Chapter 5
Meat and Meat Products Enriched with n-3 Fatty Acids

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Key Points

- Meat and meat products enriched in n-3 PUFA can partially provide the daily recommended intake of n-3 PUFA, especially of long-chain n-3 PUFA.
- The fortification of meats with n-3 PUFA is interesting in ruminant meat, which is regarded as very saturated, and in pork for its high n-6 PUFA levels.
- Plant-derived sources lead to higher α-linolenic contents, whereas marine sources provide higher long-chain n-3 PUFA contents.
- The fortification of meat products can be achieved not only by dietary supplementation but also by totally or partially replacing the animal fat by other fat sources.
- Lipid oxidation is likely the main detrimental effect of the n-3 PUFA enrichment of meat and meat products; however, the use of antioxidants delays effectively lipid oxidation.

Keywords n-3 PUFA • α-Linolenic acid • Eicosapentaenoic acid • Docosahexaenoic acid • Meat • Meat products • Nutritional value

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Abbreviations

AI	Adequate intake
BHA	Butylhydroxy anisole
BHT	Butylhydroxy toluene
C14:0	Myristic acid
C16:0	Palmitic acid
C18:0	Stearic acid
C18:1	Oleic acid
C18:2n-6	Linoleic acid
C18:3n-3	Linolenic acid
C20:5n-3	Eicosapentaenoic acid
C20:6n-3	Docosahexaenoic acid
COP	Cholesterol oxidation product
DHA	Docosahexaenoic acid
DM	Dry matter
DPA	Docosapentaenoic acid
DRI	Daily recommended intake
EL	Extruded linseed
EP	Extensive-pasture system
EPA	Eicosapentaenoic acid
EFSA	European Food Safety Authority
FAO/WHO	Food and Agriculture Organization of the United Nations/World Health Organization
IC	Intensive-concentrate system
INA	Information not available
MUFA	Monounsaturated fatty acid
n-3	Omega-3 fatty acid
n-6	Omega-6 fatty acid
PLS	Protected linseed and soybean
PSM	Protected sunflower meal
PR	Partially replaced
PUFA	Polyunsaturated fatty acid
SFA	Saturated fatty acid
TR	Totally replaced

Introduction

Meat is considered to be a good source of protein with high biological value as well as of micronutrients such as minerals (iron, zinc, selenium, …) and vitamins (B_6, B_{12}, A, D, …) with a high degree of bioavailability. These micronutrients are either not present in plant-derived food or have poor bioavailability. Consequently, consuming moderate amounts of lean meat as part of a balanced diet makes a valuable contribution to the intake of essential nutrients [1]. However, some constituents of meat, especially in red meat and meat products, have been proposed to be responsible for the development of cardiovascular disease and colon cancer. These elements include the fat content and the fatty acid composition. Meat and meat products are generally classified as “high-fat products”, although the various products available differ markedly in terms of their total fat content.

Meat is generally considered to have a fat content in the range 1–20 %, depending on the retail cut and the amount of fat trimmed. Both the fat content and fatty acid composition of meat are influenced by factors such as species, breed, sex, age/weight and diet [2]. Figure 5.1 shows the fat content and fatty acid composition of some meats and meat products obtained from different species. The fat content is usually higher in processed meat products (5–40 %), where large amounts of fatty tissue are used.

In relation to the nutritional value of meat fat, amongst 35–50 % of the fatty acids are saturated, with the major saturated fatty acids (SFA) being palmitic (C16:0) and stearic (C18:0) acids. Consumption of SFA, myristic acid (C14:0) and C16:0 but not C18:0, has been associated with increased plasma cholesterol and low-density lipoprotein levels, which have been linked to an increased risk of coronary heart disease [3]. Meat contains around 30–50 % of monounsaturated fatty acids (MUFA), with oleic acid (C18:1) being both the main MUFA and the fatty acid most frequently found in meat. Oleic acid is considered hypolipidaemic as it reduces cholesterol and triglycerides in plasma [4]. The proportion of polyunsaturated fatty acids (PUFA) in meat varies in the range 7–30 %. The PUFA, linoleic (C18:2n-6) and α-linolenic (C18:3n-3) acids are regarded as essential fatty acids for humans, and n-3 PUFA are considered relevant for normal growth and development, besides delaying the development of chronic diseases including cardiovascular disease, hypertension and diabetes [5]. The beneficial health effects of n-3 PUFA are mainly attributed to long-chain n-3 PUFA such as eicosapentaenoic acid (C20:5n-3, EPA) and docosahexaenoic acid (C22:6n-3, DHA). Indeed, numerous studies have demonstrated the beneficial effects of n-3 long-chain PUFA on recognized cardiovascular risk factors, such as a reduction of plasma triacylglycerol concentrations, platelet aggregation and blood pressure [4].

The daily intake of long-chain PUFA varies from country to country. Thus, the dietary intake of EPA + DHA in Europe is around 30–420 mg/day [4], whereas in countries with high fish consumption, such as Japan, the dietary intake is approximately 1,600 mg/day [6].

Since changing the eating habits of consumers is likely to be difficult, supplementing products that are already accepted with n-3 PUFA would appear to be a more successful strategy for improving the nutritional quality of food [7]. Modifying the fatty acid composition of animal products, such as meat and meat products, by increasing the n-3 PUFA content has been suggested [8, 9] as a good means of improving their dietary value. Likewise, supplementation of animal diets with n-3 rich sources has been shown to be an efficient method for increasing the n-3 PUFA content in animal muscles [8]. In meat products, the utilization of enriched meat or the inclusion of n-3 PUFA sources during the processing of meat products are the most common ways of improving their nutritional value.

Enhancing the n-3 PUFA Composition of Meat by Dietary Supplementation

Many authors have studied different ways of changing the fatty acid composition of meat, mainly increasing the proportion of n-3 PUFA, by dietary manipulation. Most of these methods have involved the addition of vegetables sources, particularly oil seeds, which are usually a good source of α-linolenic acid. However, the conversion of α-linolenic acid to its longer chain metabolites EPA and docosapentaenoic acid (DPA) seems to be limited, thus resulting in only a small increase in the deposition of EPA and DPA in muscle. Marine products such as fishmeal, fish oil and microalgae have therefore been used to supplement animal diets since they seem to stimulate deposition of both EPA and DHA to a greater extent [10]. In ruminants, dietary fats are hydrogenated in the rumen before intestinal absorption thus meaning that muscle fatty acids in ruminants are more saturated than those in non-ruminants.

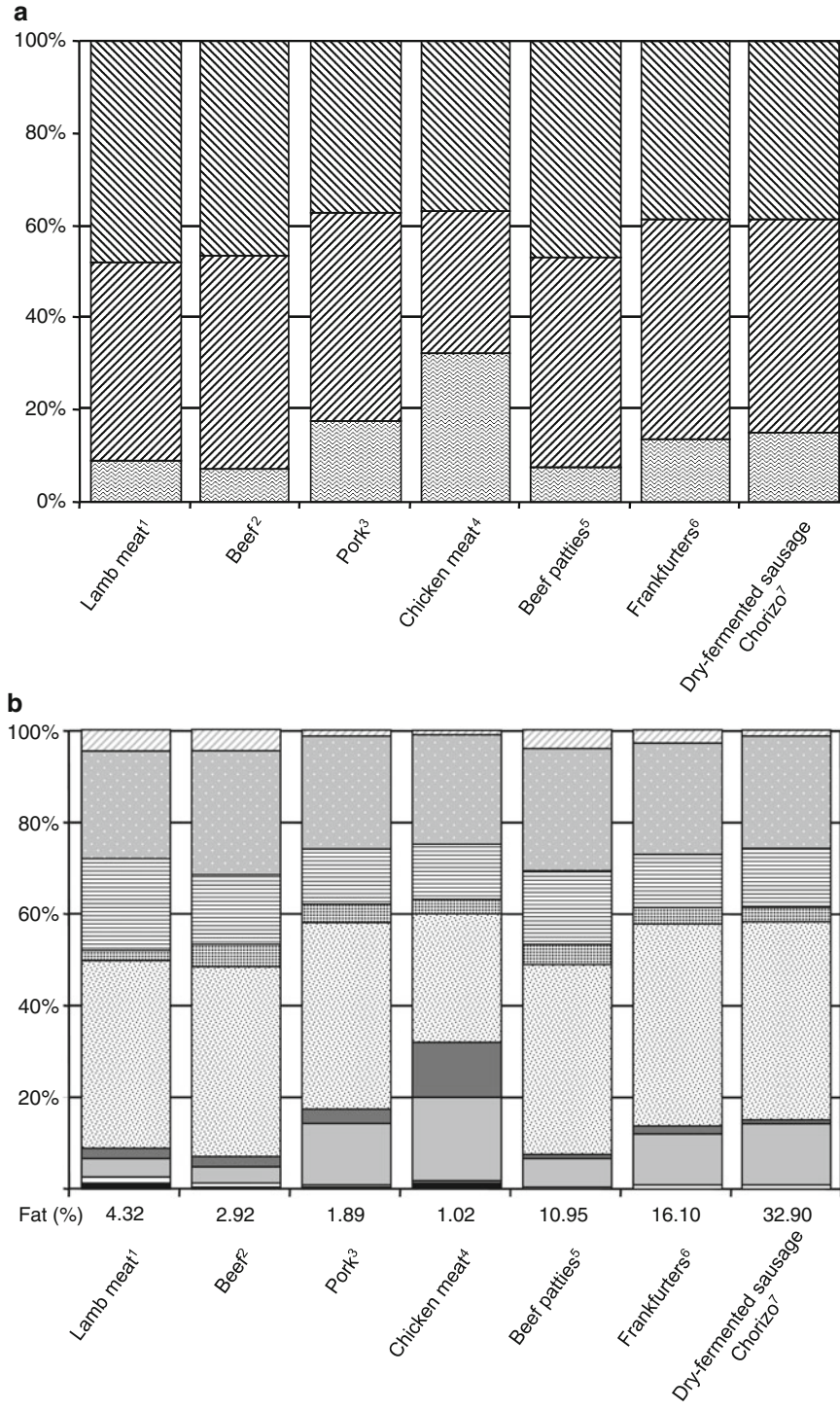

Fig. 5.1 Total fatty acid composition (**a**) and main fatty acid contents (**b**) of meats and meat products. *Legend*: *Bars* represent the contents (expressed as g/100 g of fatty acids) of: ▨ SFA, ▨ MUFA, ▨ PUFA, ▨ minor SFA, ▨ C16:0, ▨ C18:0, ▨ minor MUFA, ▨ C18:1, ▨ minor PUFA, □ C18:2n-6, □ C18:3n-3, ■ Long-chain n-3 PUFA. [1]Lamb and [2]Cattle raised at pasture and supplemented with concentrate; [3]Fattening pigs feeding *ad libitum*; [4]Cereal-based feed. Adapted from the following sources: [1][2], [2][13], [3][41], [4][18], [5][9], [6][42], [7][23]

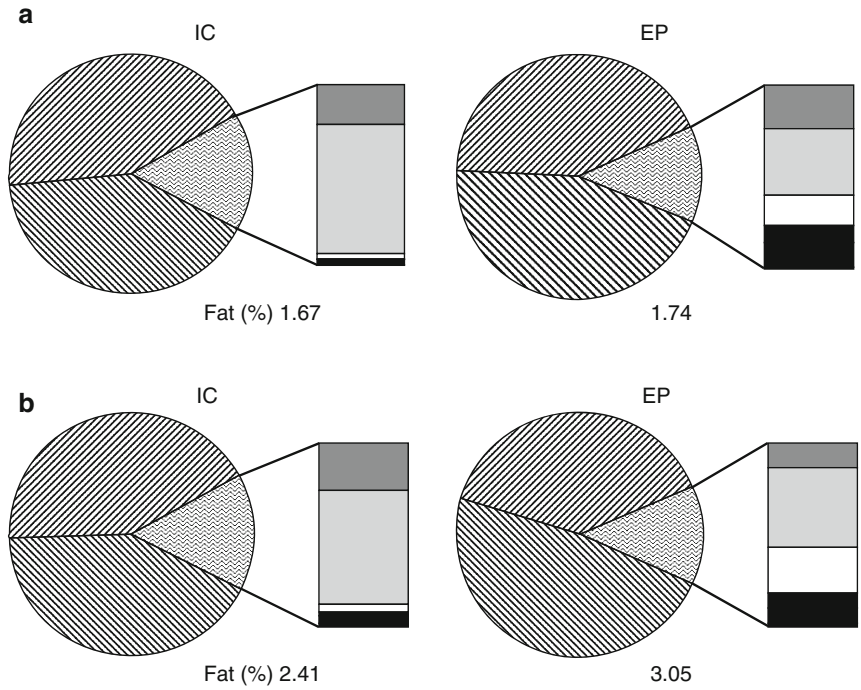

Fig. 5.2 Fatty acid composition in beef[1] (**a**) and lamb[2] (**b**) reared under intensive-concentrate (IC) or extensive-pasture (EP) systems. *Legend*: *Sectors* and *bars* represent the contents (expressed as mg/100 g of muscle) of: ▧ SFA, ▨ MUFA, ▩ PUFA, ▩ minor PUFA, □ C18:2n-6, □ C18:3n-3, ■ Long-chain n-3 PUFA. Adapted from the following sources: [1][13], [2][2]

Ruminant Meat

There is a marked difference between the dietary fatty acid composition and the absorbed fatty acids in ruminants. As noted above, unsaturated fatty acids are extensively metabolized in the rumen in successive steps. The first step in this process is the lipolysis of the dietary lipids, which releases free fatty acids that are subsequently isomerized and hydrogenated by bacterial enzymes [11]. De novo fatty acid synthesis by bacteria and protozoa in the rumen has also been described [11].

Different nutritional approaches to modifying the fatty acid composition of ruminant meat have been widely studied. The inclusion of forage in the diet of ruminants, for example, enhances the n-3 PUFA content as green forage is a good source of α-linolenic acid [12] with a linear increase in the proportion of C18:3n-3 in pasture-fed animals with increasing grazing periods being observed [12]. These authors also suggested a relationship between the duration of feeding on a diet rich in C18:3n-3 and the concentration of long-chain n-3 PUFA in muscle and adipose tissue.

Differences in the fatty acid composition of meat from animals reared predominantly on grain- and grass-based diets have been widely reported (Fig. 5.2). Thus, cattle grazing on pasture accumulated a four to five fold higher concentration of total n-3 PUFA (approximately 34.3 mg of long-chain PUFA/100 g of muscle) than those fed only on concentrate (approximately 7 mg of long-chain n-3 PUFA/100 g of muscle) [13]. In lambs, it was found that the muscle of lambs fed grass on an extensive system had 3 times more total n-3 PUFA, mainly C18:3n-3, than those reared intensively on concentrates [2]. However, although the long-chain n-3 PUFA content was twice as high in lambs reared on extensive-pasture compared to those reared on an intensive-concentrate system, this content was nevertheless low, at only 40 mg/100 g of muscle for the extensive-pasture animals [2]. Another way for increasing the n-3 PUFA content in ruminant meat is to include supplementary lipids in the animals' diet (Table 5.1). Thus, plant-based sources such as different oils and oilseeds, which mainly

Table 5.1 Literature examples of the use of different sources rich in n-3 PUFA in the diet of ruminants

			Increment with respect to control					
Species	Sources	Levels in diet	C18:3n-3	C20:5n-3 (EPA)	C22:5n-3 (DPA)	C22:6n-3 (DHA)	Total n-3 PUFA	Reference
Beef	Linseed	15.4 %	1.60-fold	1.14-fold	1.07-fold	1.31-fold	1.24-fold	[43][a]
	Linseed + fish oil	7.7 + 2.6 %	1.39-fold	1.39-fold	1.12-fold	2.57-fold	1.39-fold	
	Protected fish oil	6.9 %	1.33-fold[b,c]	2.45-fold[d,c]	INA	2.08-fold[d,e]	0.92-fold[d,c]	[44]
		13.8 %	1.21-fold[b,c]	2.57-fold[d,c]	INA	1.63-fold[d,e]	1.66-fold[d,c]	
		27.5 %	1.74-fold[b,c]	3.77-fold[d,c]	INA	1.60-fold[d,e]	2.67-fold[d,c]	
	Whole linseed	21.3 %	1.95-fold	1.45-fold	1.05-fold[f]	1.09-fold[f]	1.49-fold	[34]
	Fish oil	5.4 %	1.18-fold[f]	2.09-fold	1.20-fold[f]	2.09-fold	1.41-fold	
	Linseed + fish oil	10.6 + 2.7 %	1.36-fold[f]	1.36-fold[f]	1.05-fold[f]	2.23-fold	1.28-fold	
	Linseed	21.3 %	1.94-fold	1.50-fold	1.05-fold[f]	1.08-fold	1.85-fold	[14]
	Fish oil	5.4 %	1.38-fold	2.40-fold	1.26-fold[f]	2.12-fold	2.38-fold	
	Linseed + fish oil	10.6 + 2.7 %	1.44-fold	1.60-fold	1.05-fold[f]	2.20-fold	1.78-fold	
Lamb	Extruded linseed	3 % dry matter (DM)	1.98-fold	0.93-fold[f]	0.90-fold[f]	0.83-fold[f]	1.43-fold	[45][g]
		6 % DM	2.30-fold	1.06-fold[f]	0.90-fold[f]	0.83-fold[f]	1.64-fold	
		9 % DM	3.45-fold	1.30-fold[f]	1.13-fold[f]	1.00-fold[f]	2.32-fold	
	Linseed oil	4.3 %	91.80	24.30	22.80	7.30	142	[46][h]
	Fish oil	4.3 %	57.42	47.92	32.45	22.73	132	
	Protected linseed and soybean (PLS)	11.1 %	140.70	20.96	24.75	5.42	189	
	Fish oil + algae	2.1 + 15.5 %	30.86	85.26	45.26	93.80	254	
	PLS + algae	5.5 + 15.5 %	98.31	44.73	27.26	86.55	257	
	Extruded linseed (EL)	12.5 %	6.36-fold[d]	2.75-fold[d]	1.60-fold[d]	2.10-fold[d]	3.23-fold[i]	[8][j]
	Fish oil	3.3 %	0.47-fold[d,f]	7.06-fold[d]	1.74-fold[d]	6.34-fold[d]	5.09-fold[i]	
	EL + microalgae	10.7 + 4 %	4.74-fold[d]	2.45-fold[d]	1.39-fold[d]	2.44-fold[d]	2.76-fold[i]	
	Fish meal	9 % DM	0.71-fold	1.12-fold[f]	0.98-fold[f]	2.02-fold	0.87-fold[f]	[47][a]
	Fish oil	1.5 % DM	1.03-fold[f]	2.62-fold	1.23-fold	3.29-fold	1.42-fold	
	Protected sunflower meal (PSM)	10.5 % DM	0.68-fold	0.98-fold[f]	0.98-fold[f]	0.87-fold[f]	0.80-fold	
	Fish oil + PSM	1.5 + 9.02 % DM	0.74-fold	2.28-fold	1.13-fold[f]	3.11-fold	1.32-fold	

INA information not available
[a]Data referred to the analysis of phospholipids
[b]Data referred to the analysis of neutral lipid fraction
[c]Data are adjusted to a linear effect
[d]Data referred to the analysis of polar lipid fraction
[e]Data are adjusted to a quadratic effect
[f]Improvement statistically not significant
[g]Data referred to male lambs
[h]Data calculated from the sum of neutral and phospholipid fatty acids, expressed as mg/100 g of muscle. No control diet
[i]Data referred to total intramuscular lipids
[j]Mean values of days 0 and 7

provide α-linolenic acid, and marine products, which are major sources of EPA and DHA, have been added to the diets of ruminants. Thus, supplementation of ruminant diets with sources rich in n-3 PUFA produces an increase of 1.5–5 times more total n-3 PUFA in meat, linseed supplementation increases the C18:3n-3 content, whereas marine sources (algae and/or fish) supplementation leads to higher levels of EPA and DHA.

Supplemented steers with linseed or fish oil contained 82.4 and 77.6 mg of n-3 PUFA/100 g of meat and 18.4 and 27.6 mg long-chain n-3 PUFA/100 g of meat, respectively, whereas the corresponding values for non-supplemented steers were 55.2 and 13.2 mg/100 g of meat for n-3 PUFA and long-chain n-3 PUFA, respectively [14]. These contents represented the 3.7 and 5.5 % of the daily recommended intake (DRI) for long-chain n-3 PUFA for linseed and fish oil supplemented steers, respectively, whereas the values for control steers represented only 2.6 % of the DRI for these fatty acids.

In lambs, it has been reported that meat from animals fed on a non-n-3 enriched diet contained 38 mg of n-3 PUFA and 25 mg of long-chain n-3 PUFA/100 g of muscle [8], which represents only about 5 % of the DRI for long-chain n-3 PUFA (500 mg/day, according to [15]). However, when fish oil was added to the lambs' diet, values of 183 and 170 mg of n-3 PUFA and long-chain n-3 PUFA/100 g of muscle, respectively, were obtained. These latter values represented around 34 % of the DRI for long-chain n-3 PUFA. In the diets supplemented with linseed and microalgae, values of 99 and 116 mg of n-3 PUFA and 48 and 50 mg of long-chain n-3 PUFA/100 g of muscle were obtained, respectively, which represent nearly 10 % of the DRI for long-chain n-3 PUFA [8].

Non-Ruminant Meat

The fatty acid composition of muscle and fat tissues in non-ruminants can be significantly modified by incorporating the appropriate oil source in the feed, as dietary fatty acids are absorbed intact in the small intestine and then incorporated into tissue lipids in these species.

Various nutritional strategies for modifying the fatty acid composition of pork and poultry meat have been studied extensively. Pigs fed with fresh grass and herbs rather than with a more conventional feeding regime have a higher PUFA content in muscle and fat, including total n-3 PUFA and mainly C18:3n-3 [16].

In free-range broilers, it has been reported that the restriction on cereal-based feed intake produced an increment in pasture intake that raised 1.4 times more total n-3 PUFA in breast meat in comparison with meat from broilers that did not have any access to pasture or when feed was restricted [17].

Several studies involving the rearing of pigs and poultry on a diet rich in n-3 PUFA have been carried out, and the results have shown their subsequent incorporation into meat lipids (Table 5.2). Vegetable sources (linseed, flaxseed) and their oils mainly contain C18:3n-3. Although animals are able to synthesize long-chain PUFA from C18:3n-3, the literature shows that diets incorporating marine sources (algae and/or fish) result in significantly higher deposition of EPA and DHA than diets containing vegetable sources. The increased n-3 PUFA content in pork mainly arises upon supplementation with fish oil (Table 5.2). However, supplementation with linseed did not produce as marked an increase in n-3 PUFA content as that achieved with fish oil. Thus, [18] the meat from pigs fed a control diet contained 19.0 mg of n-3 PUFA and 11.5 mg of long-chain PUFA/100 g of muscle, representing only 2.3 % of the DRI for long-chain n-3 PUFA [16]. This content was doubled when crushed linseed was added to the animal diet (41 mg of n-3 PUFA and 22 mg of long-chain PUFA/100 g of muscle). However, when fish oil was supplemented to pigs' diet, 66 and 58 mg of n-3 PUFA and long-chain n-3 PUFA, respectively, were found per 100 g of muscle, thus representing 11.5 % of the DRI for long-chain n-3 PUFA.

In poultry, similarly to cattle, lamb and pig, the highest increment in C18:3n-3 content was achieved through linseed supplementation, whereas the highest increase in long-chain PUFA was obtained upon fish oil supplementation [11] (Table 5.2). However, due to the low amount of adipose tissue

Table 5.2 Literature examples of the use of different sources rich in n-3 PUFA in the diet of non-ruminants

Species	Sources	Levels in diet (%)	Increment with respect control					Reference
			C18:3n-3	C20:5n-3 (EPA)	C220:5n-3 (DPA)	C220:6n-3 (DHA)	Total n-3 PUFA	
Pig	Tuna oil	2	1.22-fold[a]	0–0.27[b]	INA	41.2-fold	2.64-fold	[48]
	Crushed linseed	3	2.25-fold	2.45-fold	1.60-fold	1.29-fold[a]	1.97-fold	[18]
	Fish oil	6	0.85-fold[a]	6.23-fold	1.74-fold	7.29-fold	4.64-fold	
	Extruded linseed	5	3.08-fold	3.08-fold	1.69-fold	2.00-fold	2.17-fold	[42]
	Marine algae	0.25[c]	0.92-fold[a]	1.04-fold[a]	1.20-fold[a]	3.43-fold	1.08-fold[a]	[49]
		0.25[d]	1.09-fold[a]	0.82-fold[a]	0.85-fold[a]	2.14-fold	0.93-fold[a]	
		0.5[d]	1.01-fold[a]	0.89-fold[a]	1.05-fold[a]	3.29-fold	0.97-fold[a]	
Poultry	Tallow	10	0.94-fold[a]	0.57-fold[a]	2.10-fold	2.40-fold	1.16-fold[a]	[50]
	Olive oil	10	0.83-fold[a]	0.57-fold[a]	0.86-fold[a]	1.45-fold[a]	0.85-fold[a]	
	Sunflower oil	10	0.67-fold[a]	0-fold	0-fold	1.64-fold[a]	0.45-fold	
	Linseed oil	10	29.20-fold	17.00-fold	3.55-fold	3.55-fold	24.50-fold	
	Fish oil	1	0.44-fold[a]	5.60-fold	0.66-fold	4.40-fold	3.27-fold	[51]
		2	1.36-fold	8.20-fold	1.30-fold	15.93-fold	4.29-fold	
		3	1.51-fold	10.13-fold	1.90-fold	25.30-fold	5.28-fold	
	Linseed oil	2	5.83-fold	1.10-fold[a]	1.42-fold	1.70-fold	4.52-fold	[52]
		4	8.55-fold	1.95-fold	2.00-fold	2.50-fold	7.19-fold	
	Soybean bean oil	3	3.65-fold	2.00-fold[a]	2.60-fold	2.20-fold[a]	3.25-fold	[19]
	Linseed oil	3	23.70-fold	14.30-fold	8.40-fold	5.20-fold	19.70-fold	
	Fish oil	3	1.65-fold[a]	85.00-fold	20.40-fold	41.20-fold	10.20-fold	
Duck	Dried microalgae (rich in DHA)	0.5	INA	INA	INA	2.84-fold	1.86-fold	[53]

INA information not available
[a]Improvement statistically not significant
[b]Non-detected values in control
[c]Dietary supplementation during finishing period
[d]Dietary supplementation during last half of finishing period

associated with poultry meat, the influence of dietary lipids on muscular fatty acid composition is especially important [11]. Indeed, C18:3n-3 increases between 5 and 30-fold have been achieved upon linseed oil supplementation (Table 5.2), and C22:6n-3 increases of between 4 and 41-fold have been obtained upon fish oil supplementation.

The reported findings [19] show values of 68 mg of n-3 PUFA and 13 mg of long-chain PUFA/100 g of chicken meat when the birds were fed a palm fat diet, and 221 and 30 mg of n-3 PUFA and long-chain PUFA, respectively, per 100 g of chicken meat when the birds were fed a soybean oil enriched diet. Similarly, meat from birds fed with a linseed oil enriched diet contained 1,339 mg and 111 mg of n-3 PUFA and long-chain PUFA, respectively, and that from those fed with a fish oil enriched diet contained 695 and 611 mg of n-3 PUFA and long-chain PUFA, respectively, per 100 g of chicken meat. These long-chain PUFA contents for palm fat, soybean oil, linseed oil and fish oil diets represent 2.6 %, 6 %, 22 % and 122 %, respectively, of the DRI for long-chain n-3 PUFA.

Enhancing the n-3 PUFA Composition of Meat Products by Technological Processes

The link between the consumption of some meat constituents, such as saturated fats, and the risk of several diseases, together with the consumption of convenience meat products, which are considered

Table 5.3 Literature examples of the improvement of the fatty acid profile of meat products by using raw materials from animal production practices

			Improvement with respect to control					
Meat product	Source	Levels in diet	C18:3n-3	C20:5n-3 (EPA)	C22:5n-3 (DPA)	C22:6n-3 (DHA)	Total n-3 PUFA	Reference
Dry-fermented sausage "Salami"	Rice bran	38 % (w)	2.51-fold	1.28-fold[a]	0.86-fold[a]	0.96-fold[a]	2.27-fold	[23]
Dry-cured ham[b]	Linseed oil	3 %	5.59-fold	3.15-fold	1.12-fold[a]	0.78-fold[a]	3.17-fold	[54]
	Linseed + olive oils (1/1, w/w)	3 %	4.17-fold	1.96-fold[a]	1.04-fold[a]	0.81-fold[a]	2.36-fold	
Dry-fermented sausage "Salchichón"	Linseed oil	3 %	7.81-fold	2.00-fold	1.75-fold	0.98-fold[a]	5.38-fold	[24]
	Linseed + olive oils (1/1, w/w)	3 %	6.78-fold	2.00-fold	1.50-fold	0.86-fold[a]	4.68-fold	
Chicken frankfurters	Fish oil	2 %	0.86-fold[a]	3.73-fold[a]	INA	1.61-fold[a]	1.33-fold	[25]
		4 %	0.81-fold[a]	7.54-fold	INA	2.47-fold	1.88-fold	

INA information not available
[a]Improvement statistically not significant
[b]Data from analysis performed on *Biceps femoris*

to be one of the leading sources of fat, in developed countries makes the modification of the fatty acid profile of these products especially interesting. Two main strategies are commonly used to improve the nutritional value of the fatty acid profile of meat products. The first involves the use of raw materials (meat and/or fat) from animals fed on n-3 enriched diets to manufacture different meat products (Table 5.3). Dietary supplementation and the sources commonly used for that purpose have been discussed above. The second strategy involves increasing the n-3 fatty acid content by modifying the lipid fraction during the formulation of meat products, as shown in Table 5.4. Thus, the animal fat is partially or totally replaced with another fat source with a healthier fatty acid composition. Vegetable oils, plants rich in n-3 fatty acids (corn, palm, peanut, walnuts, soybean, high-oleic acid sunflower, linseed, olive, etc.), and marine sources (fish oil and algae) have been used as animal fat substitutes [20]. The type of fat substitute greatly influences the fatty acid profile of the resulting product. Thus, the partial replacement (25 %) of pork backfat with fish oil during the manufacture of Spanish dry-fermented sausage "chorizo" led to a 6,300 and 4,500 % increase in the levels of EPA and DHA, respectively [21], whereas a similar treatment in the same type of product resulted in a 433 and 5,060 % increase in the EPA and DHA contents, respectively, when an algae-derived oil was used [22].

It should be noted that the number of meat products manufactured using partial substitution of animal fat is far greater than those elaborated from raw materials obtained from n-3 supplemented animals. The reason for this is mainly financial. Thus, it must be taken into account that, in supplemented animals, the proportion of fatty acids deposited within the muscle in relation to the ingested amount is generally low. The use of raw materials from supplemented animals is therefore usually limited to the manufacture of meat products made from identifiable pieces of meat, such as dry-cured ham, although some exceptions can be found in the literature [23, 24]. The direct addition of n-3 sources is far more profitable in meat products processed through structural breakdown procedures such as chopping, mincing or any kind of homogenization during their manufacture.

The resulting product may show some differences with respect to the traditional product depending on the fat used, especially as far as the sensory properties are concerned. For example, the use of fish oil might result in the perception of some unpleasant notes, described as "fishy" in the resulting product. Nevertheless, the results in this respect in the literature are diverse. Thus, some authors have found no detrimental effect of fish oil supplementation on the sensory properties of chicken frankfurters

Table 5.4 Literature examples of the improvement of the fatty acid profile of meat products by partial (PR) or total replacement (TR) of animal fat with other fat sources. Reformulation practices

				Improvement with respect to control					
Meat product	Source		Levels	C18:3n-3	C20:5n-3 (EPA)	C22:5n-3 (DPA)	C22:6n-3 (DHA)	Total n-3 PUFA	Reference
Low-salt (0.5 %), low-fat (10 %) cooked beef patties[a]	Olive oil emulsion		5 % (PR)	1.58-fold	1.23-fold[b]	1.00-fold[b]	0[c]	1.42-fold	[9]
			10 % (TR)	1.77-fold	0.89-fold[b]	0.87-fold[b]	0–2.44[c]	1.55-fold	
	Wakame (*Undaria pinnatifida*) seaweed		3.39 %	1.28-fold[b]	2.64-fold	0.86-fold[b]	0[c]	1.25-fold	
Dry-fermented sausage "Chorizo"	Pre-emulsified fish oil		6.25 %	1.1-fold	64.0-fold	INA	46.0-fold	INA	[21]
	Algae oil from *Schizochytrium* sp.		3.75 %	0.90-fold	4.33-fold	1.14-fold	39.20-fold	3.65-fold	[22]
			6.25 %	0.90-fold	5.33-fold	1.09-fold	51.60-fold	4.54-fold	
Frankfurters	Low-fat (6 %)	Walnut	25 %	11.81-fold	INA	INA	0.71-fold[b]	12.82-fold	[42]
	Normal-fat (16 %)			13.34-fold	INA	INA	0.83-fold[b]	12.68-fold	
Bologna-type sausages "mortadella"	Low-fat (10 %)	Fish oil	1, 2, 3, 4 and 6 %	INA	33–220-fold	INA	1.56–5.11-fold	3.13–9.13-fold	[26]
	Normal-fat (30 %)			INA	35–330-fold	INA	4.50–20.00-fold	1.63–5.52-fold	
Low-fat pork liver pâtés	Mixture of oils		7.5 % (PR)	15.89-fold	0–1.22-fold[c]	0–0.24-fold[c]	0–1.00-fold[c]	10.78-fold	[30]
	44.39 % olive oil 37.87 % linseed oil 17.74 % fish oil		15.0 % (TR)	32.35-fold	0–2.69-fold[c]	0–0.46-fold[c]	0–1.80-fold[c]	20.69-fold	

INA information not available
[a]Data from cooked sample
[b]Improvement statistically not significant
[c]Non-detected values in control

[25] whereas other studies have reported some fishy notes in low-fat bologna-type sausages [26] or in dry-fermented sausages formulated with fish oil [21], although the overall acceptability remained unaffected in both cases. Likewise, the use of seaweed as an n-3 source may result in an atypical flavour in the final product depending on both the type of seaweed and dose. The use of walnuts in meat products might also affect the resulting product. Hence, the sensory panel detected a walnut-like flavour after the addition of walnuts to restructured beefsteaks, although they showed better texture than the controls [27]. In any case, the effect of animal fat substitutes on the sensory quality of meat products might be masked or toned down by means of seasonings, when applicable.

The replacement of animal fat can also entail several technological problems. An increase in the PUFA content may lead to "soft" meat and meat products [28]. However, no significant differences with respect to control were observed in salami manufactured from pigs fed on diets composed of maize and rice bran [23] or in chorizo in which 25 % of pork backfat was replaced with fish oil [21]. In fact, the latter study reported better juiciness of the modified sausages. Another possible effect concerns emulsion stability in products such as pâtés. Thus, the substitution of animal fat by another more unsaturated source of fat (with lower melting points than those of SFA) may reduce the emulsion stability of these products [29]. However, this effect was not observed in pork liver pâtés formulated with partial or total replacement of pork backfat by a combination of oils rich in n-3 PUFA [30].

The main problem as far as dry-fermented sausages are concerned is that there appears to be a limit for backfat substitution above which a significant drip fat-loss occurs during ripening, thus leading to a decrease in the MUFA and/or PUFA contents [31].

Problems with n-3 PUFA-Enriched Meat and Meat Products

Modification of the fatty acid profile of meat can entail some negative effects, amongst which lipid oxidation is likely the most relevant. Indeed, lipid oxidation is one of the major degradation processes responsible for losses in food quality. The rate and extent of lipid oxidation in muscle-containing foods is affected by many factors, including exposure to light, oxygen, temperature or the presence of both anti- and pro-oxidants, as well as the lipid content, the degree of unsaturation of the fatty acids and the presence of enzymes in meat [32]. Besides these factors, any processes that cause cell-membrane rupture (such as deboning, mincing or cooking) facilitate the interaction of pro-oxidants with unsaturated fatty acids, thereby resulting in the formation of free radicals and propagation of the oxidative reaction [33].

As mentioned above, the degree of unsaturation of fatty acids greatly influences lipid oxidation with more unsaturated fatty acids being more susceptible to oxidation. Indeed, highly unsaturated fatty acids seem to form free radicals, which then promote the breakdown of other fatty acids [34]. Thus, high levels of fatty acids containing a large number of double bonds (such as DHA which has six) in meat seem to enhance rapid lipid oxidation processes.

Lipid oxidation causes some detrimental effects in terms of the quality of muscle-based foods. Thus, the deterioration of essential fatty acids, together with the loss of nutrients such as the liposoluble vitamins A and E, results in a decrease in the nutritional value of meats and meat products [20]. In addition, various products that may exert different biological effects depending on the conditions of oxidation at the time of eating might be formed. According to some authors [35], lipid oxidation products are cytotoxic and genotoxic.

The oxidation of fats, mainly PUFA, also affects the organoleptic quality of meats and meat products by producing a loss of colour uniformity and unpleasant tastes and odours, amongst other effects. Lipid oxidation is highly correlated with pigment oxidation and hence with the colour of muscle-containing foods [36], as the free radicals formed during lipid oxidation seem to act either directly by promoting the oxidation of meat pigments or indirectly by acting on the reduction systems of the pigments [37].

The oxidative breakdown of lipids during cooking or ripening usually contributes to development of desirable odours in meats and meats products by forming volatile compounds such as aldehydes, ketones and alcohols. However, an increase in the levels of some of these compounds, especially those with low-odour threshold such as aldehydes or ketones, may cause an imbalance in the volatile profile of meats and meat products. An increase in the PUFA content in meats and meat products may result in the development of unpleasant notes in the final product as a consequence of higher levels of lipo-oxidation volatile compounds.

Cholesterol present in meat can also be oxidized by many factors in similar manner to unsaturated fatty acids to produce cholesterol oxides that affect human health. Cholesterol oxidation products (COPs) exhibit specific deleterious effects such as cytotoxicity, mutagenicity, carcinogenicity, atherogenicity, inhibition of sterol biosynthesis and modulation of immune function [38]. The intake of processed foods of animal origin containing high levels of COPs seems to be the greatest source of these products found in the human body [39].

It should be noted that all these adverse effects might, of course, be enhanced under some storage conditions. Hence, storage under appropriate conditions may minimize the harmful effects of lipid oxidation. Moreover, the oxidation of meat and meat products rich in n-3 PUFA can be limited by including of synthetic or natural antioxidants in either the dietary treatment or in the formulation. For example, the antioxidant effects of vitamin E by animal diet [36] or a mixture of butylhydroxy anisole (BHA) and butylhydroxy toluene (BHT) [21] have been successfully proven in n-3 enriched meat and meat products, respectively.

Guidance on Safe Levels

Minor differences in the DRI of n-3 fatty acids amongst countries and/or international health organizations may be found. Previously, health organizations and government agencies used to recommend intakes of 0.6–1 g/day for n-3 PUFA, from which 100–200 mg/day corresponded to long-chain PUFA. These values were then readjusted. For example, Food and Agriculture Organization of the United Nations/World Health Organization (FAO/WHO) or the American Health Association have suggested adequate intakes (AI) for total n-3 PUFA of 1.5–2.5 g/day and specifically for long-chain n-3 PUFA in the range 140–600 mg/day [40].

In Europe, the European Food Safety Authority (EFSA) has proposed DRI separately for C18:3n-3 and for long-chain n-3 PUFA (EPA and DHA), since the conversion of the former into the latter may be affected by numerous factors. Thus, an AI of 250 mg/day for C18:3n-3 was initially proposed [4], although soon after the recommended level was considered to be too low [15]. The AI for long-chain PUFA has been set on 500 mg/day [15]. It should be noted that all these levels are suggested for healthy individuals, but additional levels are recommended in some particular cases such as pregnancy or lactation.

In the United States, there are no official recommendations but some suggestions based on scientific research have been made. Thus, 1.6 and 1.1 g/day of AI for C18:3n-3 were recommended for males and females, respectively, aged between 31 and 50. Concerning Canada, the DRI has been made for total n-3 PUFA which should be between 1.2 and 1.6 g/day. Scientific research suggests that DRI should be reviewed in order to consider health promotion rather than avoid deficiency symptoms.

Conclusions

Fortification of meats and meat products with n-3 fatty acids is an effective way of assuring the DRI of these fatty acids, especially in western countries, in which changing the eating habits of consumers may be difficult. Pasture rearing systems provide meat with higher contents in C18:3n-3. Nevertheless,

dietary supplementation enhances the nutritional fatty acid profile of meat more efficiently than grazing systems. The inclusion of vegetable sources in the animal's diet leads to an increase in the levels of C18:3n-3, whereas the use of marine sources provides higher contents of both EPA and DHA. Meat products can be enriched by either supplementation or by using healthier fat sources through formulation. Some adverse effects might be observed in fortified meats and meat products, although there are promising technological alternatives to reduce them.

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