Preferential incorporation of *sn*-2 lysoPC DHA over unesterified DHA in the young rat brain

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Thiès, F., C. Pillon, P. Molière, M. Lagarde, and J. Lecerf. Preferential incorporation of *sn*-2 lysoPC DHA over unesterified DHA in the young rat brain. *Am. J. Physiol.* 267 (*Regulatory Integrative Comp. Physiol.* 36): R1273–R1279, 1994.—The uptake and metabolism of [^{3}H]docosahexaenoic acid (DHA) esterified at the *sn*-2 position of lysophosphatidylcholine (lysoPC DHA) and in the unesterified form, both bound to albumin, was studied in 20-day-old rats. LysoPC DHA was preferentially recovered in the brain (4–5% of the injected radioactivity) over the unesterified form of DHA (0.3–0.4%). Conversely, the lysoPC form was taken up less than or at the same extent as the unesterified form by the liver, heart, and kidney. In the brain, DHA was mainly recovered in phosphatidylethanolamine whether the esterified or the unesterified form was used, although DHA from lysoPC was esterified at the same extent in phosphatidylcholine and phosphatidylethanolamine after 2.5 min. The uptake of labeled palmitic, oleic, linoleic, and arachidonic acids, esterified or not in lysophosphatidylcholine, was also studied in brain, liver, heart, and kidney. Only the brain preferentially incorporated unsaturated (but not saturated) lysoPC, with the uptake increasing with the degree of unsaturation of the fatty acid moiety. These results strongly suggest that the young rat brain specifically utilizes albumin-lysoPC-containing polyunsaturated fatty acids.

sn-2-docosahexaenoyl lysophosphatidylcholine; docosahexaenoate; albumin; fatty acids; liver; kidney; heart; acylation; lipid metabolism

THE BRAIN CONTAINS high amounts of docosahexaenoic acid (DHA), mainly in the gray matter and synaptosomes (8, 9, 23). This n–3 essential fatty acid plays a structural and metabolic role in the retina (13, 20, 29) and seems also to be involved in the development of learning activity and exploratory behavior in the young animal (7, 30, 31, 33).

Although the brain possesses enzymes responsible for chain elongation/desaturation of linolenic acid, the essential fatty acid precursor of DHA, an exogenous supply of DHA seems nevertheless necessary, at least during the developmental period (6, 20, 23, 30). In rat, the linoleic-to-linolenic acid ratio of the maternal diet is critical for the DHA accretion in brain of the young suckling rat (32). In mice, it has been shown that DHA is formed from linolenic acid in the pup liver and secreted into the blood for redistribution to other organs (3). Therefore it is likely that DHA is mainly supplied to the brain and other organs by blood during the period of brain development in mammals, even in species such as rodents whose milk contains negligible amounts of this fatty acid.

Brain utilization of unesterified DHA bound to albumin used as a delivery form has been investigated (1, 11, 21, 24), and orally administered DHA seems to be particularly redistributed to the retina and brain via albumin (17). However, albumin also carries lysophosphatidylcholine (lysoPC) (26), the polyunsaturated species of which are secreted by the liver (14, 22). On the other hand, we previously showed that unsaturated fatty acids are preferentially taken up by the developing brain in the lysoPC form over the corresponding unesterified forms (28). This was observed for oleic acid and for n–6 fatty acids, such as linoleic and arachidonic acids. However, opposite and reciprocal effects of n–6 and n–3 fatty acids on the brain fatty acid composition are well known (6, 20). The present study was therefore designed to test the uptake of lysoPC-containing DHA vs. the uptake of its unesterified form by the developing brain. We have also investigated whether other organs would be able to preferentially take up DHA or other unsaturated fatty acids esterified at the *sn*-2 position of lysoPC bound to albumin. The present results show that the developing rat brain preferentially took up DHA from lysoPC DHA compared with unesterified DHA. In contrast, other organs, such as the liver, kidney, and heart, did not exhibit such a preference. Similar results could be observed for other unsaturated fatty acids.

MATERIALS AND METHODS

Chemicals. [4,5-^{3}H]DHA (23 Ci/mmol) and *sn*-1-palmitoyl-2-[1-^{14}C]arachidonyl phosphatidylcholine (50 mCi/mmol) were purchased from NEN (Du Pont, France). [1-^{14}C]palmitic, [1-^{14}C]oleic, and [1-^{14}C]linoleic acids (50–56 mCi/mmol) were purchased from CEA (Saclay, France). [^{3}H]methyl iodide (10 Ci/mmol) was purchased from the Radiochemical Centre (Amersham, UK). Palmitic, oleic, linoleic, and arachidonic acids; DHA; 1-acyl-lysophosphatidylcholine from egg yolk; *sn*-1,2-dipalmitoyl, dioleoyl, and dilinoleoyl phosphatidylcholine; 1,1′-carbonyldiimidazole; dimethyl sulfoxide; tetrahydrofuran; and the lipase from *Rhizopus arrhizus* were purchased from Sigma Chemical (St. Louis, MO). Sodium or lithium bis(trimethylsilyl)amide was purchased from Aldrich (Milwaukee, WI).

LysoPC preparation. Phosphatidylcholine species (PC) with specific fatty acid esterified at the *sn*-2 position were synthesized by acylation of *sn*-1-acyl,2-lysoPC. The acyl moiety at the *sn*-1 position was then removed by enzymatic hydrolysis.

To minimize oxidative degradations, the acylation of *sn*-1-lysoPC by [^{3}H]DHA was performed as follows. DHA imidazolide was formed in a microvial by addition of 12.25 μmol of carbonyldiimidazole to 150 μl of a hexane solution containing 10 μmol of labeled DHA and 25 nmol of butylated hydroxytoluene (BHT) as an antioxidant. The reaction was carried out for 40 min at room temperature under argon atmosphere. Then 10 μmol of dry egg yolk *sn*-1-lysoPC were added to the reaction vial with 10 μmol of sodium or lithium bis(trimethylsilyl)-amide for 10 min. The reaction mixture was then neutralized with 1 N HCl, lipids were extracted with a 1:1 (vol/vol)

mixture of chloroform-methanol, and the labeled PC was purified as previously described (28).

sn-1,2-Diacyl(palmitic, oleic, linoleic, or arachidonic acid) phosphatidyl[*methyl*-^{3}H]cholines were prepared by demethylation of corresponding PC and remethylation by [^{3}H]methyl iodide, and PC with labeled palmitic, oleic, linoleic, or arachidonic acid esterified at the *sn*-2 position were prepared by acylation of *sn*-1-lysoPC, as previously described (28). *sn*-2-Acyl,1-lysoPC were obtained by selective hydrolysis of the ester bound at the *sn*-1 position of appropriately labeled PC by the lipase from *R. arrhizus*. Each resulting lysoPC was extracted, purified, and checked for isomeric purity as previously reported (28).

Physiological experiments and lipid analysis. Physiological experiments and lipid analysis of different tissues were conducted as previously described (28) and are summarized briefly. Lyophilized delipidated bovine serum albumin was coated with labeled lysoPC or with labeled free fatty acids (FFA), solubilized in 0.15 M NaCl solution, and filtered. Anesthetized 20-day-old Sprague-Dawley male rats (Iffa-Credo, l'Arbresle, France) weighing 40–45 g were perfused with 100 μl of the solution into the tail vein for 30 s. The injected solution contained 1 μCi of lysoPC with [^{3}H]DHA (12 nmol) or 0.5 μCi of ^{14}C (acyl) and 2 μCi of ^{3}H (methyl choline) double-labeled lysoPC (50 nmol for arachidonate and 100 nmol for others). The same quantities of corresponding unesterified labeled fatty acids were used for control experiments in another group of rats. To control the accuracy of the injected dose, the radioactivity of an aliquot was measured before injection, each syringe-needle set was rinsed with saline after the injection, and the recovered radioactivity was determined. Within each type of experiment, the injected doses differed ≤5%. Animals were killed 2.5, 5, 7.5, 15, 30, and 60 min (2 animals at each time) after the end of perfusion, the vascular system was washed, and organs were removed and frozen in liquid nitrogen. Plasma kinetics were also studied in two animals from each group by collection of a few drops of blood at the tail tip at various times. Plasma volume was calculated from the body weight-to-plasma volume ratio previously measured in 10 control rats. The radioactivity remaining in the plasma at the different times was measured on 20- to 50-μl samples of plasma. This radioactivity was always >800 disintegrations/min.

Nonlipoprotein plasma proteins were isolated by density gradient ultracentrifugation to determine the concentration of fatty acids bound to albumin in the unesterified form and esterified in lysoPC. Two milliliters of NaCl in water at density (*d*) of 1.005, 0.2 ml of KBr in water at *d* of 1.18, 1 ml of plasma containing 5 nmol of BHT ajusted to *d* of 1.21 with use of solid KBr, and 0.2 ml of KBr solution at *d* of 1.24 were layered in a 3.4-ml centrifuge tube. Tubes were sealed and centrifuged for 1 h at 6 × 10^6 *g* in a Kontron rotor TFT 80.490 with use of a Kontron Centrikon T-1190 ultracentrifuge. This gradient ultracentrifugation allowed separation of all main lipoprotein fractions. Lipids of the nonlipoprotein plasma protein fraction were extracted (12) in the presence of 5 nmol of BHT, heptadecanoic acid, and *sn*-1-heptadecanoic acid-lysoPC as internal standards. FFA and lysoPC were separated by thin-layer chromatography (see below) with chloroform-methanol-water (65:25:4 vol/vol) under nitrogen atmosphere. Elution bands were scraped off, and powder was directly transmethylated with 5% sulfuric acid in methanol. Methyl esters were analyzed by gas-liquid chromatography.

Tissue lipids were extracted (12), and lipid classes were separated by preparative thin-layer chromatography (silica gel G 0.75 mm) by three elution systems: hexane-diethyl ether-acetic acid-methanol (90:20:2:3 vol/vol/vol/vol) for neutral lipids, chloroform-methanol-water (65:25:4 vol/vol/vol), and chloroform-methanol-methyl amine (40% in aqueous solution; 60:20:5 vol/vol/vol) (15) for phospholipid classes. The radioactivity of lipid classes and of aliquots from total lipids or the lipid extract aqueous phase was determined by liquid scintillation.

RESULTS

Figure 1 illustrates kinetics of plasma radioactivity after infusion of unesterified DHA and DHA esterified in lysoPC (lysoPC DHA) bound to albumin. The unesterified DHA quickly disappeared within the first 5 min, and in a second phase (after 15 min) some radioactivity was redistributed within plasma FFA and triglygerides (TG) after the lipoprotein secretion by the liver (not shown). The lysoPC DHA disappeared more slowly than the unesterified form after three phases: the first phase within the first 8 min, a slower second phase, and a very slow third phase after 50 min. All lysoPC and their corresponding unesterified fatty acids exhibited analogous kinetics (not shown), and the disappearance of radioactivity from choline and the fatty acid moiety of lysoPC was identical throughout the study period. More than 96% of the plasma radioactivity was still located in the lysoPC fraction 30 min after the injection.

The radioactivity recovery within lipids of different organs after the infusion of unesterified DHA or lysoPC DHA is reported in Fig. 2. The percentage of radioactivity from DHA recovered in brain lipids after lysoPC infusion was 10- to 12-fold higher than that observed when DHA was delivered in the unesterified form. Conversely the percentage of radioactivity from lysoPC

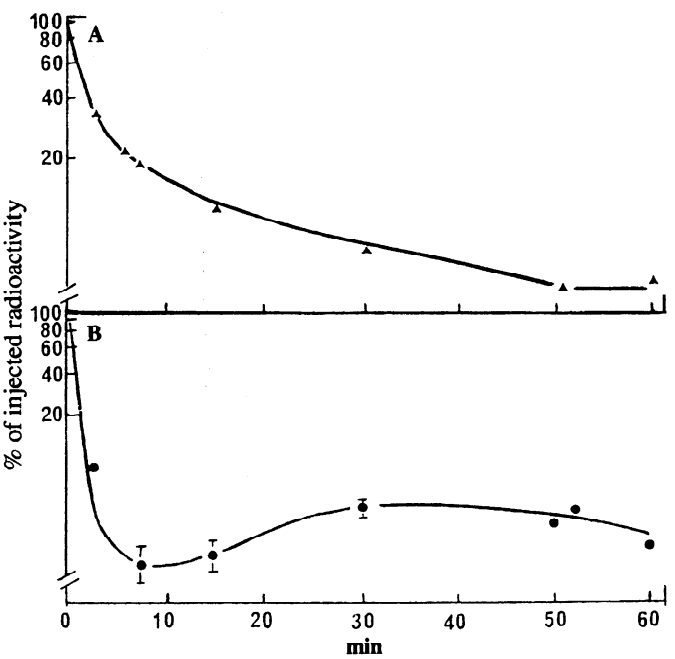

Fig. 1. Plasma decay curves of ^{3}H radioactivity after perfusion of 20-day-old rats with docosahexaenoic acid esterified at *sn*-2 position of lysophosphatidylcholine (*sn*-2-[^{3}H]lysoPC DHA; *A*) or unesterified [^{3}H]DHA (*B*) bound to albumin for 30 s. Values are means ± SD (n = 2). Analogous curves were observed when palmitic (16:0), oleic (18:1), linoleic (18:2), and arachidonic acids (20:4) were used in unesterified form or esterified in lysoPC.

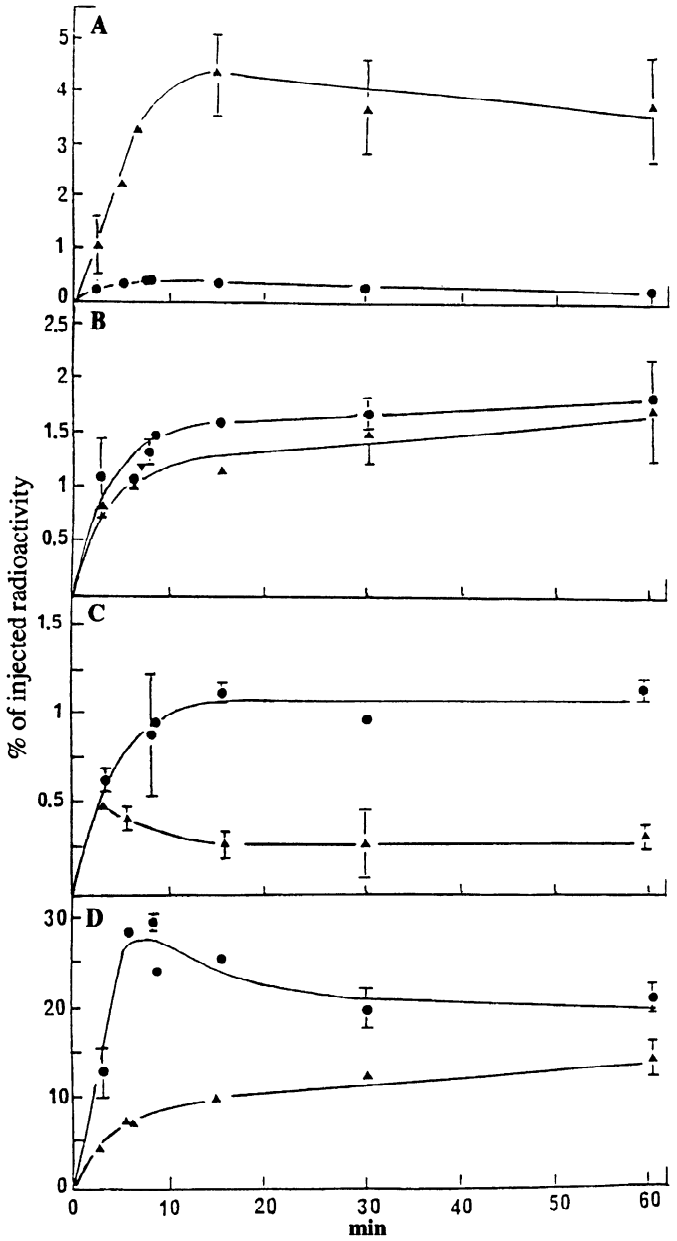

Fig. 2. Time course of recovery of ^3H radioactivity in total lipids of brain (*A*), kidney (*B*), heart (*C*), and liver (*D*) after perfusion of 20-day-old rats with *sn*-2-[^3H]lysoPC DHA (▲) or unesterified [^3H]DHA (●) bound to albumin for 30 s. Values are means ± SD (n = 2).

DHA recovered in other organs was lower than (liver and heart) or approximately the same as (kidney) that observed when unesterified DHA was used.

Time courses of the radioactivity from DHA within the main lipid classes of the heart, kidney, liver, and brain are reported in Figs. 3, 4, 5, and 6, respectively.

When lysoPC DHA was used, cardiac PC (Fig. 3*A*) was quickly and highly labeled, and its radioactivity decreased after 2.5 min, as did that of lysoPC, suggesting a direct uptake and acylation of lysoPC DHA. Cardiac FFA exhibited a time course analogous to that of PC, whereas the radioactivity of phosphatidylethanolamine (PE) was lower and increased progressively to reach a plateau from 15 min. Conversely, when unesterified DHA was injected (Fig. 3*B*), PE was, together with FFA,

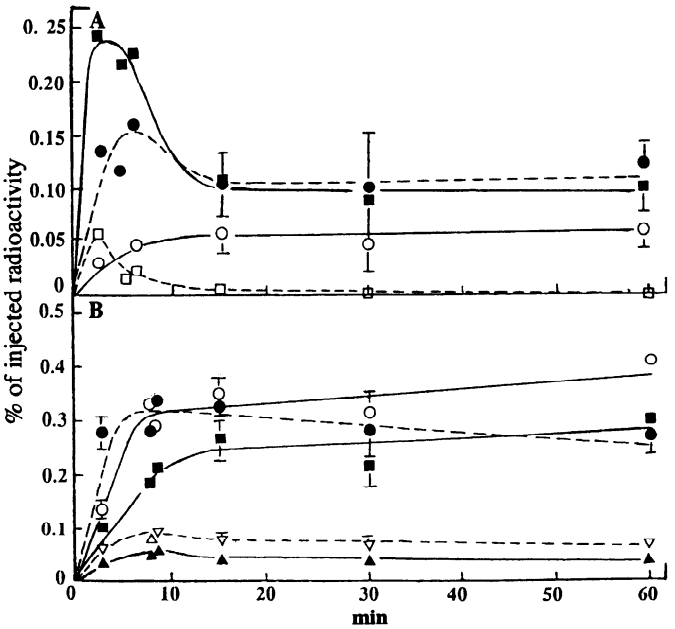

Fig. 3. Time course of ^3H radioactivity in lipid classes of heart after perfusion of 20-day-old rats with *sn*-2-[^3H]lysoPC DHA (*A*) or unesterified [^3H]DHA (*B*) bound to albumin for 30 s. ■, Phosphatidylcholine; □, lysophosphatidylcholine; ○, phosphatidylethanolamine; ●, free fatty acid; ▽, diglyceride; ▲, triglyceride. Values are means ± SD (n = 2).

the most labeled lipid class, with the PC radioactivity following a similar time course at a lower level. Moreover the radioactivity was also observed in diglycerides (DG) and TG, whereas the labeling of these fractions was very weak when lysoPC DHA was injected (and therefore not reported in Fig. 3*A*).

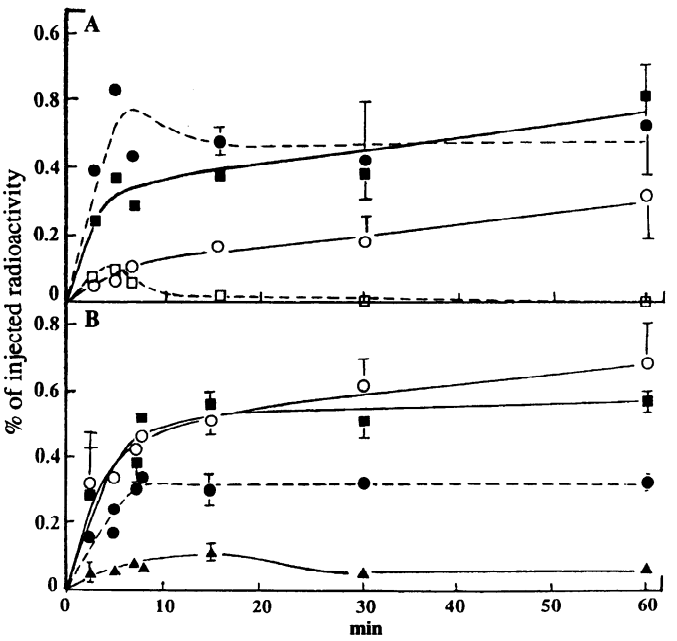

Fig. 4. Time course of ^3H radioactivity in lipid classes of kidney after perfusion of 20-day-old rats with *sn*-2-[^3H]lysoPC DHA (*A*) or unesterified [^3H]DHA (*B*) bound to albumin for 30 s. Symbols as in Fig. 3. Values are means ± SD (n = 2).

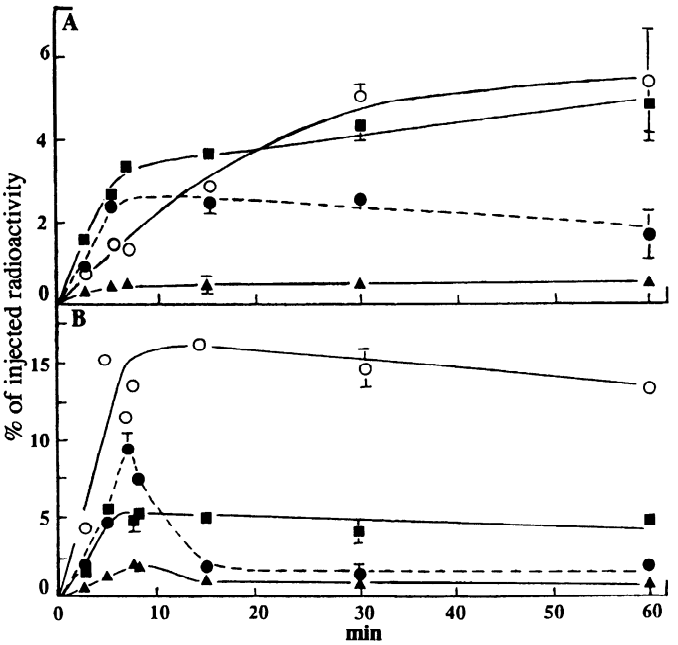

Fig. 5. Time course of ^3H radioactivity in lipid classes of liver after perfusion of 20-day-old rats with *sn*-2-[^3H]lysoPC DHA (*A*) or unesterified [^3H]DHA (*B*) bound to albumin for 30 s. Symbols as in Fig. 3. Values are means ± SD (n = 2).

The preferential incorporation of radioactivity into PC after injection of lysoPC DHA was also observed in the kidney (Fig. 4*A*), whereas PC and PE were labeled to the same extent when unesterified DHA was used (Fig. 4*B*). Only weak proportions of radioactivity were observed in TG after lysoPC DHA infusion (not shown), whereas this fraction was substantially labeled when unesterified DHA was used.

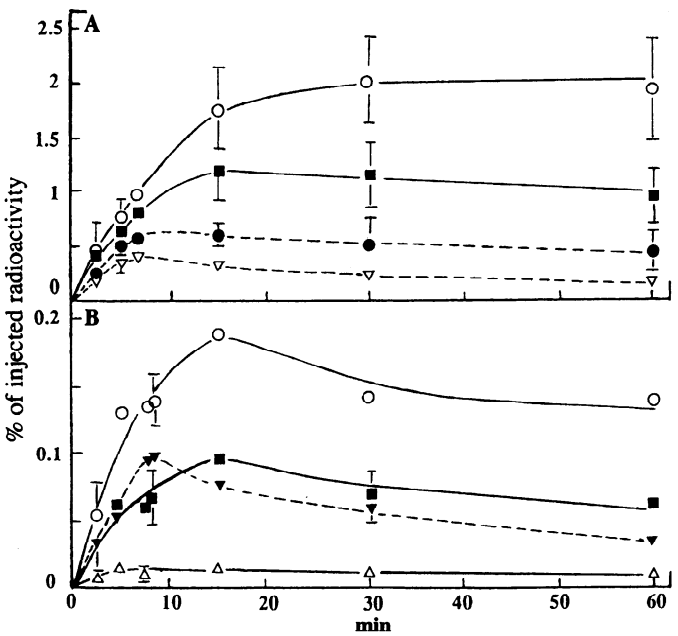

Fig. 6. Time course of ^3H radioactivity in lipid classes of brain after perfusion of 20-day-old rats with *sn*-2-[^3H]lysoPC DHA (*A*) or unesterified [^3H]DHA (*B*) bound to albumin for 30 s. △, Phosphatidylinositol; ▼, neutral lipid; other symbols as in Fig. 3. Values are means ± SD (n = 2).

In the liver, incorporation of unesterifed DHA was threefold higher in PE than in PC (Fig. 5*B*), whereas the use of lysoPC DHA (Fig. 5*A*) resulted in a preferential incorporation of DHA into PC within the first 15 min, with the labeling of PE quickly reaching the level observed in PC within 15 and 30 min after infusion. Hepatic synthesis of TG was slightly more important and transient when DHA was administered in the unesterified form than in the lysoPC form.

The differences between PC and PE according to the administration form of DHA were less clear in the brain than in the other organs. The percent radioactivity from total brain lipids recovered in PC and PE 2.5 min after injection of lysoPC DHA was 25.6 ± 1.2 and 28.2 ± 2, respectively, and 18.8 ± 1 and 37.5 ± 1.9 when the unesterified DHA was used. After 2.5 min, labeling was always about twofold higher for PE than for PC whether the esterified or unesterified form was used (Fig. 6). However, when lysoPC DHA was injected (Fig. 6*A*), PE was the sole lipid class in which the radioactivity increased after 15 min, whereas the radioactivity of the others (PC, DG, and FFA) slightly decreased. In all organs studied and for both supply forms, the radioactivity observed in phosphatidylinositol and phosphatidylserine was low (not shown), although the kidney exhibited a significant labeling of phosphatidylserine, and the proportion of phosphatidylinositol labeling was noticeable in the brain when the unesterified DHA was supplied (Fig. 6*B*).

When palmitic, oleic, linoleic, and arachidonic acids were used instead of DHA, all corresponding lysoPC species were taken up by the heart, kidney, and liver with the ratio between the radioactivity of the choline moiety and that of the fatty acid moiety being 0.9–0.8 in PC for any of these organs 2.5 min after injection (not shown). The initial ratio being 1, this suggests that lysoPC were mainly taken up without prior hydrolysis and then acylated at the *sn*-1 position. Unlike DHA, other fatty acids were preferentially incorporated in PC in all studied organs when they were supplied in the unesterified form. Table 1 shows that plasma concentrations of unesterified fatty acids bound to albumin differ from that of the corresponding lysoPC. The amounts of polyunsaturated lysoPC are higher than those found previously (28), presumably because the new method used in the present study for isolation of lysoPC prevented loss of polyunsaturated species. Instead of reporting all results

Table 1. *Plasma concentrations of 16:0, 18:1, 18:2, 20:4, and 22:6 bound to albumin in unesterified form or esterified in lysoPC*

	Unesterified Fatty Acid	LysoPC
16:0	126.8 ± 16.5	139.4 ± 2.6
18:1	121.8 ± 4.1	32.1 ± 5.3
18:2	131.1 ± 11.6	80.2 ± 16.7
20:4	24.4 ± 4.3	24.5 ± 6.7
22:6	9.7 ± 2.8	5.5 ± 0.4

Values are means ± SD in μM of 4 samples. Samples were mixtures of 2 ml of plasma from 5 20-day-old control rats (20 rats in total). LysoPC, lipophosphatidylcholine; 16:0, palmitic acid; 18:1, oleic acid; 18:2, linoleic acid; 20:4, arachidonic acid; 22:6, docosahexaenoic acid.

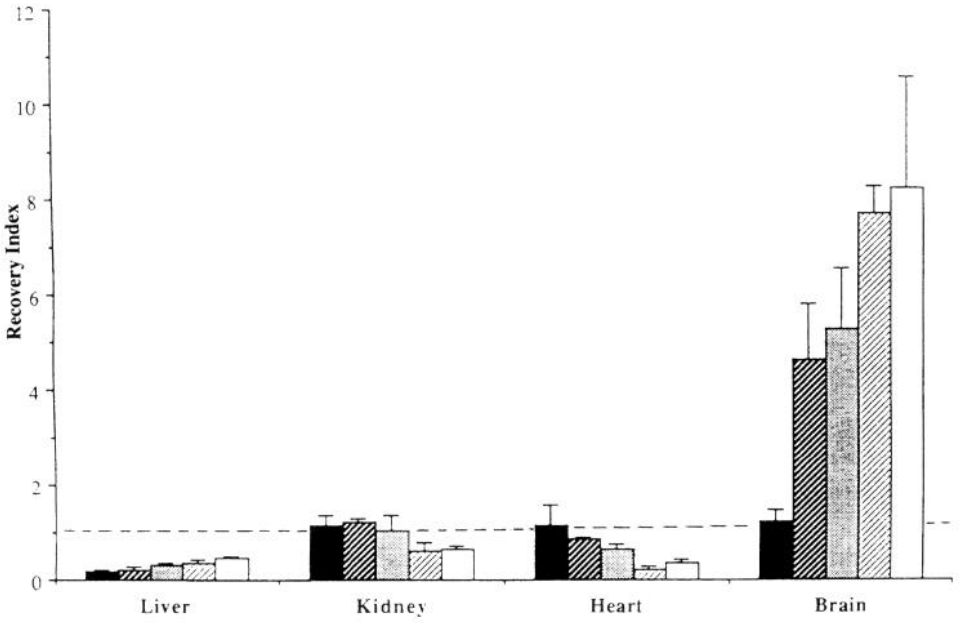

Fig. 7. Ratios of amounts of fatty acid moiety recovered in each organ after lysoPC infusion to amounts obtained after infusion of corresponding unesterified fatty acid: 16:0 (filled bars), 18:1 (dark hatched bars), 18:2 (stippled bars), 20:4 (light hatched bars), and 22:6 (open bars). Amounts were arbitrarily calculated by multiplying percent recovery in each organ by plasma content of each injected molecule (endogenous plus exogenous). Values are means ± SD ($n = 2$).

obtained for each organ and delivery form of each fatty acid, we have summarized the main differences between organs on the basis of the form of delivery and the nature of the fatty acid, i.e., the preference of organs for a specific form of supply (Fig. 7). The comparison was done 30 min after infusion, because the lipid radioactivity of most organs was at the plateau, except the liver, in which the radioactivity slightly decreased after 15 min. For example, at 30 min, 3.6, 3.2, 2.7, 2, and 0.34% (mean values) of the injected radioactivity from 22:6-, 20:4-, 18:2-, 18:1-, and 16:0-lysoPC, respectively, and 0.26, 0.46, 0.37, 0.19, and 0.31% of the injected radioactivity from the corresponding unesterified fatty acids, respectively, were recovered in the brain. Comparisons between organs were made on the basis of the ratios of the recovery of the lysoPC form to the recovery of the unesterified form. Unfortunately the amounts of fatty acids bound to albumin (as unesterified or lysoPC form) were too weak to be accurately determined on aliquots from plasma of small rats. Moreover, because the molecules were infused and not instantaneously injected as a bolus (to prevent tail vein rupture), it was not possible to calculate the integral of the radioactivity disappearance. Therefore the exact amount of fatty acid taken up by organs could not be determined. However, if the percentage of recovery observed in each organ at 30 min is applied to the number of circulating molecules (exogenous plus endogenous), an arbitrary mass of fatty acids recovered can be calculated for each type of experiment. Under these conditions, the ratio of the calculated amount obtained when lysoPC was used to the amount obtained when the unesterified form was used (circulating lysoPC × %recovery/circulating unesterified fatty acid × %recovery) reflects the ratio of the amounts present in the organ. A ratio >1 reflects a preferential utilization of the lysoPC form. Ratios reported in Fig. 7 show that the liver, heart, and kidney incorporated the unesterified form of all fatty acids preferentially or at the same extent, because all these ratios were ≤ 1. Conversely, the brain showed a net and very large preference for the lysoPC form of unsaturated fatty acids that increased with the degree of unsaturation. Moreover the brain did not exhibit such a preference when the fatty acid was saturated, the ratio for palmitic acid being ~ 1.

DISCUSSION

LysoPC DHA was preferentially utilized by the brain of young rats over unesterified DHA, and brain incorporation was slightly higher for lysoPC DHA than for lysoPC arachidonate. This extends to DHA our previous results showing that brain preferentially incorporates unsaturated lysoPC (28). DHA is mainly esterified in brain PE of many animal species (9), and injections of radioactive DHA bound to albumin showed that DHA is quickly incorporated into this brain lipid class (1, 11, 21, 24). This agrees with the preferential esterification of unesterified DHA into the brain PE.

We previously showed that the brain takes up lysoPC containing 16:0, 18:1, 18:2(n–6) or 20:4(n–6) without hydrolysis and converts them to PC preferentially to hydrolyze them for redistribution of their fatty acid moiety (28). Therefore the rapid incorporation of radioactivity from DHA into the brain PE after infusion of lysoPC DHA was relatively unexpected. Moreover the distribution of radioactivity from DHA among the brain lipid classes was very similar after 60 min in both types of experiment showing a preferential esterification of DHA into PE. However, it is likely that lysoPC DHA was nevertheless taken up by the brain without prior hydrolysis for the following reasons: *1*) incorporation of intact exogenous lysoPC within organs is a general phenomenon (4, 5, 16, 19, 27, 28); and *2*) brain PC radioactivity was close to that of PE 2.5 min after infusion of lysoPC DHA, whereas PE radioactivity was always twofold higher than PC radioactivity at any time when unesterified DHA was used.

Preferential acylation of lysoPC to form PC was observed for lysoPC DHA in other organs; e.g., the PE radioactivity reached that of PC in the liver after only 30 min. It is interesting to note that the heart, which is known to possess an active lysophospholipase A_2 (2), is the organ in which the radioactive lysoPC was the highest and persisting for a long time, suggesting a slower activity of lysophospholipase and/or acyltransferase activities for lysoPC DHA.

In all organs studied, the radioactivity of the FFA fraction was always high, even when DHA was supplied via lysoPC. This suggests that lysophospholipase was generally active, although it is possible that part of the PC formed by acylating lysoPC was also hydrolyzed and contributed to the FFA labeling. This is true at least for the brain PC, because the radioactive PC formed after lysoPC infusion slightly decreased and the radioactive PE increased after 15 min. However, this does not imply that the labeling of brain PE results only from the free DHA, because PE could also be formed by the deacylation/reacylation process. DHA proportions recovered in TG were low in all organs, and these proportions were always lower when lysoPC DHA was infused. The latter point could be an in vivo extension to DHA of results from a recent study (18) showing that the incubation of lysoPC with brain membranes inhibits the incorporation of arachidonic acid into TG in the presence of DG.

The main result of our study is the particular ability of the developing brain to incorporate exogenous unsaturated lysoPC, especially that containing DHA. In the absence of accurate measurements of kinetic parameters, a discussion of their respective influence would be speculative. It seems more judicious to examine the role of the blood-brain barrier. This incorporation cannot be related to an incomplete achievement, an eventual rupture of the blood-brain barrier, or an incorporation via choroid plexus, because brain-derived cells (neurons, oligodendrocytes, and fibroblasts from astroglia) have been found to be the least active cells among several cultured cells (including the kidney endothelium) to incorporate exogenous lysoPC (5). It is therefore likely that the ability of the 20-day-old rat brain to incorporate unsaturated lysoPC is due to the brain vascular endothelium. Usually, lysoPC quickly binds and enters the outer leaflet of plasma membranes, but its tranfer to the inner leaflet may last for different periods of times depending on the tissue. The transfer from the outer to the inner leaflets is very slow (several hours) in erythrocytes (4, 27) and faster (a few minutes) in other cells (10). It has recently been shown that the acylation of lysoPC to form PC is the limiting step for the transmembrane movement of palmitoyl-lysoPC (5). Because lysoPC hydrolysis can also occur (27) as the final step, it can be suggested that the preferential incorporation of unsaturated lysoPC by the developing rat brain would be due to the particular ability of brain vascular endothelium to acylate and/or hydrolyze this lysoPC species. However, it seems difficult to extend the results obtained with *sn*-1-palmitoyl-lysoPC (5) to *sn*-2 unsaturated acyl-lysoPC, because the brain did not preferentially incorporate palmitoyl-lysoPC over unesterified 16:0. On the other hand, transbilayer membrane movements can be also facilitated by "flipase" (10), and a transport system for specific fatty acids through the blood-brain barrier has been reported (25). Therefore the occurrence of a transport system specific for polyunsaturated lysoPC in the brain vascular endothelium membrane should be also considered.

Perspectives

Albumin, which plays an important role in the redistribution of DHA to organs (3), carries unesterified fatty acids and lysoPC, two supply forms of fatty acids to tissues. This work shows that the brain of 20-day-old rat, in contrast to other organs examined, preferentially used the latter form rather than the former when the fatty acid was DHA or a polyunsaturated fatty acid. This raises the question of the relevance of this phenomenon in the adult rat, in which the brain requirements for DHA and polyunsaturated fatty acids are likely to be smaller. On the other hand, DHA is mainly carried by PC and PE from lipoproteins; therefore studies on the relevance of these phospholipid classes to supply the brain with DHA could also be of great interest.

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