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Review

Digestion and absorption of polyunsaturated fatty acids

H Carlier, A Bernard, C Caselli

Département de Nutrition, ENS.BANA, Campus Montmuzard, Université de Bourgogne, F21000, Dijon, France

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Summary — Polyunsaturated fatty acids play an important part in the structure and function of cellular membranes and are precursors of lipid mediators which play a key role in cardiovascular and inflammatory diseases. Dietary sources of essential fatty acids are vegetable oils for either linoleic or α-linolenic acids, and sea fish oils for eicosapentaenoic and docosahexaenoic acids. Because of the specificity of the pancreatic lipid hydrolases, triglyceride fatty acid distribution is an essential parameter in the digestibility of fats. The efficiency of the intestinal uptake depends on the hydrolysis and especially on their micellarization. n-3 polyunsaturated fatty acid ethyl ester digestion is recognized to be impaired, but n-3 polyunsaturated fatty acid triglyceride hydrolysis remains a controversial point, and to some authors explains differences observed between vegetable and fish oil absorption. So additional studies are required to investigate this intestinal step. In enterocytes, morphological and biochemical absorption processes involve reesterification of long-chain fatty acids and lipoprotein formation. At this level, specific affinity of I- and L- FABPc (cytosolic fatty acid binding proteins) to polyunsaturated fatty acids requires further investigation. A better understanding of the role of these FABPc might bring to light the esterification step, particularly the integration of polyunsaturated fatty acids into phospholipids. With reference to differences published between fish and vegetable oil absorption, longer-term absorption studies appear essential to some authors. Polyunsaturated fatty acid absorption is thought to be not very dissimilar to that of long-chain mono-unsaturated fatty acid absorption. However, several digestion and absorption specific steps are worth studying with reference to the crucial role of polyunsaturated fatty acids in the organism, and for example adaptation of possible dietary supplements.

polyunsaturated fatty acid / digestion / absorption pathway / enterocyte esterification / intestinal lipoprotein

Résumé — Digestion et absorption intestinales des acides gras polyinsaturés. *Les acides gras polyinsaturés interviennent dans la structure et la fonction des membranes cellulaires et ils participent à la synthèse des médiateurs cellulaires lipidiques dont le rôle est primordial dans la prévention des maladies cardiovasculaires et la défense de l'organisme. Les sources d'acides gras indispensables, linoléique et α-linolénique sont essentiellement d'origine végétale, alors que les acides gras essentiels, eicosapentaénoïque et docosahexaénoïque, proviennent d'animaux marins. La distribution des acides gras sur les 3 fonctions du glycérol des triglycérides, variable selon leur origine, est un paramètre important de la digestibilité des lipides et ultérieurement de leur absorption et de leur biodisponibilité. Si les éthyl esters des acides eicosapentaénoïque et docosahexaénoïque sont reconnus pour être difficilement hydrolysés dans la lumière intestinale, des études complémentaires s'avèrent cependant nécessaires, pour mieux définir les conditions de l'hydrolyse intestinale des tri-*

glycérides d'acides eicosapentaénoïque et docosahexaénoïque, compte tenu des résultats contradictoires obtenus concernant leur absorption. L'absorption intestinale des acides gras à longue chaine nécessite la mise en place de processus biochimiques et morphologiques pour la réestérification de ces acides gras et la formation des lipoprotéines permettant ainsi leur transport dans le milieu intérieur. Il semble que les deux FABPc exprimées au niveau intestinal jouent un rôle déterminant dans les processus d'absorption étant donné leur affinité différente pour les différents types d'acides gras. L'étude de la répartition chyloportale des acides gras polyinsaturés atteste de leur absorption préférentielle par la voie lymphatique, avec selon les conditions expérimentales, des caractéristiques spécifiques par comparaison avec les modalités d'absorption des acides gras à longue chaine monoinsaturés tel l'acide oléique. Ainsi, une intégration particulière dans les phospholipides des lipoprotéines intestinales est mise en évidence. Cette estérification est influencée par la quantité d'acides gras polyinsaturés administrée et le degré d'insaturation des lipides d'accompagnement. La distribution entre les VLDL et les chylomicrons est tributaire du degré d'insaturation des acides gras administrés. Les différences observées par certains auteurs dans les modalités d'absorption des huiles de poissons par comparaison avec celles d'huiles végétales invite à réaliser des expérimentations de longue durée. Toutefois l'absorption des acides gras polyinsaturés n'apparaît pas fondamentalement différente de celle des acides gras à longue chaîne, en particulier mono-insaturés, mais de nombreux points sont à approfondir, étant donné l'importance physiologique des acides gras essentiels. L'intégration des acides gras polyinsaturés, dès l'entérocyte, dans les phospholipides des lipoprotéines intestinales, revêt un intérêt nutritionnel particulier dans l'éventualité d'une supplémentation en acides gras polyinsaturés essentiels motivée par une baisse d'activité des désaturases. Par ailleurs, une meilleure connaissance des mécanismes de digestibilité, permettra de définir la forme d'administration orale adaptée aux conditions optimales de l'hydrolyse intraluminale.

acide gras polyinsaturé / digestion / voie d'absorption / estérification entérocytaire / lipoprotéine intestinale

INTRODUCTION

Polyunsaturated fatty acids play an important role in the structure of cell membranes and in the synthesis of important mediators. Cell membranes consist of a lipid bilayer composed of phospholipids and cholesterol, as well as proteins which are embedded in this lipid bilayer. Differences exist between various cell types in the incorporation and metabolization of fatty acids provided with the diet and between incorporation into specific cellular phospholipids (Galloway *et al,* 1985). Particularly n-3, polyunsaturated fatty acids have a very specific role in the membrane of the nervous system (Bourre *et al,* 1989). The maintenance of appropriate membrane physico-chemical properties is essential for life processes. Thus, modifications in dietary fatty acids change membrane phospholipid fatty acid composition and affect functions mediated by membrane proteins: receptors, transport pathways and enzymes (Goodnight *et al,* 1982; Philbrick *et al,* 1987). Polyunsaturated fatty acids are precursors of lipid mediators which play a key role in cardiovascular and inflammatory diseases. In recent years, evidence has been obtained showing that polyunsaturated fatty acids may be effective in prevention and therapy of cardiovascular diseases (Dyerberg *et al,* 1975, 1978; Bang *et al,* 1976; Kromann and Green, 1980; Goodnight *et al,* 1982). The low incidence of atherosclerosis and chronic inflammatory diseases among the Eskimos of Greenland has been related to their traditional diet rich in long-chain n-3 polyunsaturated fatty acids (Dyerberg *et al,* 1975; Bang *et al,* 1976).

The 2 real main dietary essential fatty acids are linoleic acid, C18:2, n-6 and α-linolenic acid, C18:3, n-3 because they cannot be synthesized by mammalian organisms. But mammalian organisms are generally able to convert linoleic acid (C18:2, n-6) and α–linolenic acid (C18:3, n-3) into n-6 and n-3 polyunsaturated long-chain fatty acids with the participation of desaturases and elongases respectively. On the one hand, arachidonic (C20:4, n-6) and on the other hand, eicosapentaenoic (C20:5, n-3) and docosahexaenoic (C22:6, n-3) acids are synthesized and constitute the more important physiological polyunsaturated essential fatty acids. Arachidonic and eicosapentenoic acid are precursors of eicosanoids: prostaglandins, thromboxanes and leukotrienes, the biosynthesis of which is controlled by cyclooxygenases and endoperoxidases respectively. These eicosanoids have a broad spectrum of biological activity (Goodnight *et al,* 1982; Samuelsson, 1983).

Biological effects and nutritional essentiality of polyunsaturated fatty acids

In response to numerous stimuli, phospholipase A_2 splits off the arachidonic acid (C20:4, n-6) which is metabolized by cyclooxygenase both to thromboxane A_2, a potentiator of platelet aggregation and a vasoconstrictor, and to prostaglandin I_2. Dietary eicosapentaenoic acid (C20:5, n-3), the predominant n-3 fatty acid in the platelet phospholipids of seafood consumers, is released froom phospholipids and can be transformed by cyclooxygenase both into prostaglandin I_3 which is as active as the vasodilatory and antiaggregatory prostaglandin I_2 derived from arachidonic acid, and into a small quantity of thromboxane A_3 which is almost inactive as platelet aggregator and vasoconstrictor (Fischer and Weber, 1983). Moreover, n-3 and n-6 fatty acids compete for the desaturase and elongase enzymes. As a consequence, n-3 fatty acids inhibit the synthesis of arachidonic acid from linoleic acid and a decrease in thromboxane A_2 synthesis may result (Holman, 1964). These biochemical and physiological aspects explain some of the proposed benefits of fish oils (Dyerberg *et al,* 1978; Fischer and Weber, 1983) and the intense interest developed in the biological effects and nutritional essentiality of n-6 and particularly n-3 long chain polyunsaturated fatty acids.

According to the interaction of the n-6 and n-3 polyunsaturated fatty acids, and the importance of their respective metabolism and biological effects, recommendations for dietary fat/fatty acid intake have been proposed assuming 2 600 kcal/day. Total polyunsaturated fatty acids have been evaluated at 6–7% of calories with n-6/n-3 = 4/1, linoleic acid, α-linolenic acid, and eicosapentaenoic acid and docosahexaenoic acid together at 4.8–1.0 and 0.27% of calories respectively. Saturated fatty acids have been evaluated at 6–7% of calories and total mono-unsaturated fatty acids at 12–14% of calories (Lasserre *et al,* 1985; Simopoulos, 1989). There is a limiting step in the transformation of linoleic acid and α-linoelic acid into derivatives. If a loss of desaturase activity prevents endogenous synthesis of arachidonic acid, or of eicosapentaenoic acid, the dietary doses of C18 essential fatty acids must be modulated with certain disease states and with aging (Brenner, 1981) implying a possible supply, for example, in γ-linolenic acid, C18:3, n-6.

Source of polyunsaturated fatty acids and chemical characteristics (table I)

Vegetable oils such as sunflower, soy bean, peanut, and corn oils constitute a

substantial source of linoleic acid. Moreover, some vegetable oils such as soy bean and low erucic acid rapeseed (canbra oil in table I) oils contain both n-3 and n-6, C18 polyunsaturated fatty acids. Evening primrose oil, blackcurrant oil, and borage oil provide significant amounts of C18:3, n-6 γ-linolenic acid and some C18:4, n-3 stearidonic acid, particularly blackcurrant oil (Lawson and Hughes, 1988c). The derived C20 and C22 essential fatty acids may be found in animals. While land animals provide arachidonic acid (C20:4, n-6), sea animals and especially fish oils constitute some important natural sources of n-3 polyunsaturated fatty acids: eicosapentaenoic acid (C20:5, n-3) (EPA) and docosahexaenoic acid (C22:6, n-3) (DHA). However there is a wide variation in fatty acid composition according to the species. For example, herring and mackerel depot fats are rich in gadoleic (C20:1) and erucic (C22:1) acids (17–26% and 11–16% respectively) and poor in eicosapentaenoic (C20:5, n-3) and docosahexaenoic (C22:6, n-3) acids (6–7% and 5–8%). In contrast, menhaden oil contains 16 and 8% respectively of eicosapentaenoic and docosahexaenoic acids.

Triglyceride fatty acid distribution is an important parameter in the digestibility of fats (table II). Unsaturated fatty acids occupy the 2-position of glycerol in fats of most mammals, except for pigs. Gamma-linolenic acid, C18:3, n-6, is found in 2- and 3-positions of evening primrose, blackcurrant, and borage (Lawson and Hughes, 1988c). In the same manner, saturated fatty acids are in 1- and 3-positions and linoleic acid in the 2-position of glycerol in most plant oils. Several differences occur in the marine oil triglyceride chemical structure. For example, eicosapentaenoic acid and docosahexaenoic acid are often found in the 2-position in the glycerol of mackerel, herring, and cod oils, whereas in fat of seal and polar bear eicosapentaenoic and docosahexaenoic acids occupy the 1- and 3-positions (Brockerhoff and Yurkowski, 1966; Brockerhoff *et al,* 1966a, 1968; Bottino *et al,* 1967). In MaxEPA, a purified fish oil preparation, eicosapentaenoic acid occupies both the 2- and 1-, 3-positions and docosahexaenoic acid preferentially the 2-position (Chernenko *et al,* 1989).

Intestinal availability of polyunsaturated fatty acids

The availability of these different polyunsatured fatty acids is determinant for their biological utilization. This availability is conditioned by digestion and intestinal absorption of polyunsaturated fatty acid triglycerides, since triglycerides remain the main source of dietary lipids.

The digestion and absorption of saturated and mono-unsaturated long-chain fatty acid triglycerides have been largely reviewed (Brindley, 1977; Thomson, 1978; Friedman and Nylund, 1980; Thomson and Dietschy, 1981; Tso and Simmonds, 1984; Bernard and Carlier, 1989; Thomson *et al,* 1989). Conversely, long-chain polyunsaturated fatty acid triglyceride digestion and absorption studies began only a few years ago (Nelson and Ackman, 1988) and sometimes gave conflicting results (McDonald *et al,* 1980; Chen *et al,* 1987b; Nilsson *et al,* 1987; Chernenko *et al,* 1989; Pavero *et al,* 1989). The assumption of most investigators was that polyunsaturated fatty acids are digested and absorbed through normal processes similar to those of mono-unsaturated long-chain fatty acids. However, some researchers suggested a possible substantial blood absorption pathway for polyunsaturated fatty acids with reference to their hydrosolubility properties (McDonald *et al,* 1980, 1987).

Table I. Major fatty acid components (mol %) of some vegetable oils, land and marine animal fats.

	Fatty acids											
*Depot fats * or oils*	*C14:0*	*C16:0*	*C16:1 n-7*	*C18:0*	*C18:1 n-9*	*C18:2 n-6*	*C18:3 n-3*	*C20:1 n-9*	*C20:5 n-3*	*C22:1 n-9*	*C22:5 n-3*	*C22:6 n-3*
Peanut [1, 6]		10		3	60	21		1.5				
Sunflower [6]		6		5	23	65	0.1					
Corn [1, 6]		11		2	28	59	0.9					
Soy bean [1]		10		4	24	54	8					
Canbra ** [6]		5		2	57	22	9			2		
Olive [1]		10.5	1	2	76	10	0.9					
Pig * [2]	1	23	3	12	46	13	0.9					
Beef * [2]	5	29	6	18	36	5	1					
Herring * [3]	7	12	9	1	11	2		17	8	26	2	5
Mackerel * [3]	7	17	9	2	18	2		11	6	16	2	8
Cod * [3, 6]	5	12	12	3	22	2		13	9	7	2	13
Menhaden [5, 6]	8	21	13	3	10	1		1	16		1	8
Max EPA [4, 6]	7	17	9	3	15	1	0.7	4	17	2	2	11

** Canbra = low erucic acid rapeseed oil. From: [1] Brockerhoff and Yurkowski, 1966; [2] Brockerhoff *et al,* 1966b; [3] Brockerhoff *et al,* 1968; [4] Brockerhoff *et al,* 1966a; Chernenko *et al,* 1989; [5] Yang *et al,* 1989; and [6] personal analysis.

Table II. Position distribution of fatty acids (mol %) in triglycerides of some vegetable oils, land and marine animal fats.

		Fatty acids											
*Depot fats * or oils*	*Postions*	*C14:0*	*C16:0*	*C16:1 n-7*	*C18:0*	*C18:1 n-9*	*C18:2 n-6*	*C18:3 n-3*	*C20:1 n-9*	*C20:5 n-3*	*C22:1 n-9*	*C22:5 n-3*	*C22:6 n-3*
Peanut [1]	1		14		5	59	18		1				
	2		1		< 1	58	39		< 1				
	3		11		5	57	10		–3				
Corn [1]	1		18		3	27	50	1					
	2		2		< 1	26	70	< 1					
	3		13		3	31	51	1					
Soy bean [1]	1		14		6	23	48	9					
	2		1		< 1	21	70	7					
	3		13		6	28	45	8					
Olive [1]	1		13		3	72	10	< 1					
	2		1			83	14	1					
	3		17		4	74	5	1					
Pig * [2]	1	1	14	2	23	40	12	< 1					
	2	4	58	6	3	18	8	< 1					
	3	1	1	2	11	61	23	< 1					

	1	4	41	6	17	20							
Beef * [2]	2	9	17	6	9	41							
	3	1	22	6	24	37							
	1	6	12	13	1	16	3	25	3	14	1	1
Herring * [3]	2	10	17	10	1	10	3	6	18	5	3	13
	3	14	7	5	1	8	1	20	4	50	1	1
	1	6	15	11	3	20	2	8	4	16	1	2
Mackerel * [3]	2	10	26	6	1	9	1	5	11	5	2	15
	3	3	6	7	2	24	2	14	8	24	1	4
	1	4	13	12	5	31	2	13	3	9	1	2
Cod * [3]	2	6	15	10	1	9	1	7	11	6	3	26
	3	3	6	11	1	24	2	18	12	9	1	9
	1	4	11	15		29		18	3	8	2	3
Seal * [4]	2	11	13	30		30		3	1	1	1	1
	3	1	4	14		26		16	8	7	6	10
	1	2	5	7		27		31	4	7	6	7
Polar bear * [4]	2	5	7	24		45		4	1	1	2	2
	3	1	3	8		30		25	4	2	13	13

[1] Brockerhoff and Yurkowski, 1966; [2] Brockerhoff *et al*, 1966b; [3] Brockerhoff *et al*, 1968; [4] Brockerhoff *et al*, 1966a.

The aim of this review is to expose long-chain fatty acid triglyceride digestion and absorption processes with emphasis on polyunsaturated fatty acid absorption. Several aspects are worth studying: i), Is the gastric and intestinal hydrolysis of polyunsaturated fatty acid triglycerides, or more generally of polyunsaturated fatty acid esters, similar to that of saturated and mono-unsaturated fatty acid triglycerides? ii), Does the presence of double bonds increase mucosal catabolism of these fatty acids during their absorption? iii), What is the chyloportal partition of polyunsaturated fatty acids? iv), If the lymphatic pathway remains the preferential mode of transport of polyunsaturated fatty acids, what is the chemical form of integration of these fatty acids into intestinal lipoproteins: phospholipids or triglycerides? v), What are the intestinal lipoproteins which transfer them: very low density lipoproteins (VLDL) or chylomicrons? These last 3 points are determinant for their biological utilization either as an energy supply or as precursors of lipidic mediators.

Stomach and intestinal steps of dietary lipid hydrolysis

The digestion of lipids is based on hydrolysis of the different lipid constituents of the diet. Triglycerides are the main components (96%) of human dietary lipids corresponding to 80–120 g per day; dietary phospholipids represent 2–4 g per day, enriched with 7–20 g of endogenous phospholipids from the bile and from the shed intestinal cells, and by sterols, especially cholesterol, and by fat-soluble vitamins.

In the stomach, the lingual lipase secreted by lingual glands in murine or gastric lipase in rabbit or in man (DeNigris *et al*, 1988; Moreau *et al*, 1988) starts hydrolysis of triglycerides. This hydrolysis takes place at the low pH of the stomach lumen in the absence of bile salts. In adults, due to the preferential cleaves of the 1- and/or 1- and 3-positions of triglycerides, lingual or gastric lipase mainly produces partial glycerides and free fatty acids. Such amphiphilic molecules help disperse the fat in the stomach and also in the proximal small intestine. This emulsification of lipids enhances further hydrolysis by pancreatic lipid hydrolases in the small intestine, since these enzyme activities depend on the emulsification of dietary fat. Thus, Roy *et al* (1979) demonstrated significantly improved digestion when rats deprived of salivary lipase were fed lipids microdispersed by ultrasonication. Furthermore, Liu *et al* (1987) showed that uptake of docosahexaenoic acid-fish oil was improved by dispersion in preterm infants known to have low levels of pancreatic lipase and bile salts. And Harris and Williams (1989), observed the very rapid and extensive absorption of emulsified fish oil in man.

In the proximal part of the small intestine, *ie* the end of the duodenum and the jejunum, the intestinal lumenal phase of the digestion of lipids by pancreatic lipid hydrolases takes place. In presence of bicarbonate-rich pancreatic juice and bile, triglycerides are included in the emulsion phase previously made in the stomach. Pancreatic bicarbonates increase the intestinal lumenal pH to a value allowing an optimal activity of pancreatic enzymes. Simultaneously under the detergent action of bile salts, the diameter of the emulsion oil droplets is decreased from 50 000 down to 2 000 Å. Intestinal lumen lipid emulsification is thought to be a limiting step for absorption of most long-chain fatty acids (Hofmann, 1976).

Pancreatic cholesterol ester hydrolase activated by bile salts completely hydrolyzes cholesteryl esters into free fatty acids and free cholesterol. Dietary phospholipids, less resistant than biliary phospholipids, are hydrolyzed by activated pancreatic

phospholipase A_2 in the presence of trypsin, calcium ions and bile salts. They yield 1-lysophospholipids and free fatty acids. Above the critical micellar concentration of bile salts, pancreatic colipase, activated by trypsin, strongly binds to the lipase to allow the specific enzymatic action of the pancreatic lipase on triglycerides at the oil–water interface. Thus, triglycerides are cleaved into 2-monoglycerides and free fatty acids (Chapus *et al,* 1975; Borgström, 1977).

Due to the specificity of pancreatic lipid hydrolases, the hydrolysis products included free fatty acids, free cholesterol, 1-lysophospholipids and 2-monoglycerides. Free fatty acids and 2-monoglycerides are the major constituents of the terminal phase of dietary lipid intestinal lumen digestion.

n-3, polyunsaturated fatty acid triglyceride digestion

Brockerhoff *et al* (1966a), Yurkowski and Brockerhoff (1966) and Bottino *et al* (1967) reported that marine oil has an unusual fatty acid distribution pattern and observed that certain long-chain polyunsaturated fatty acids esterified in triglyceride oils of whales were resistant *in vitro* to porcine pancreatic lipolysis. This resistance, which seems to be independent of the position of polyunsaturated fatty acids on the glycerol molecule, was attributed to the introduction of a double bond in the δ-2 through δ-5 position of the fatty acid chain. It is a controversial point *in vitro* in the rat, when lipase and colipase were in sufficient excess over the dietary load, MaxEPA fish oil triglycerides were totally hydrolyzed into monoglycerides and free fatty acids (Chernenko *et al,* 1989). However, Harris and Connor (1980), reported the decrease of postprandial hypertriglyceridemia in humans fed a fatty meal, where the source of fat was salmon oil. Vahouny (1985) and Chen *et al* (1987a) demonstrated in rats that the total fatty acids recovered in lymph after respective salmon oil or menhaden oil and fish oil concentrate feeding were significantly lower than levels found after corn oil feeding. Lawson and Hughes (1988a) showed that in man, eicosapentaenoic and docosahexaenoic acids from fish oil free fatty acids were found to be completely absorbed, but only two-thirds of eicosapentaenoic and docosahexaenoic acids from the fish oil triglycerides were found to be absorbed. They observed that absorption of docosahexaenoic acid but not of eicosapentaenoic acid from fish oil triglycerides was significantly improved, from 68 to 90%, by coingestion with a high-fat meal. This improvement was attributed to an enhanced pancreatic lipase activity. *In vivo,* in the rat, Yang *et al* (1989) observed that hydrolysis of menhaden oil resulted in a preferential retention of a high proportion of the polyunsaturated long chain fatty acids in the sn-2 monoglycerides and in the residual triglycerides; in contrast, the digestion of rapeseed oil led to a preferential release of free long-chain mono-unsaturated fatty acids. They concluded that the differential lumenal release of long-chain and polyunsaturated fatty acids from fish and rapeseed oils is largely due to their characteristic distribution between the primary and secondary positions in the glycerol molecule and to a much lesser extent to a chain length discrimination by pancreatic lipase. Additional studies are required to investigate intestinal modalities of polyunsaturated fatty acid triglyceride hydrolysis, especially C20, and C22, n-3 polyunsaturated fatty acid triglycerides.

n-3, polyunsaturated fatty acid ethyl ester digestion

Despite the fact that ethyl ester forms of fatty acids are not natural components of

the human diet, enriched preparations of eicosapentaenoic and docosahexaenoic acids of fish oils in the form of ethyl esters have been proposed as dietary supplements. Opinions differ about fish oil C20 and C22, n-3 polyunsaturated fatty acid triglyceride hydrolysis compared to vegetable oil, C18:1, n-9 and C18:2, n-6 unsaturated fatty acid triglyceride hydrolysis. The poor intestinal absorption of ethyl esters of C20 and C22, n-3 polyunsaturated fatty acids compared to free fatty acid absorption of eicosapentaenoic and docosahexaenoic acids, is unanimously attributed to their impaired intestinal hydrolysis (Hamazaki *et al,* 1982; El Boustani *et al,* 1987; Lawson and Hughes, 1988a, b). It is now demonstrated that intestinal absorption of eicosapentaenoic and docosahexaenoic acids administered as ethyl esters takes place after their intestinal hydrolysis. Indeed, no eicosapentaenoic or docosahexaenoic ethyl esters were found either in 4-h lymph samples collected from rats receiving ethyl ester, or in blood of man or rats after ethyl ester administration (Hamazaki *et al,* 1982, 1987; Terano *et al,* 1983; Reicks *et al,* 1990). In man, Lawson and Hughes (1988a), noted that the ethyl esters of eicosapentaenoic and docosahexaenoic acids were absorbed in a range of 20% as well as the free fatty acids. Furthermore, they showed (Lawson and Hughes, 1988b), that absorption of both eicosapentaenoic acid and docosahexaenoic acid, from fish oil ethyl esters was increased 3-fold to ≈ 60% by co-ingestion with a high-fat meal, indicating that absorption of polyunsaturated fatty acid ethyl esters is highly dependent on the amount of co-ingested fat. *In vivo* in the rat, Yang *et al* (1989), showed that the methyl and ethyl esters are hydrolyzed ≈ 4 times more slowly than the corresponding triglycerides, which is sufficient to maintain a saturated micellar solution of fatty acids in the intestinal lumen during digestion.

Diffusion across the unstirred water layer of intestinal hydrolysis products (fig 1)

As intestinal hydrolysis progresses, the hydrolysis products can move from the oil–water interface into the aqueous phase of the lumenal content allowing pancreatic lipid hydrolases to pursue their action. Particularly ionized free fatty acids which constitute more than half of the fatty acids at the pH of the jejunum and 2-monoglycerides enter into bile micelles, to form with phospholipids mixed micelles, multimolecular

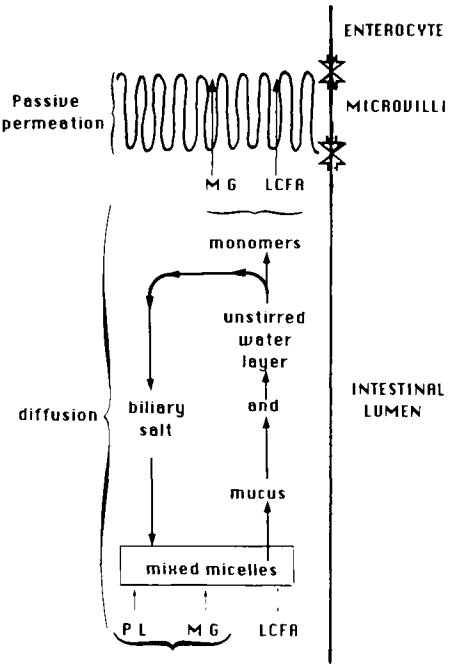


Fig 1. Micellar solubilization and passive permeation of long-chain acids. The mixed micelles diffuse towards the microvillous membrane and act as a solvent to convey apolar molecules up to the surface. The role of bile micelles is to solubilize large amounts of lipids to overcome the resistance of the unstirred water layer (PL = phospholipids; MG = monoglycerides; LCFA = long-chain fatty acids).

aggregates of 30 to 50Å. They constitute the micellar phase of lipid absorption which enables apolar lipid monomers such as saturated and mono-unsaturated long-chain fatty acids to cross the unstirred water layer and mucus lining enterocyte microvillus of the intestinal villus. The transfer of the lipolysis products from the oil phase to the micellar phase appears as a critical step in the absorption of dietary fatty acids. Micellar solubilization increases uptake of lipid digestion products by increasing their aqueous concentration gradients across the unstirred water layer (Wilson *et al,* 1971). The partition of free fatty acids and 2-monoglycerides between the aqueous phase and the micellar phase depends on fatty acid polarity, in particular the chain length and the degree of unsaturation (Hofmann, 1976; Dietschy, 1978). The concentration of long-chain fatty acids in the water phase is low compared to that of short and medium-chain fatty acids and rises with polyunsaturated fatty acids. Mixed micelles, indispensable for the diffusion of apolar lipids across the unstirred water layer and mucus, diffuse less rapidly and release their solubilized products less readily than polar fatty acid monomers, such as short and medium-chain fatty acids and perhaps some polyunsaturated fatty acids.

Uptake of long-chain fatty acids

On the one hand the thickness of the intestinal water layer is the lowest at the distal part of the villus (Thomson and Dietschy, 1981); on the other hand, as the maturity of enterocyte increases as cells migrate from the villus crypts to the villus tips, the enzyme activity involved in the monoglyceride pathway simultaneously increases (Johnston, 1976). Thus, the site of lipid absorption is the proximal part of the small intestine, in an area corresponding to the upper third of the villus. Dissociation of mixed micelles occurs prior to lipid absorption and only monomers stemmed from the lipid lumenal hydrolysis are able to be taken up by enterocytes. This dissociation is permitted by the low pH microclimate which is maintained by the presence of mucus all along the lumenal side of the enterocyte microvillous membrane (Shiau *et al,* 1985; Shiau, 1990).

Uptake of long-chain fatty acids is believed to be an energy-independent process. This passive permeation increases with the lipophilic properties of fatty acids; consequently it increases with the chain length and decreases with the degree of unsaturation. However, at low concentration, polyunsaturated fatty acids such as linoleic and arachidonic acids may be absorbed through a facilitated diffusion mechanism without energy requirement (Chow and Hollander, 1978a,b). The role of a specific long-chain fatty acid binding protein (FABPpm) acting as a membrane receptor is not excluded (Stremmel *et al,* 1985). Fatty acid absorption would represent a dual, concentration-dependent uptake mechanism consisting of a passive diffusional transport process and an active carrier-mediated translocation mechanism which would be predominant at low substrate concentration as demonstrated by recent works with some long-chain fatty acids, among them linoleic and linolenic acids (Stremmel, 1988). At the cytosolic side of the enterocyte microvillous membrane, cytosolic FABPs may be responsible for the removal of long-chain fatty acids from their strong binding to the lumenal enterocyte membrane.

Mucosal phase of fatty acid absorption

The chain length of fatty acids appears to be determinant for absorption processes and particularly the absorption pathway

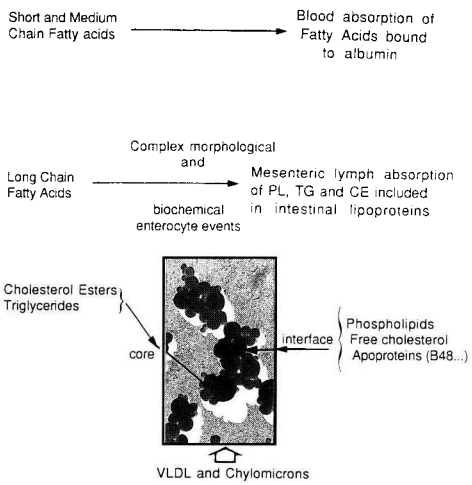


Fig 2. Chyloportal partition of fatty acids according to their chain length. Absorptive processes differ depending on the chain length of the fatty acids. On the one hand, short and medium-chain fatty acids enter the blood stream through the mesenteric portal vein bound to albumin. On the other hand, long-chain fatty acids are absorbed through lymph in esterified form included in very low density lipoproteins (VLDL) and chylomicrons.

(fig 2). Short-chain fatty acids are transported by the portal blood as free fatty acids bound to albumin, while it has been recognized for a long time that long-chain fatty acids are mainly esterified into triglycerides and delivered to the lymph included in lipoproteins: VLDL and chylomicrons (Bloom *et al,* 1951; Blomstrand, 1955; Borgström, 1955; Clement *et al,* 1963; Greenberger *et al,* 1966; Hyun *et al,* 1967; Carlier, 1971; Carlier and Bezard, 1975; Vallot *et al,* 1985; Bugaut and Carlier, 1987). Intestinal absorption of saturated and monounsaturated long-chain fatty acids involves correlated complex biochemical and morphological enterocyte

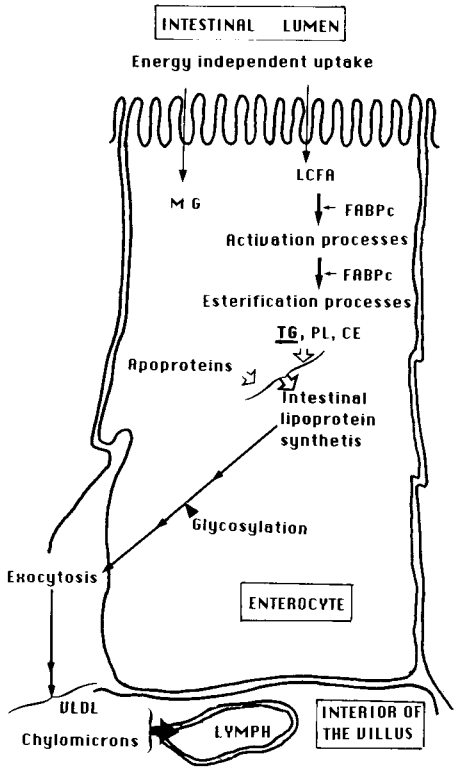


Fig 3. Long-chain fatty acid intestinal absorption steps. Cytosolic fatty acid binding proteins (FABPc), allow the transfer of long-chain fatty acids (LCFA) towards their ultrastructural sites of activation and esterification into phospholipids (PL), cholesteryl esters (CE) and triglycerides (TG). Then association with apoproteins of reesterified long chain fatty acids give very low density lipoproteins (VLDL) and chylomicrons. After glycosylation, the newly synthesized intestinal lipoproteins are secreted in the intercellular spaces and travel towards lacteals.

events (fig 3). Intestinal lipoprotein synthesis by the masking of the hydrophobic groups of apolar long-chain fatty acids permits their transfer in the aqueous media of the lymph and then of the blood vascular compartments.

Cytosolic fatty acid binding proteins (FABPc) bind to the apolar long-chain fatty acids and transfer them through the aqueous compartment of the enterocyte cytosol from the microvillous membrane to the endoplasmic reticulum vesicles. In 1976, Ockner and Manning attributed to the fatty acid binding protein Z isolated from the intestine a higher affinity towards polyunsaturated long-chain fatty acids compared with saturated long-chain fatty acids. In fact, a few years later, 2 cytosolic fatty acid binding proteins were identified in the enterocyte: the I-FABPc (15124 Da), specific of the intestine and the L-FABPc (14273 Da) expressed in the enterocyte but expressed alone in the liver (Bass, 1985). These FABPc seem to have no affinity for short- and medium-chain fatty acids. According to the work of Lowe *et al* (1987) I-FABPc may have an equal affinity for palmitic and arachidonic acids, whereas L-FABPc may have a higher affinity only for the arachidonic acid.This point needs further investigation because of the interest in the fate of polyunsaturated fatty acids in the organism. These cytosolic fatty acid binding proteins transfer long-chain fatty acids towards their site of activation, then the activated long-chain fatty acids towards their sites of esterification in the enterocyte, that is the smooth and rough endoplasmic reticulum. The triglycerides are resynthesized mainly through the 2-monoglyceride pathway (Clark and Hubscher, 1960), rather than the glycerol-3-phosphate pathway (Kern and Borgström, 1965). Simultaneously with the triglyceride synthesis within the smooth endoplasmic reticulum, phospholipid and cholesteryl ester synthesis takes place within the rough endoplasmic reticulum in which an active protein synthesis is also observed, particularly the synthesis of apoprotein B48. The triglyceride synthesis through the 2-monoglyceride pathway requires the presence of 2-monoglycerides as acceptors for activated fatty acids. Lumenal hydrolysis of dietary triglycerides provides enterocytes in exogenous 2-monoglycerides. They are acylated by 2 acyltransferases (a monoacylglycerolacyltransferase and a diacylglycerolacyltransferase) to 1,2-diglycerides and triglycerides respectively. These esterification processes might partly explain the impairment of the absorption of ethyl esters of eicosapentaenoic and docosahexaenoic acids administered alone, that is, without either triglycerides or monoglycerides, since an improvement in their absorption is observed when co-ingested with a high- fat meal in man (Lawson and Hughes, 1988a,b). However, in rats, Reicks *et al* (1990), showed that addition of olive oil to the eicosapentaenoic and docosahexaenoic ethyl ester lipid emulsion did not significantly influence lymph eicosapentaenoic and docosahexaenoic acid recoveries.

The vesiculation observed in the smooth endoplasmic reticulum and afterwards in the rough endoplasmic reticulum saccules of the supranuclear area of the enterocytes takes place simultaneously with the resynthesis of triglycerides, cholesteryl esters and phospholipids (Cardell *et al,* 1967; Carlier, 1971). During their transfer and progression within the rough endoplasmic reticulum, triglycerides, cholesteryl esters and phospholipids are integrated in VLDL and chylomicrons by addition of apoproteins (Bisgaier and Glikman, 1983). These newly synthesized lipoproteins are transferred towards the Golgi complex. Structural changes in the enterocytes during lipid absorption affect the Golgi apparatus, which exhibits a major enlargement of its vesicles filled with the nascent lipoproteins (Cardell *et al,* 1967; Carlier, 1971). This organelle appears to be an essential step for the final struturation of the intestinal lipoprotein particles, particularly their glycosylation. This packaging is necessary in order to allow the

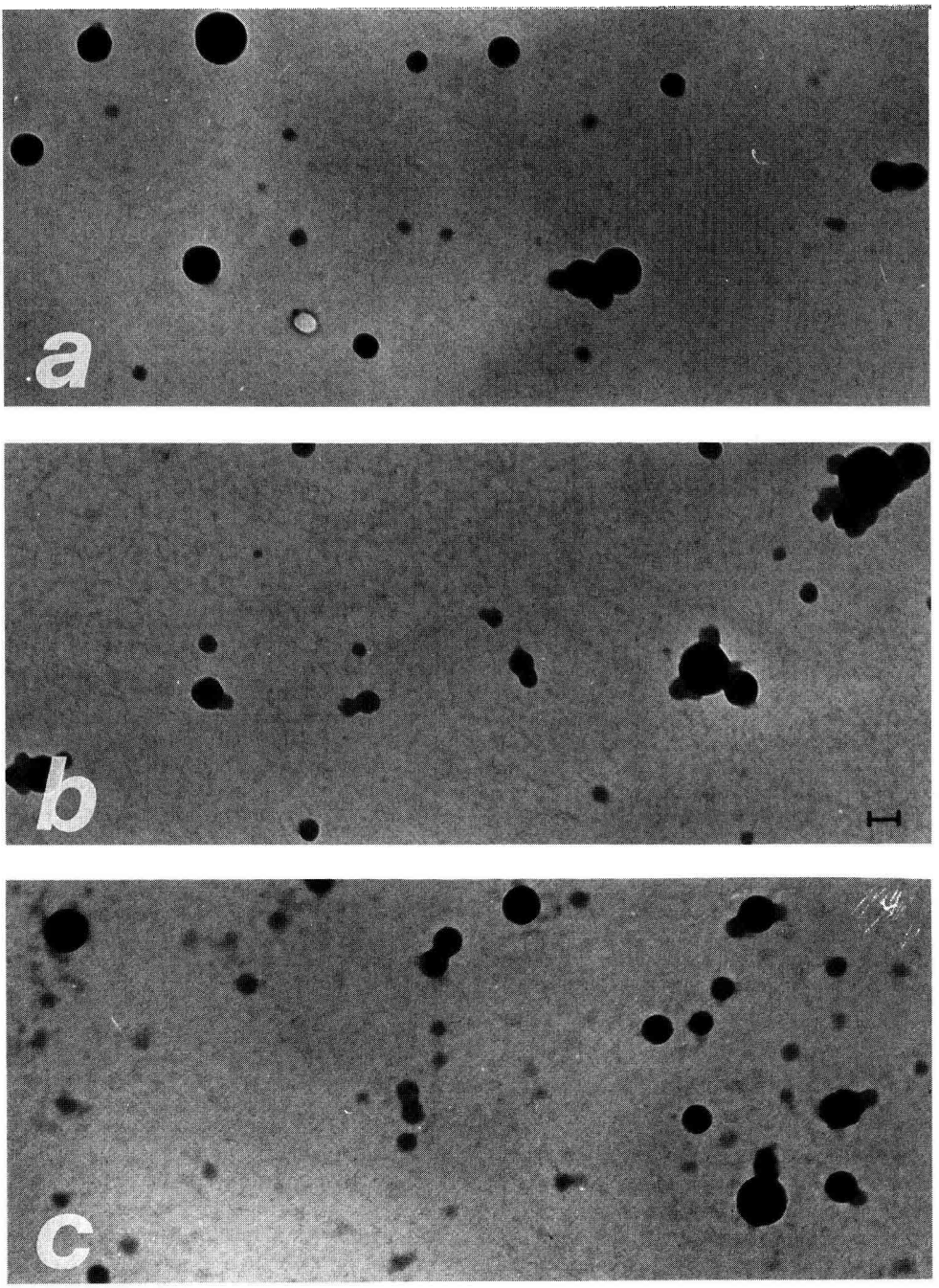
transport of lipoproteins within secretory vesicles towards the lateral plasma membrane and to their exocytosis from the enterocyte into the intercellular spaces, involving the participation of the microtubules for this migration (Sabesin, 1976; Bernard *et al,* 1979a). Intestinal lipoproteins of the intercellular spaces travel towards the basement membrane where opening gaps allow a direct communication with the lamina propria. Their transfer into the lacteals takes place mainly through gaps between adjacent endothelial cells (Cardell *et al,* 1967; Carlier, 1971; Sabesin, 1976; Bernard *et al,* 1979b). Electron microscope micrographs of rat enterocytes after administration of canbra oil (57% oleic acid, 22% linolenic acid, 9% linolenic acid) (Bernard *et al,* 1979a,b) and electron microscope micrographs of rat intestinal lymph particles after administration of either corn oil (30% oleic acid, 52% linoleic acid) or menhaden oil (9% oleic acid, 16% eicosapentaenoic acid, 7% docosahexaenoic acid) or cod liver oil (21% oleic acid, 10% eicosapentaenoic acid, 9% docosahexaenoic acid) (Caselli *et al,* 1979; Rayo *et al,* 1990) (fig 4), seem to indicate that absorption processes of polyunsaturated fatty acids are similar to those of saturated and mono-unsaturated fatty acids.

To clarify such an assumption, we will expose the actual state of knowledge about polyunsaturated fatty acid absorption with reference to both medium-chain fatty acid and to saturated and mono-unsaturated long-chain fatty acid absorption.

Polyunsaturated fatty acid absorption

In rats under vascular perfusion, 24–35% of the infused ^{14}C decanoic acid radioactivity was recovered in the mesenteric portal venous blood during the hour which followed the intraduodenal infusion of either 90 μmol of an equimolar mixture of ^{14}C decanoic acid, oleic acid and monopalmitin or of ^{14}C decanoic acid alone (Vallot *et al,* 1985). In contrast, in the same experimental conditions, only 1.8–2.2% of infused ^{14}C linoleic acid and 2.6 to 3% of infused ^{14}C arachidonic acid were recovered in the mesenteric portal venous blood (Bernard and Carlier, 1991). As suggested by McDonald *et al* (1980), we observed with ^{14}C linoleic acid administered alone, for infused loads equal or inferior to 30 μmol, a significant increase in blood absorption as linoleic acid doses decreased, but all the values remained lower than 5% of the infused radioactivity (Bernard *et al,* 1991) (fig 5). In rats with fistulated main mesenteric lymphatic duct using the same labelled lipid emulsions (fig 6), *ie* either 90 μmol of ^{14}C fatty acid alone or 90 μmol of an equimolar mixture of ^{14}C fatty acid, oleic acid and monopalmitin, only 0.36–3% of the infused ^{14}C decanoic acid was recovered in the lymph instead of 32–48% of the infused ^{14}C linoleic acid (Vallot *et al,* 1985; Bernard *et al,* 1991) and of 37–43% of the infused ^{14}C arachidonic acid during the 6 h following duodenal lipid infusion (Pavero *et al,* 1989). In the lymph, we observed a slightly higher recovery of ^{14}C linoleic acid radioactivity rather than that

Fig 4. Electron micrographs of particles of rat mesenteric lymph collected at the peak of absorption, after ingestion of either 0.5 ml or corn oil (a) or menhaden oil (b) or cod liver oil (c) (Rayo *et al,* 1990). Ingestion of corn oil, menhaden oil or cod oil is followed by an increase of the intestinal formation of lipoproteins (see fig 8). Relative proportion of chylomicrons appears to be of the same range whatever the oil ingested, indicating a similar lymph transport pathway both for fish oil triglycerides and corn oil triglycerides. Scale bar = 0.1 μm.


a
b
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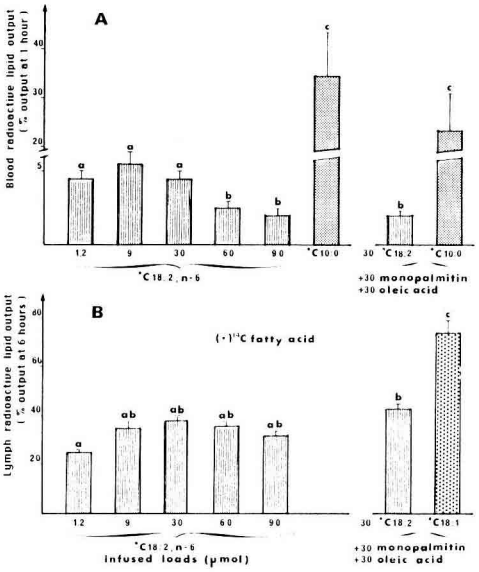

Fig 5. Blood (A) and mesenteric lymph (B) absorption of intraduodenally infused ^{14}C linoleic or ^{14}C decanoic acids either for 1 h in rats under vascular perfusion (A) or for 6 h in main mesenteric lymphatic duct fistulated rats (B) (Vallot *et al*, 1985; Bernard *et al*, 1991). Values are means (SEM) for 4 rats per group. Either ^{14}C linoleic acid acid was administered alone at different loads from 1.2 to 90 μmol or either 30 μmol of ^{14}C linoleic acid or ^{14}C decanoic acid were infused in presence of 30 μmol of oleic acid and 30 μmol of monopalmitin. Bars assigned with different letters are significantly different ($P < 0.001$). In comparison to decanoic acid and to oleic acid intestinal absorption, a preferential lymph absorption pathway occurs for linoleic acid, whatever the dose administered.

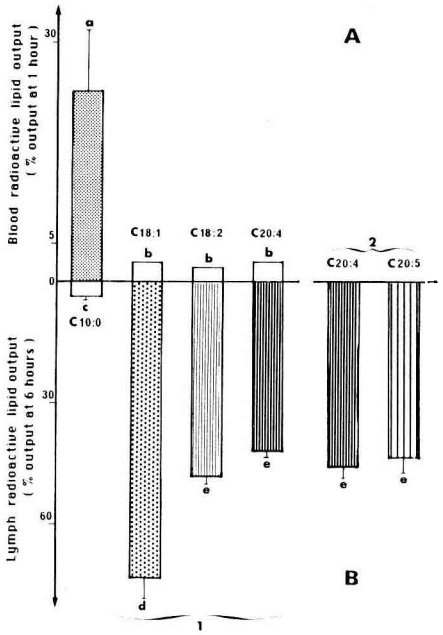

Fig 6. Blood (A) and lymphatic (B) intestinal absorption of intraduodenally infused ^{14}C fatty acids either for 1 h on rats under vascular perfusion (A) or for 6 h on main mesenteric lymph duct fistulated rats (B) (Vallot *et al*, 1985; Pavero *et al*, 1989; Bernard and Carlier, 1991; Bernard *et al*, 1991). Either 30 μmol of ^{14}C fatty acid were administered in presence of 30 μmol of oleic acid and 30 μmol of monopalmitin (1), or 30 μmol of eicosapentaenoic acid or docosahexaenoic acid were administered in presence of 30 μmol of linoleic acid and 30 μmol of monoolein (2). Values are means (SEM) for 4 to 5 rats per group. Bars assigned with different letters are significantly different ($P < 0.01$). Whatever the polyunsaturated fatty acid studied, results reveal their preferential lymph absorption pathway.

of ^{14}C arachidonic acid, as observed by Nilsson *et al* (1987).

Twenty-four h after gastric administration of 300 μmol of either oleic acid or arachidonic acid or eicosapentaenoic acid, in rats with fistulated left thoracic lymphatic channel Chen *et al* (1985) demonstrated that the overall appearance of arachidonic and eicosapentaenoic acids in the lymph was quantitatively equivalent to that of oleate, although there were apparent differences in the rates of lymphatic absorption of these fatty acids. In these experiments the lymph recovery was 76.2–

85.6% of the quantify infused. Studies carried out on rats with fistulated main mesenteric lymphatic duct, with 90 µmol of equimolar mixtures of oleic acid, monopalmitin and either ^{14}C arachidonic acids or ^{14}C eicosapentaenoic acid, gave similar recoveries of labelled lipids in lymph (45.1 and 43.7% respectively of the radioactivity infused intraduodenally 6 h before) (fig 6). The same similitude appeared in the results when the infusates were composed of 5 µmol of either ^{14}C arachidonic acid diluted with 25 µmol of linoleic acid or ^{14}C eicosapentaenoic acid diluted with 25 µmol of arachidonic acid in the presence of 30 µmol of oleic acid and 30 µmol of monopalmitin (Pavero *et al,* 1989). Whatever the experimental conditions and the dose administered, unesterified polyunsaturated fatty acids are efficiently absorbed through the lymphatic pathway, even if some differences occur in the absorption profiles, as noted by Chen *et al* (1985). Thus in our data, compared to oleic acid absorption, the peak of absorption was delayed by 30 min for arachidonic acid and that of linoleic acid by 60 min, and the lymph recovery was significantly lower (Pavero *et al,* 1989). Such differences might be explained by the integration of these polyunsaturated fatty acids into pools of mucosal phospholipids before their effective integration in VLDL and chylomicrons.

Polyunsaturated fatty acid integration in lymph phospholipids

Most investigators noted a significant increase of labelled lymph phospholipids after labelled polyunsaturated fatty acid administration by comparison for example with a mono-unsaturated fatty acid such as oleic acid. Despite the fact that oleic, arachidonic and eicosapentaenoic acids were largely recovered in lymphatic triglycerides, particularly when administered alone (Chen *et al,* 1985; Nilsson *et al,* 1987; Pavero *et al,* 1989), Chen *et al* (1985) showed a greater incorporation of arachidonic and eicosapentaenoic acids into lymphatic phospholipids (respectively 4.5 and 4.3% of the radioactive lipids) than oleic acid (2.1%). Can such an esterification pathway be explained, as in the liver, by a preference of 1-lysophosphatidylacylCoA transferase for unsaturated fatty acids (Nisson *et al,* 1987)? *In vitro,* with mouse jejunal explants incubated for 15 min at 37 °C in an oxygenated culture medium enriched with lipids (1.2 mM: equimolar in monopalmitin, oleic acid and either ^{14}C oleic or ^{14}C linoleic acid) ^{14}C oleic acid was integrated mainly into triglycerides: 82.5 nmol per mg of explant proteins *versus* 37 nmol/mg of explant proteins into phospholipids, whereas ^{14}C linoleic acid was mainly integrated into phospholipids: 74.5 nmol per mg of explant proteins *versus* 49 nmol/mg of explant proteins into triglycerides (Bernard and Carlier, 1991; in press). This integration in lymph phospholipids is particularly enhanced in our experimental conditions *in vivo* in the rat, when 30 µmol of monopalmitin of ^{14}C fatty acids were administered with 30 µmol of oleic acid and 30 µmol of monopalmitin. Thus, at the maximum of the radioactive lymph recovery (from the 30th to the 120th min after the intraduodenal infusion), 1.1 to 3.1% of the lymph lipid radioactivity was recovered in phospholipids, after ^{14}C oleic acid infusion, 7.3 to 19% of the lymph lipid radioactivity was found on phospholipids after ^{14}C arachidonic acid administration (Pavero *et al,* 1989) (fig 7).

The integration of polyunsaturated fatty acids into lipoprotein phospholipids appears to be influenced by the quantity of lipids infused and the degree of unsaturation of the other lipids of the lipid emulsions. Thus, in the rat, Nilsson *et al* (1987), observed that the incorporation of both linoleic and arachidonic acids into different lipid classes varied with the proportion of unsaturated fat in the meal: the proportion

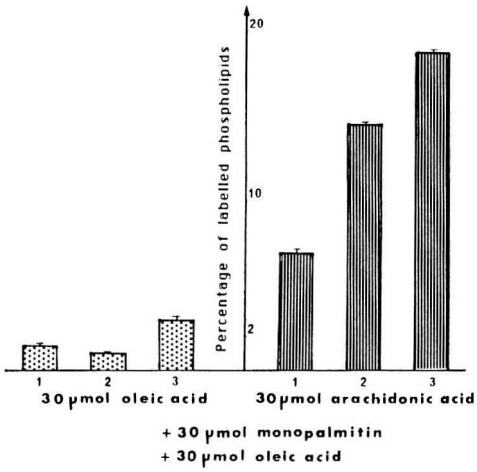

Fig 7. ^{14}C oleic and ^{14}C arachidonic acid integration into lymph phospholipids (Pavero *et al*, 1989). Results are percentages *versus* the total radioactive lymph lipids of the radioactivity recovered into lymph phospholipids. Values are means (SEM) for the second (1), the third (2) and the fourth (3) half h following the intraduodenal infusion of 30 µmol of either ^{14}C oleic acid or ^{14}C arachidonic acid in the presence of 30 µmol of oleic acid and 30 µmol of monoplamitin. A significant increase in labelled lymph phospholipids appears after labelled arachidonic acid administration compared with labelled oleic acid administration.

of the 2 polyunsaturated fatty acids in phospholipids was higher when fed in highly saturated fat *ie* cream instead of intralipid. Some of our results corroborated this observation: at the maximum of the radioactive lymph recovery, the integration of ^{14}C arachidonic acid into phospholipids was significantly decreased when this fatty acid was administered in the presence of linoleic acid and mono-olein rather than when ^{14}C arachidonic acid was administered with oleic acid and monopalmitin (2.8–6.5% and 7.3–19% respectively of the lymph lipid radioactivity was recovered in the phospholipids) (Pavero *et al*, 1989).

Marine oil absorption

The above discussion concerns absorption studies of unesterified polyunsaturated fatty acids administered alone or with other lipids. What happens with vegetable oils rich in C18 polyunsaturated fatty acids, preferentially of the n-6 family, and with marine oils rich in C20 and C22 polyunsaturated fatty acids of the n-3 family? Harris and Connor (1980) observed that the ingestion of salmon oil did not give a typical fat tolerance curve in human subjects, compared to those given a control meal containing animal and vegetable fats. These results were in agreement with Vahouny's data in the rat (Vahouny, 1985). Then Chen *et al* (1987b) found that corn oil was better absorbed than menhaden oil or fish oil concentrate: 24 h after duodenal infusion of 170 mg of each emulsified oil respectively 219.7–164.4 and 152.2 mg of fatty acids were recovered in rat thoracic lymph. In contrast, Chernenko *et al* (1989) did not observe any significant difference in the lymphatic absorption of 0.5 ml of MaxEPA or olive oil given intraduodenally in a bolus to Sprague–Dawley rats, over 6 and 24 h, over which times ≈ 40 and 70% of the administered dose was recovered in lymph triglycerides. In the same manner Rayo *et al* (in press) did not observe any significant difference in lymph triglyceride output of rats fed with 0.5 ml of either corn oil, or menhaden oil or cod liver oil, but the profiles of lymph absorption were different (fig 8). Menhaden oil was more rapidly absorbed than cod liver oil; these differences might be explained by the presence of gadoleic and erucic acids in cod liver oil. Undoubtedly, differences between authors are to be attributed to differences in experimental conditions; the role of lingual or gastric lipase must be also taken into account, as well as the fractionation delivery of the chyme from the stomach. All these

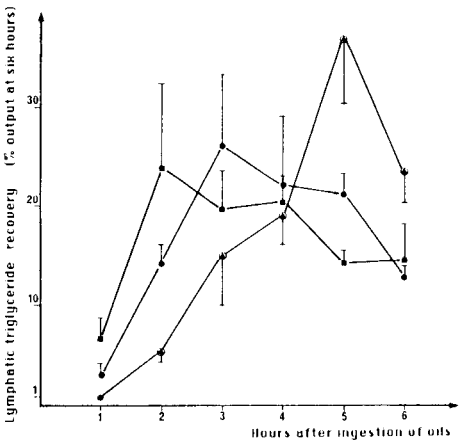

Fig 8. Lymphatic triglyceride recovery as a percentage of total lymphatic triglyceride output over 6 h (Rayo *et al,* 1990). Profiles of lymphatic absorption of corn oil (●), menhaden oil (■) and cod liver oil (▲) during the 6 h following the ingestion of 0.5 ml of one of these oils in rats. Menhaden oil and corn oil are absorbed more rapidly than cod liver oil.

studies dealt with short-term absorption; longer-term animal studies appeared essential for some authors to conclude about the bioavailability of polyunsaturated fatty acids (Hamazaki *et al,* 1987).

Polyunsaturated fatty acids and lymph lipoprotein formation

Few works have dealt with the morphologic aspect of lymph lipoprotein particles. Chen *et al* (1985) subjected lymph samples to ultra-centrifugal separation and noted that the distribution of labelled oleic, arachidonic and eicosapentaenoic acids among major lymph lipoproteins were similar. The same researchers (1987a), demonstrated that the overall clearances of the EPA-enriched and oleate-enriched chylomicrons from the circulation are essentially the same. In agreement with biochemical results of Chen *et al* (1985), electron micrographs of lymph lipoprotein particles reveal no significant differences after administration of corn, or menhaden or cod liver oil (fig 4). In fact, percentage and size of chylomicrons appear correlated with the degree of unsaturation of the lipids administered. For example, when arachidonic acid was infused with oleic acid and monopalmitin (90 μmol: 30/30/30 mol/mol/mol) at peak absorption, 6.3% of the lymph lipoprotein particles were chylomicrons, while when arachidonic acid was infused with linoleic acid and mono-olein (90 μmol: 30/30/30 mol/mol/mol) at peak absorption, 11.2 were chylomicrons. When arachidonic acid (5 μmol) was diluted with linoleic acid (25 μmol) and infused with oleic acid and monopalmitin (30/30 mol/mol) at peak absorption, 7.3% of the lymph particles were chylomicrons, while, as in previous experiments, when 5 μmol of arachidonic acid and 25 μmol of linoleic acid were infused with linoleic acid and mono-olein (30/30 mol/mol), at peak absorption, 11.3% of the lymph particles were chylomicrons. In the 2 cases, the substitution of oleic acid and monopalmitin by linoleic acid and mono-olein was followed by an increase in both the proportion of chylomicrons and in their size (fig 9). These observations on the influence of the unsaturation degree of fatty acids corroborate previous results obtained with vegetable oils (Caselli *et al,* 1979).

Enterocyte metabolism of polyunsaturated long-chain fatty acids

Chernenko *et al* (1989) indicated that fish oil is absorbed from the rat intestine without any major alteration in the acyl chain of the triglycerides. The results of the diges-

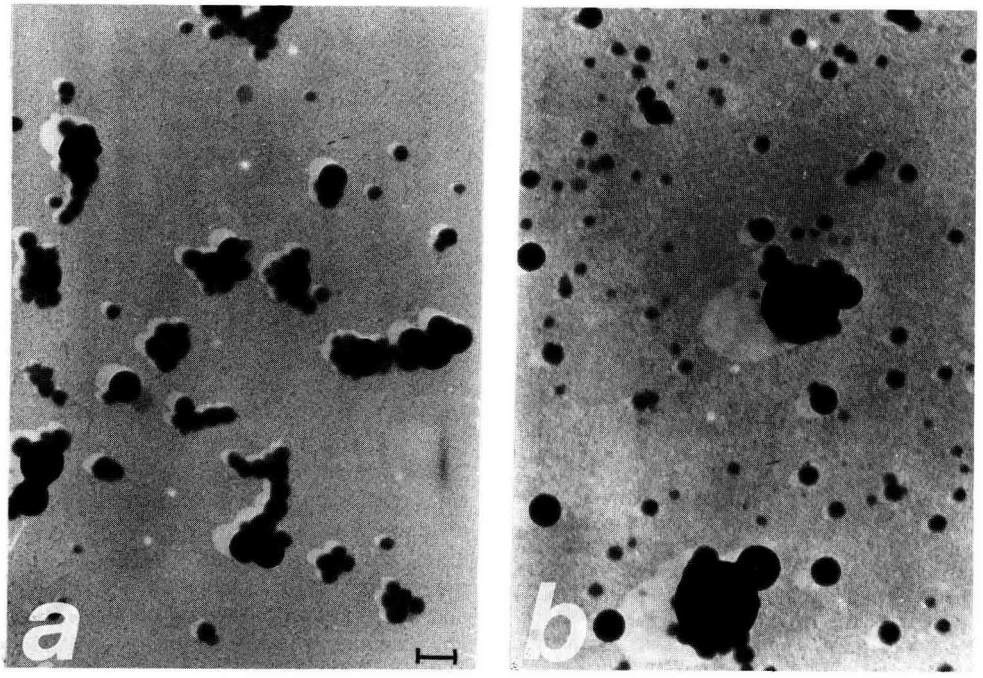

Fig 9. Electron micrographs of rat mesenteric lymph lipoprotein particles. Rat mesenteric lymph lipoprotein particles visualized at the peak of absorption of ^{14}C lipid absorption after intraduodenal infusion of 5 μmol of arachidonic acid either in presence of 25 μmol of linoleic acid, 30 μmol of oleic acid and 30 μmol of monopalmitin (a), or in presence of 55 μmol of linoleic acid and 30 μmol of mono-olein (b) (unpublished results). Large-size chylomicrons appear in the lymph when the degree of unsaturation of lipids administered increases. Scale bar = 0.1 μm.

tion of the lymph samples with lipase indicated that the positional distribution of characteristic fish oil fatty acids is similar in both the MaxEPA and the intestinal lymph. Information concerning eventual oxidation and remodeling of fatty acids in the enterocyte during their absorption is limited. Some authors have shown that endogenous plasma fatty acids were more oxidized in the enterocyte than exogenous lumenal fatty acids (Gangl and Ockner, 1975). Although the intestine was an actively metabolizing tissue, it oxidized very little of the exogenous fatty acids. the study of catabolic products recovered in mesenteric portal venous blood of rats (Bernard and Carlier, 1991) shows that the mucosal catabolism of polyunsaturated fatty acids, even higher than the mucosal catabolism of saturated and monounsaturated long-chain fatty acids, remains low compared to their absorption. In fact, this oxidation appeared in relation with the chain length, decanoic acid is significantly more oxidized than long-chain fatty acids, and with the degree of unsaturation, pal-

mitic acid and erucic acids were significantly less oxidized than oleic, linoleic and particularly arachidonic acids (Greenberger *et al,* 1965; Vallot *et al,* 1985; Bernard and Carlier, 1991).

Christiansen *et al* (1986) revealed that the microsomal fraction from rat small intestine contains a fatty acid elongation activity; they showed that the activity towards saturated and mono-unsaturated fatty acids may seem similar in liver and small intestine, and that highly polyunsaturated fatty acids are markedly poorer substrates for the intestinal system. Furthermore, Garg *et al* (1988) showed the presence of some desaturase enzymes in the rat enterocyte. Particularly, they demonstrated that rat small intestine possesses desaturase activity to convert palmitic acid into palmitoleic acid and linoleic acid into linolenic acid. They suggested that a significant amount of arachidonic acid may originate from *de novo* synthesis within the enterocyte *via* desaturation and chain elongation of linoleic acid. In agreement with this hypothesis, labelled arachidonic acid appeared in the intestinal lymph of rats during labelled linoleic absorption, the best transformation efficiency occurring at peak absorption (Bernard *et al,* 1991).

A chain shortening, more or less associated to chain lengthening, of erucic acid, was described when erucic acid was administered in free form either with triglycerides (Thomassen *et al,* 1985), or with monoglycerides (Pavero *et al,* 1990).

It may be concluded that the enterocyte remains essentially an absorptive cell.

CONCLUSION

In conclusion, polyunsaturated long-chain fatty acid absorption does not appear to be very dissimilar to that of long-chain fatty acid absorption, particularly monounsaturated long-chain fatty acid absorption. Indeed, it has been accepted that saturated long-chain fatty acids are poorly absorbed (Bloom *et al,* 1951; Bernard *et al,* 1987). However, polyunsaturated long-chain fatty acid absorption processes require further investigation to elucidate some particularities of the lumenal phase with reference to the polarity of polyunsaturated fatty acids and some specific absorption processes of the mucosal phase, in particular esterification modalities. Better information on the emulsification, hydrolysis and micellarization of polyunsaturated long-chain fatty acids is essential for the adaptation of possible dietary supplements. Furthermore, a better understanding of the role of the enterocyte fatty acid binding proteins towards polyunsaturated fatty acids might bring to light the esterification step crucial for the fate of the polyunsaturated fatty acids in the organism. Indeed, the energetic or regulator further utilization of these essential fatty acids might depend on their integration into phospholipids or triglycerides as soon as they are absorbed.

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