as compared to raw fish ($p > 0.05$).	Pressurised fish can be used to store or total fat and omega-3	Asmah <i>et al.</i> (2014)

	Yellow-tail catfish and long-tail shad	(ii) The total content of pressurised fish decreased but was not significant as compared to raw fish ($p > 0.05$).	preservations in human daily dietary intake.	
11.	Frying, grilling, baking: Anchovy	(i) PUFA and MUFA higher in baked fish as compared to fried and grilled fish. (ii) SFA decreased in grilled and fried samples significantly but not in the baked sample. (iii) MUFA increased significantly ($p < 0.05$) in the baked and fried samples, and decreased in the grilled sample. (iv) Frying did not alter the fatty acid concentration in the sample ($p > 0.05$).	Baking was the best cooking method for anchovy.	Uran and Gokoglu (2014)
12.	Boiling, roasting, grilling: Farmed meagre	(i) The SFA, MUFA, and PUFA contents increased but not significantly in grilled and roasted fish ($p > 0.05$). (ii) No significant difference was found in the $n-3:n-6$ ratio in boiled, grilled, and roasted fish as compared to raw fish. (iii) Culinary processes produced no EPA and DHA degradation.	Grilled treatment was the best culinary treatment to retain EPA and DHA in farmed meagre.	Costa <i>et al.</i> (2013)
13.	Boiling, roasting, frying: Indian mackerel	(i) The total fat significantly increased in fried fish (13%) as compared to boiled (7.5%), roasted (9.6%), and raw fish (5%) samples. (ii) The fat content did not differ significantly in boiled and roasted fish as compared to the raw sample.	Roasted and boiled fish were beneficial for health.	Solanki and Lende (2020)
14.	Baking in aluminium foil, broiling, microwaving: European sea bass	(i) Total MUFA/Total SFA decreased in oven broiled fish, and increased in the microwaved sample. (ii) Baking in foil did not change the fatty acids compositions in fish. (iii) The highest true retention values (TRVs) was found in the baking-in-foiled sample. (iv) Broiling and microwaving produced the lowest TRVs.	(i) Baking in foil, broiling, and microwaving had modest effect on fatty acids. (ii) Baking-in-foil provided the highest TRVs as compared to broiling and microwaving.	Badiani <i>et al.</i> (2013)
15.	Pan-frying, oven-baking, and grilling: Carp, cod, pike, and herring	(i) The amount and ratio of $n-3$ and $n-6$ PUFA decreased in codfish. (ii) Baking had the most negligible effect on PUFA in pike, herring, and carp fish. (iii) Oven-baking was the mildest heat treatment for PUFA preservation.	(i) The effect of heat treatments on lipid compositions in fish tissue was species-specific. (ii) Oven-baking preserved PUFA as compared to grilling and pan-frying.	Schneedorferová <i>et al.</i> (2015).

An investigation by Cano-Estrada *et al.* (2018) examined the impact of four of Mexico's most popular cooking methods such as steaming, deep-frying, baking, and grilling on rainbow trout FA content. For boiling, the samples were boiled for 10

min at 94°C in a pot of 350 mL of water. For frying, the samples were immersed in 180°C safflower oil for 5 min. For steaming, the samples were wrapped in 25 × 25 cm aluminium foil, and placed in the middle of the 85 - 93°C pot layers for 10 min. For grilling, the

samples were grilled on a low-heat flat pan for 12 min until the samples' internal temperature reached 75 - 80°C. Results showed that the DHA and linoleic acid increased significantly in the grilled samples. The PUFA were retained in the steamed samples. In fried and boiled samples, the EPA (C20:5n3) decreased significantly. Similarly, DHA (C22:6n3) showed a significant decrease when cooked with the frying method. The total amount of PUFA showed a significant difference due to potential FA oxidation to heat exposure. However, there was no significant difference in n3:n6 ratio for all cooking treatments applied. In conclusion, steaming and grilling were suggested as the best methods to preserve the PUFA content of cooked fish.

The excellent effect of moist-heat cooking was shown in a study by Choo *et al.* (2018) which revealed that the PUFA retention was significantly lower in fish fillets cooked with two moist-heat cooking methods as compared to raw fish fillets ($p < 0.05$). The long-chain omega-3 DHA+EPA (mg/100 g) retention was the highest in steamed yellow stripe scad fillets than raw fillets, followed by foil-baking, grilling, and deep-frying. Nevertheless, the EPA and DHA levels decreased significantly in fish fillets cooked with dry-heat methods (deep-frying and grilling). The study then suggested the two moist-heat cooking methods, steaming and baking in foil, as the best cooking methods for DHA and EPA retentions in related samples.

The study is similar to an investigation on moist-heat cooking on FA retention by Bastías *et al.* (2017). They investigated the FA retention of salmon (*Salmo salar*) and Chilean jack mackerel fillet (*Trachurus trachurus*) comparable in sizes (35 - 45 cm). The fishes were cooked with four different cooking methods; microwaving, oven-baking, canning, and steaming. The samples were baked in an oven at 250°C for 25 min, whereas the samples were cooked at 1,500 W for 12 min for the microwaving method. The electronic steamer was used to steam the sample at 25 min with $83.5 \pm 2^\circ\text{C}$ as the final sample temperature; and for the canning procedure, the canned fish was put in tin cans and then filled with 2% saline solution. An exhaustor was used to remove oxygen, and samples were finally sealed and sterilised using the autoclave for 45 min until the central temperature reached 121°C. As a result, omega-3 FA exhibited significant changes in some treatments. In contrast to the controls, a significant increase in omega-3 was observed in steamed

(21.1%) and canned (18.97%) salmon samples. In addition, a significant increase was also observed in the canned Chilean jack mackerel by 17.84%, followed by microwaved sample by 19.96%, and steamed sample by 18.34% as compared to the controls.

Stancheva *et al.* (2014) studied the effect of steaming on rainbow trout and bluefish, and found that steaming increased PUFA by 45.8% in rainbow trout as compared to raw fish. Steaming, however, had a slight effect on PUFA in bluefish. The PUFA:SFA ratio increased by 8.33%, but did not affect bluefish. They then suggested the less aggressive steaming method as the ideal cooking technique for a healthy dietetic plan.

However, a few studies found that the DHA and EPA contents did not vary significantly in the fish samples after cooking. For example, a study by Neff *et al.* (2014) found no significant difference in the EPA and DHA contents (mg/100 g DW) in cooked fillets as compared to raw fillets ($p > 0.05$). In the analysis, skinless and boneless fillets of fish species (chinook salmon, common carp, lake trout, white sucker, and walleye) from the Great Lakes tributaries or connecting rivers were cooked using broiling, baking, and frying methods. As a result, it was found that cooking treatments had little effect on n-3 fatty acids in the cooked samples as compared to raw parts of the same fillet. However, n-6 and MUFA contents in frying and baking in canola oil samples increased as compared to the uncooked and broiled samples. The increased n-6 and MUFA contents in fried samples were possibly due to the cooking oil (canola). Canola oil consists only of ALA, and does not contain either EPA or DHA for its n-3 FA content (Neff *et al.*, 2014). In conclusion, baking and broiling were the healthiest forms of cooking as they contribute to lower levels of less desirable fatty acids than frying (Neff *et al.*, 2014).

Comparable to Neff *et al.* (2014), a study by Leung *et al.* (2018) showed no significant reduction in PUFA in boiled, pan-fried, and oven-baked salmon as compared to raw salmon. None of the cooking methods in the salmon samples decreased either omega-3 or omega-6 PUFA significantly, thus suggesting that both PUFA were altered after cooking. However, there was an increase in oxidised products. The PUFA oxidised products, the 4-HHE and 4-HNE, PV, and TBARS showed alterations after cooking. Pan-frying increased the MDA concentration, and became the most extreme method

to cause oxidation among all cooking methods in the study (Leung *et al.*, 2018). Deep-frying also significantly decreased king salmon's DHA, EPA, and DPA ($p < 0.05$) relative to all other cooking approaches (Larsen *et al.*, 2010). This study determined the effect of different cooking methods on FA profile, particularly the omega-3 FA, in king salmon fish according to conventional New Zealand cooking methods; poach in sunflower oil, pan-fry, steam, oven bake, no added-oil deep-fry, and microwave. No significant changes were reported in the percentage of DHA, EPA, or DPA across all the cooking methods applied to king salmon samples, other than the deep-fried method. The omega-6, however, significantly increased in the deep-fried sample, which might be due to the linoleic acid content in sunflower oil used. For the SFA level, there was no significant difference between all cooking treatments applied to the sample. Overall, the omega-3 FA stabilised in all cooking methods, and responded differently to heat treatments.

Marimuthu *et al.* (2012) published similar findings that showed no substantial difference in fat contents between baked, grilled, boiled, and raw fish fillets in the study ($p > 0.05$). The lipid content was observed to be significantly higher (10.7 ± 1.85) in fried fillets, followed by grilled fillets (8.6 ± 0.67), as compared to baked, boiled, and raw snakehead fish fillet ($p < 0.05$). However, the increase in fat content in the samples might be attributed to the absorption of cooking oil used for cooking treatment (frying), as stated previously by Neff *et al.* (2014). The increase in fat content was also due to oil absorption into the sample after evaporation, in which the water was partially lost (Saguy and Dana, 2003). They concluded that the best cooking method for healthy eating was grilling.

In a study by Islam *et al.* (2020), the influence of four widely used cooking methods in Bangladesh on FA compositions of freshwater mud eel muscles (FWME) was investigated. There was a slight impact on PUFA content by the different thermal processes applied (frying, grilling, boiling, and microwaving). In all cooked samples, the total omega-3 PUFAs content was lower than raw FWME muscle. During cooking, the thermal effect decreased the omega-3 PUFAs content to 10.95, 7.54, 11.11, and 11.11% in grilled, boiled, fried, and microwaved samples, respectively. On the other hand, the thermal effect during cooking treatments increased the SFA content marginally in grilled, boiled, fried, and microwaved

samples from 41.71 to 42.84, 46.07, 43.27, and 43.07%, respectively. The increase in SFAs was due to a decrease in the content of $\sum\omega$ -3 PUFAs in the samples. Zhang *et al.* (2013) and Choo *et al.* (2018) also showed an increase in SFA content when the $\sum\omega$ -3 PUFAs decreased in fish muscles. The DHA content in samples decreased significantly, whereas the EPA content was found in a low quantity ranging from 0.53 to 0.79% in raw and cooked samples. The decrease in DHA might be due to the instability of the double bond of DHA, which is vulnerable to high temperature oxidation (Islam *et al.*, 2020).

The marginal impact on total fat and omega-3 retention was also found by Asmah *et al.* (2014) who studied the effect of a pressurised method (moist-heat) on total fat and omega-3 retention in freshwater and marine fish. The omega-3 content was lower in cooked freshwater (catfish) and marine (long tail shad) fish as compared to the raw fish. For the pressurised method, the samples were cooked for 6 min using a pressure cooker. As a result, there was no significant difference observed in total fat and omega-3 content in pressurised samples as compared to the raw sample in freshwater and marine fish. The decrease in FA might be due to the action of heat (Asmah *et al.*, 2014). This study is different from Ersoy (2011) that found a significant increase in omega-3 content in cooked fish ($p < 0.05$). The increase in fat content in samples following different cooking methods might be explained by the decrease in water, thus implying a negative relationship between water content and other nutritional components (Ersoy, 2011; Cano-Estrada *et al.*, 2018).

Uran and Gokoglu (2014) found no significant differences in PUFA contents between cooking treatments applied (fried, baked, and grilled) on fish samples at two different temperatures, 160 and 180°C. The PUFA content in baked samples was higher than fried and grilled samples. Baked samples also exhibited the highest values of MUFA. The SFA decreased in grilled and fried samples significantly, but not in the baked samples. Grilling and baking were then suggested as the healthier cooking methods. However, baking was concluded as the best cooking method to preserve PUFA and MUFA in anchovy, as the texture and sensory samples were acceptable.

Costa *et al.* (2013) studied the effect of cooking methods (boiling, grilling, and roasting) on farmed meagre (*A. regius*) fish. The study observed no degradation of EPA and DHA during the culinary

processes even in the most aggressive grilling phase. The study also showed that the grilled and roasted samples increased in the SFA, MUFA, and PUFA contents. The omega-3 level was higher than the omega-6 level in all samples, except the roasted meagre. However, there was no significant difference in the omega-3:omega-6 ratio observed between raw and cooked samples. Of all the culinary treatments, grilled was the best cooking method suggested in retaining EPA, DHA, and protein in cooked fish.

Another study by Jora *et al.* (2020) investigated three cooking methods, roasting, frying and boiling, on Indian mackerel (*Rastrelliger kanagurta*) fish. Boiling was performed for 10 - 15 min at 97°C (water temperatures), whereas frying was done at 150 - 160°C for 10 - 12 min in cottonseed oil. As a result, the total fat increased significantly (13 ± 0.57) in fried fish, followed by roasted fish (9.6 ± 0.57), boiled, and raw fish. However, no significant changes were found in the total fat content between boiled and roasted fish as compared to raw fish.

Badiani *et al.* (2013) investigated the effect of three cooking treatments (microwaving, baking in foil, and oven-broiling) on European seabass (*Dicentrarchus labrax*) fish. It was shown that the highest true retention values (TRVs) of FA were found in baked in foil samples as compared to oven-broiled samples. As compared to the raw fish, baked in aluminium foil samples retained most nutrient content in the fish's flesh. The moderate TRVs came from microwaved samples, with those from oven-broiled yielding the lowest TRVs. Therefore, baking retains more FA in samples as compared to oven-broiling and microwaving.

Similar studies by Schneedorferová *et al.* (2015) showed the best effect of baking on PUFA retention, which compared pan-fried, oven-baked, and grilled effect on PUFAs content of four fish species from the Czech Republic namely pike, carp, cod, and herring. Results showed no change in the proportion of individual PUFAs in oven-baked herring. However, a decrease in the amount and ratio of *n*-3 and *n*-6 PUFA was observed in cod samples. Therefore, baking for pike, herring, and carp fish appeared to have minor effect on PUFA abundance. As demonstrated by multivariate data analysis, this study showed that the effect of heat treatments on the lipid composition of fish tissue were species-specific.

The above studies showed that moist-heat cooking methods, particularly steaming, retained PUFA effectively as compared to dry-heat methods

(Stancheva *et al.* 2014; Bastías *et al.*, 2017; Cano-Estrada *et al.*, 2018; Choo *et al.*, 2018). Frying is the oldest food preparation method, and the quickest and easiest method as compared to boiling and roasting (Bognár, 1998; Solanki and Lende, 2020). The omega-3 FA retention in fried samples is usually lower than in raw samples (Cano-Estrada *et al.*, 2018). The decrease in the sample's EPA and DHA might be attributed to the breakdown of double bonds for oxidation (Leung *et al.*, 2018). The longer the frying time, the more oxidation of PUFAs will occur, and could cause loss of PUFA afterwards. However, some studies showed an increase in fat content in fish samples using the frying method (Marimuthu *et al.*, 2012). The increase in the fat content of fried and roasted fish during cooking is related to the absorption of oil used during cooking. Besides, the increase in fat content might be due to the penetration of oil into food after water is partially lost by evaporation (Saguy and Dana, 2003). The water loss during the cooking treatments (deep-frying, steaming, boiling, and roasting) could increase the total fat-to-protein ratio in samples (Cano-Estrada *et al.*, 2018). The temperature of the heating medium, geometric dimensions of the food, desired cooking degree, and initial food temperature also contribute to frying (Sinha and Bhargav, 2015).

Boiling is one of the most common fish cooking methods besides frying (Islam *et al.*, 2020). It is also considered a mild method of cooking. Therefore, the temperature produced by boiling is the lowest as compared to other cooking methods, where the limit is the water boiling point of 100°C (Leung *et al.*, 2018). A decrease in several FA, particularly EPA, might be found after boiling (Castro-González *et al.*, 2015). These changes are related to the most common lipids released into the water throughout boiling (Cano-Estrada *et al.*, 2018). However, the decrease in FA might be lower as compared to other cooking methods. The low peroxide and reactive aldehyde levels generated in boiled fish as compared to raw fish might be explained by the short cooking time and low temperature used throughout boiling (100°C) as compared to other methods such as pan-frying and baking (Leung *et al.*, 2018).

During baking, the temperature applied is often as high as during frying. However, the EPA and DHA oxidation are lesser than frying, which could be attributed to the heat transfer method during baking (Choo *et al.*, 2018; Leung *et al.*, 2018). The air convection and radiation are the heat transfer methods

during baking, which are not as successful as boiling and frying, and contribute to lower oxidation in baking. Furthermore, the air in the oven is a thousand times less dense than water; so, there is negligible energy transfer between hot air molecules and food collisions (Leung *et al.*, 2018). Therefore, more time is needed for the food in the oven to heat up, and the internal food temperature is much lower than the surrounding temperature. As a result, lipid peroxidation is always lower in baking than frying, even though the temperature used is the same as frying, and the cooking time is longer (Leung *et al.*, 2018). The use of aluminium foil during baking also helps to increase the cooking yield, although same temperature is used during frying (Choo *et al.*, 2018).

Grilling and steaming maintained the omega-3 FA in most studies (Cano-Estrada *et al.*, 2018; Choo *et al.*, 2018; Solanki and Lende, 2020). The omega-3 retention in grilling and steaming might be attributed to the temperature used in the process, and the time needed to complete the process. The highest cooking yields were in steamed fillet, followed by baked in foil, grilled, and fried samples. The higher cooking yield in steaming might be due to the temperature applied (100°C) as compared to frying (180°C) and grilling (180°C) (Choo *et al.*, 2018).

In general, factors like cooking method, time, temperature, and species could contribute to retaining the nutritional quality of fish FA (Bastías *et al.*, 2017). For example, weight changes during cooking are related to water and fat changes (Bognár, 1998). Meanwhile, proteins, fats, vitamins, minerals, and sensory characteristics such as colour, taste, texture, and general appearance are the most compositional fish components influenced by processing methods (Abraha *et al.*, 2018). The lipid content might decrease slightly or significantly after being exposed to various cooking methods. These effects might be attributed to broken protein-lipid binding during heating, thus making the lipids easier to be removed (Larsen *et al.*, 2010), then deactivates the enzymes that prohibit the secretion of free FA from lipase action in cooked samples (Zhang *et al.*, 2013).

In addition to changes in PUFA and other FA, heating can produce different types of oxidised products that can be helpful and detrimental to human health. The high temperature applied in cooking could lead to the formation of free radicals and reactive oxygen species (ROS), and the release of products like peroxides (Leung *et al.*, 2018). Zakipour Rahimabadi *et al.* (2011) studied the impact

on total volatile profile in fish samples related to cooking treatments such as microwaving, grilling, steaming, shallow-fat frying, and reheating by microwave. Reheating by microwave increased the total volatile compounds the most (from 72.8 to 111.06 µg/kg), followed by grilling (74.25 µg/kg), steaming (112.19 µg/kg), and shallow-fat frying (92.37 µg/kg). The ketones, which resulted from oxidation, were higher in fried than steamed *S. commerson* fish.

The high degree of unsaturation in PUFA (EPA and DHA) makes them susceptible to be oxidised during culinary treatments, which in turn making them the most unstable FA (Weber *et al.*, 2008). Therefore, the unbalanced benefit-to-harm ratio in fish during cooking, such as the lower level of DHA and EPA to the higher level of trans fat used during frying, might increase the CVD risk despite the residual confounding factors of our lifestyle (Rimm and Mozaffarian, 2006).

There is also a concern about the probability of methylmercury and polychlorinated biphenyls (PCBs) accumulation in fatty fish. Methylmercury concentrations in aquatic animals are caused by mercury exposure in fish that change into methylmercury by microbial activity. The methylmercury content in fish might be affected by the environmental pollution levels, predatory nature, and lifespan of the species (Rimm and Mozaffarian, 2006). The shorter-lived species such as salmon and shellfish might have a lower mercury concentration than larger/longer-lived species such as swordfish and sharks. However, concerning this matter, women who might become pregnant, are pregnant, or nursing, and young children are suggested to consume 8 to 12 ounces lower mercury levels of seafood weekly as recommended by the 2015-2020 Dietary Guideline of Americans (FDA, 2019). Meanwhile, the European Food Safety Authority and others have pointed the risk of PCB exposure from fish intake to be similar to other meats and dairy products (Schechter *et al.*, 2001).

A good selection of cooking methods could significantly impact the overall FA and other nutrient content in cooked fish. It could also improve health and energy levels, make digestion better, and improve nutrient absorption. Baking, broiling, and steaming are recommended healthy cooking methods that have less impact on PUFA content after cooking (Neff *et al.*, 2014; Choo *et al.*, 2018). Though frying has better acceptability in terms of flavour and structure than

baking and other cooking methods, it could never be suggested to those with limitations on dietary fat intake due to diet and health concerns. Through the penetration of used oil into the sample, the fat absorption during frying could increase fish's SFA content and contribute to CVD risk factors (Uran and Gokoglu, 2014).

Conclusion

Cooking could affect the fatty acid retention in fish, especially the long-chain omega-3. Nevertheless, the effect of cooking on fatty acid retention is influenced by the temperature and time applied during cooking, and the abundance of the specific fatty acid components in the fish, which are mainly and always species-specific.

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