Biosynthesis of Phosphatidylcholine from a Phosphocholine Precursor Pool Derived from the Late Endosomal/Lysosomal Degradation of Sphingomyelin*

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Previous studies suggest that the steps of the CDP-choline pathway of phosphatidylcholine synthesis are tightly linked in a so-called metabolon. Evidence has been presented that only choline that enters cells through the choline transporter, and not phosphocholine administered to cells by membrane permeabilization, is incorporated into phosphatidylcholine. Here, we show that [14C]phosphocholine derived from the lysosomal degradation of [14C]choline-labeled sphingomyelin is incorporated as such into phosphatidylcholine in human and mouse fibroblasts. Low density lipoprotein receptor-mediated endocytosis was used to specifically deliver [14C]sphingomyelin to the lysosomal degradation pathway. Free labeled choline was not found either intracellularly or in the medium, not even when the cells were energy-depleted. Deficiency of lysosomal acid phosphatases in mouse or alkaline phosphatase in human fibroblasts did not affect the incorporation of lysosomal [14C]sphingomyelin-derived [14C]phosphocholine into phosphatidylcholine, supporting our finding that phosphocholine is not degraded to choline to be incorporated into phosphatidylcholine. Inhibition studies and analysis of molecular species showed that exogenous [3H]choline and sphingomyelin-derived [14C]phosphocholine are incorporated into phosphatidylcholine via a common pathway of synthesis. Our findings provide evidence that, in fibroblasts, phosphocholine derived from sphingomyelin is transported out of the lysosome and subsequently incorporated into sphingomyelin without prior hydrolysis of phosphocholine to choline. The findings do not support the existence of a phosphatidylcholine synthesis metabolon in fibroblasts.

De novo synthesis of phosphatidylcholine (PC) usually occurs via the CDP-choline pathway (1). Choline can enter the cell via the ATP-dependent choline transporter present in the cell membrane and is subsequently phosphorylated to phosphocholine (PCho) by the cytosolic enzyme choline kinase (EC 2.7.1.32). PC is then synthesized in two steps, catalyzed by the rate-limiting cytosolic and membrane-bound enzyme CTP:phosphocholine cytidylyltransferase (CT; EC 2.7.7.15) and the membrane-bound enzyme CDP-choline:s-1,2-diacylglycerol cholinephosphotransferase (EC 2.7.8.2). For some cell types, including fibroblasts, it has been reported that the steps of PC synthesis are tightly linked in a so-called metabolon (2–4). In these cell types only choline that entered the cells through the choline transporter was incorporated into PC. In contrast, PCho administered to cells by partial membrane permeabilization was not incorporated into PC (2, 4).

A possible endogenous source of PCho and choline is provided by the lysosomal degradation of PC and sphingomyelin (SM) by acid sphingomyelinase (5). We hypothesized that if only choline, and not PCho, can enter the PC metabolon and be incorporated into PC in human fibroblasts, then PCho that is formed during the degradation of SM by acid sphingomyelinase in the lysosome must be degraded to choline to be incorporated into PC. Candidate enzymes for intralysosomal hydrolysis of PCho to choline are lysosomal acid phosphatase (LAP) and tartrate-resistant acid phosphatase (TRAP) (6–8). Their in vivo substrates and functional roles are unclear. LAP is ubiquitously expressed, and TRAP is predominantly expressed in alveolar macrophages and osteoclasts (9), suggesting a specific function of the latter enzyme in these cell types. Another possible route for lysosomal SM-derived PCho to transport across the lysosomal membrane (by an unknown transporter), followed by hydrolysis to choline and subsequent incorporation of choline into the PC synthesis metabolon. A candidate enzyme for the hydrolysis of cytosolic PCho is alkaline phosphatase. In human skin fibroblasts, alkaline phosphatase is predominantly located at the plasma membrane (10, 11). Phosphoethanolamine and pyridoxal-5'-phosphate have been described as physiological substrates for this enzyme (10). We have investigated the possible role of LAP, TRAP, and alkaline phosphatase in the hydrolysis of PCho derived from the late endosome/lysosomal degradation of SM.

In this study we compared exogenous choline and late endosome/lysosome-derived PCho as substrates for PC synthesis. Hexadecylphosphocholine (HePC) was used as an inhibitor of PC synthesis (12–16). The results indicate that PCho derived from lysosomal SM is not degraded to choline but is incorporated into PC as such. We conclude that the PC synthesis
metabolon either does not exist in fibroblasts or is not as inaccessible to PCho as indicated by previous studies.

**EXPERIMENTAL PROCEDURES**

**Materials**—Culture flasks, Ham's F-10, Dulbecco's modified Eagle's medium with Glutamax, fetal bovine serum (FBS), penicillin, and streptomycin were obtained from Life Technologies, Inc. Culture plates (6 wells) were obtained from Greiner, Nuringen, Germany. Recombinant human apolipoprotein E3 (apoE) was obtained from PanVera Corporation (Madison, WI). HePC, 2-deoxyglucose, digitonin, and bovine serum albumin were obtained from Sigma. HPTLC plates (number 5633) and NaF were from Merck. [Choline-methyl-14C]sphingomyelin (specific activity, 54.5 Ci/mmol; >99% purity) was obtained from ICN Biomedicals, Costa Mesa, CA, and [methyl-2-14C]choline chloride (specific activity, 81 Ci/mmol) was obtained from PerkinElmer Life Sciences.

**Cell Culture**—Human fibroblasts from three healthy subjects and from hypophosphatasia (alkaline phosphatase deficiency), familial hypercholesterolemia (low density lipoprotein (LDL) receptor deficiency), and Niemann-Pick A (NP-A) (acid sphingomyelinase deficiency) patients were cultured as described previously (5). Primary embryonic (embryonic day 12.5) fibroblasts from LAP/TRAP double-deficient, and control mice were cultured in Dulbecco's modified Eagle's medium supplemented with 10% FBS, penicillin (10 IU/ml), and streptomycin (10 IU/ml). LAP/TRAP double-deficient mice are viable but more severely affected than single-deficient mice.

**14C]SM Incubations**—Cells were plated in 6-well plates (1.5–2.0 × 105 cells/well) and cultured for 4–5 days until confluence. They were then starved on Ham's F-10 or Dulbecco's modified Eagle's medium with 10% lipoprotein-deficient serum for 24 h to enhance LDL receptor expression. Subsequently, the medium was replaced with medium containing 0.8 nmol of [14C]SM (dissolved in ethanol; final concentration, 0.2%, v/v) in complex with apoE (5 µg/ml) ([14C]SM:apoE). Cells were incubated for 30 min at 37 °C, then the medium was replaced by medium (10% FBS) without [14C]SM:apoE, and cells were kept at 18 °C for 30 min to enable internalization of the lipid:apoE complex. Medium was replaced again with fresh medium (10% FBS) for chase studies at 37 °C for various periods of time. In some experiments 50 µM 2-deoxyglucose was present during the chase. For energy depletion, 10 mM NaN3 and 10 mM 2-deoxyglucose were dissolved in the chase medium. After incubations cells were trypsinized and pelleted.

**1HCholine Labeling**—Fibroblasts were plated as described above and incubated with 2 µCi of [1H]choline per well (dissolved in ethanol) in medium (10% FBS) for 2 h. In some experiments 50 µM HePC or 10 mM NaF + 10 mM 2-deoxyglucose was present during the incubation.

**Digitonin Permeabilization**—Digitonin permeabilization was done according to Vanders et al. (19), with slight modifications. Briefly, cells were incubated for 30 min with [14C]SM:apoE, washed, trypsinized and taken up in buffer (final pH 7.4) containing 150 mM KCl, 25 mM Tris-HCl, 2 mM EDTA, 10 mM KH2PO4, 0.1% (w/v) bovine serum albumin, and digitonin at different concentrations. After a 5-min incubation at 37 °C, the cells were pelleted by centrifugation. Hexosaminidase activity was measured in the pellets and the supernatant to determine the intactness of the lysosomal membrane (20). The protein concentration was determined according to Lowry et al. (21).

**Determination of Degradation Products**—The lipids in the pellets, supernatant, or medium were separated from aqueous metabolites with Folch extraction (22). The radiolabeled lipids were separated by HPTLC (23). Quantification of each radiolabeled product was achieved by liquid scintillation counting in 10 ml of Ultima Gold (Packard Instrument Co.).

**Analysis of the Molecular Species of PC, Synthesized from Exogenous [1H]Choline and from 14C]SM-Derived 14C]PC**—Fibroblasts were incubated with [1H]choline or [14C]SM:apoE as described above. The fibroblast pellets were homogenized, and lipids were extracted with chloroform:methanol (2:1, v/v) in the presence of 0.01 % butylated hydroxytoluene and washed according to Folch et al. (22). The lower phase was dried under N2, dissolved in 100 µl of chloroform:methanol (2:1, v/v), and PC was purified by preparative TLC as described (24). The PC molecular species were then separated by reverse-phase HPLC (25), and their radioactivity was determined by liquid scintillation counting. The identity of the major species was determined based on the retention time of standards, as well as by electrospray mass spectrometry, as described elsewhere (26).

**RESULTS**

**Validation of the Specific Uptake of [14C]SM:apoE by the LDL Receptor and Its Lysosomal Degradation**—To study the fate of lysosomal SM-derived PCho, fibroblasts were incubated with [14C]SM:apoE. The lipid:apoE complex was specifically taken up by the LDL receptor. This was verified by measurement of the uptake of [14C]SM:apoE by LDL receptor-deficient fibroblasts from a familial hypercholesterolemia patient. In these cells the uptake of [14C]SM was less than 8% compared with normal fibroblasts. When normal fibroblasts were incubated with [14C]SM, not in complex with apoE but in the presence of bovine serum albumin, the uptake of [14C]SM was negligible. Furthermore, LDL receptor-mediated uptake of [14C]SM:apoE ensured exclusive delivery of the labeled lipid to the lysosomal compartment. This was verified by incubating NP-A fibroblasts, which are deficient in the lysosomal enzyme acid sphingomyelinase, with [14C]SM:apoE. In these cells, [14C]SM:apoE was taken up in similar amounts as in normal cells, but [14C]SM was not degraded. Even after prolonged incubation, no labeled PCho or PC was formed (Fig. 1). These experiments show that [14C]SM:apoE uptake was mediated by the LDL receptor. [14C]SM was subsequently transported to the late endosomal/lysosomal compartment and degraded by acid sphingomyelinase in normal fibroblasts. Thus, incubation of fibroblasts with [14C]SM:apoE as described results in the hydrolysis of [14C]SM by acid sphingomyelinase in the lysosomal compartment, producing lysosomal SM-derived (endogenous) [14C]PCho.

**Lysosomal Degradation of [14C]SM**—The degradation of [14C]SM was determined after a 30-min incubation with [14C]SM:apoE (pulse) and during a 3-h chase in normal medium (Fig. 1). During the pulse period 36% of total intracellular SM was degraded to PCho. During the initial phase of the chase, production of labeled PCho was prominent. Thereafter, the amount of radiolabeled PCho gradually decreased following the appearance of labeled PC. At all time points the amount of radioactivity in the medium was negligible. The recovery of total radioactivity for all cell lines at each time point was 98.7 ± 3.2% as compared with t = 0. Free choline was not

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detected during pulse or chase, neither intracellularly nor in the medium.

Role of Acid Phosphatases in Incorporation of SM-derived PCho into PC—To determine the role of LAP and TRAP in the incorporation of lysosomal \[^{14}C\]SM-derived PCho into PC, primary embryonic fibroblasts from LAP-deficient, TRAP-deficient, or LAP/TRAP double-deficient mice were pulsed with \[^{14}C\]SM and apoE and subsequently chased. In each deficient cell type, \[^{14}C\]SM was similarly degraded compared with cells from normal mice (Fig. 2A). The degradation of \[^{14}C\]SM produced \[^{14}C\]PCho in all cell types (Fig. 2B). No labeled free choline was detected. Furthermore, labeled PCho was incorporated into PC in the deficient cells at rates comparable with or even faster than in normal cells (Fig. 2C). Thus, in mouse fibroblasts, acid phosphatase activity was not required for the incorporation of lysosomal \[^{14}C\]SM-derived PCho into PC.

Role of Alkaline Phosphatase in Incorporation of SM-derived PCho into PC—The role of alkaline phosphatase in the incorporation of lysosomal \[^{14}C\]SM-derived PCho into PC was determined with alkaline phosphatase-deficient fibroblasts. These cells were pulsed with \[^{14}C\]SM and apoE and subsequently chased. \[^{14}C\]SM was degraded to PCho, which was then incorporated into PC (Fig. 3). Labeled free choline was found neither in the cellular fraction nor in the medium. These results were similar to the degradation of \[^{14}C\]SM in normal fibroblasts (Fig. 1). This indicated that alkaline phosphatase did not play a role in the incorporation of lysosomal \[^{14}C\]SM-derived PCho into PC.

Effect of HePC and Energy Depletion on the Incorporation of Exogenous Choline and SM-derived PCho into PC—Administration of radiolabeled choline to normal fibroblasts via the culture medium resulted in its incorporation in intracellular PCho and PC (Fig. 4). The presence of the PC synthesis inhibitor HePC caused accumulation of PCho and decreased PC synthesis, in agreement with the reported specific inhibition by HePC of CT, the rate-limiting enzyme of PC synthesis (12–14, 16). Energy depletion, accomplished by the addition of NaN\(_3\) and 2-deoxyglucose, caused accumulation of intracellular choline and decreased the amounts of labeled PCho and PC (Fig. 4). The total choline uptake in the presence of NaN\(_3\) and 2-deoxyglucose was ~25% of the uptake in the absence of these additives (with or without HePC), showing that choline uptake is energy-dependent.

Addition of HePC after the incubation with \[^{14}C\]SM and apoE had no effect on the degradation of SM, but it inhibited PC synthesis from the endogenous PCho (Fig. 5). Energy depletion by the addition of NaN\(_3\) and 2-deoxyglucose during the chase resulted in slower degradation of SM and slower synthesis of labeled PC (Fig. 6). Thus, the incorporation of SM-derived PCho into PC is energy-dependent. Under conditions of energy depletion labeled free choline was detected neither intracellularly nor in the medium.

Subcellular Localization of \[^{14}C\]SM-derived PCho—To study the subcellular localization of \[^{14}C\]SM-derived PCho, normal human fibroblasts, pre-labeled with \[^{14}C\]SM and apoE, were...
derived PCho was present in the cytoplasm. Notably, the ad-
collection of exogenous [3H]choline into PCho and PC. Control fibroblasts were incubated with [3H]choline for 2 h without additives (control) or in the presence of 50 μM HePC or 10 mM NaN₃ + 10 mM 2-deoxyglucose (NaN3+2-DG). The percentage of total cell-associated radioactivity in PC (black bars), PCho (gray bars), and choline (white bars) is depicted. The mean total cell-associated radioactivity were 42.7 × 10⁴ ± 14.0 × 10³ dpm/mg cellular protein (control); 39.3 × 10⁴ ± 12.8 × 10³ dpm/mg cellular protein (HePC), and 11.9 × 10⁴ ± 2.1 × 10³ dpm/mg cellular protein (NaN3+2-DG). Results are depicted as means ± S.D. from three independent experiments.

We have shown that the uptake of [14C]SM and apoE for 30 min as described under “Experimental Procedures.” At indicated times during the chase, in the absence (closed symbols) or presence (open symbols) of energy poisons (10 mM NaN₃ + 10 mM 2-deoxyglucose), radioactivity in intracellular SM (circles), PCho (triangles), and PC (squares) was determined. Data are expressed as percentages of total cell-associated radioactivity at t = 0. The mean total cell-associated radioactivity was 32.9 × 10³ dpm/mg cellular protein. Values are given as means ± S.D.

Analysis of the Molecular Species of PC, Synthesized from [3H]Choline and [14C]SM—To investigate further whether SM-derived PCho could enter the PC synthesis metabolon, the molecular species of PC synthesized from [3H]choline or [14C]SM were analyzed. As can be seen in Fig. 8, the distribution of the radioactivity among PC species synthesized from both precursors is nearly identical, suggesting a common biosynthetic pathway.

**DISCUSSION**

In the present study we describe the incorporation of labeled PCho derived from the late endosomal/lysosomal degradation of SM into PC via the CDP-choline pathway of PC synthesis (1). We have shown that the uptake of [14C]SM-apoE was LDL receptor-mediated and that [14C]SM was exclusively metabolized in the late endosomal/lysosomal compartment, because NP-A fibroblasts, which are deficient in acid sphingomyelinase, did not metabolize [14C]SM. Also, in this cell line no evidence was found for the incorporation of SM-derived [14C]choline into PC, even after prolonged incubation. This finding excludes the

![Effect of HePC on the incorporation of SM-derived PCho into PC.](image1)

![Effect of energy depletion on the incorporation of SM-derived PCho into PC.](image2)

![Subcellular localization of radiolabeled PCho.](image3)
involvement of the reverse reaction catalyzed by SM synthase (27) in our experimental setup. In addition we have clearly shown that during the initial degradation of \( ^{14} \text{C}\text{SM} \) to \( ^{14} \text{C}\text{P}-\text{Cho} \), \( ^{14} \text{C}\text{PC} \) synthesis was negligible, indicating a lag phase in the process of PC synthesis from SM-derived PCho.

In these \textit{in situ} experiments, we did not observe production of radiolabeled free choline from SM, indicating that SM-derived PCho was not hydrolyzed to choline prior to its incorporation into PC. Even under conditions of energy depletion, when resynthesis of PCho from choline would be inhibited, no radiolabeled free choline was detected. These results corroborate our previous \textit{in vitro} studies with fibroblast homogenates at acidic pH, where we found PCho, and not choline, as an end product of PC degradation. PCho, which was predominantly generated by acid sphingomyelinase activity, was apparently not further degraded by acid phosphatases (5). Furthermore, in another \textit{in vitro} study the incubation of purified rat liver lysosomes with PC or PCho at acidic pH levels produced virtually no free choline (28).

The possible role of acid phosphatases in the degradation of PCho in the present \textit{in situ} study was investigated with embryonic fibroblasts from LAP and/or TRAP (double)-deficient mice. These mice have been developed to study the physiological role of the acid phosphatases (17, 18).\textsuperscript{2} The natural substrates for these enzymes have not been elucidated. However, an early study reported that PCho was a poor substrate for a purified rat liver acid phosphatase \textit{in vitro} (29). Our study shows that PCho is not an \textit{in situ} substrate for acid phosphatases present in mouse fibroblasts.

Because we have not found any indication that SM-derived PCho was hydrolyzed to free choline, it is likely that PCho is an end product of lysosomal degradation. However, most of the intracellular SM-derived PCho was present in the cytoplasm, not in the lysosomes, as was shown by digitonin-induced membrane permeabilization. This suggests that PCho must be transported out of the lysosomes by an as yet unknown transporter. Subsequently, it can serve as a substrate for PC synthesis. The experiments with alkaline phosphatase-deficient human fibroblasts indicate that PCho is not hydrolyzed in the cytosol by alkaline phosphatase. In conclusion, all results indicate that PCho is not hydrolyzed prior to its incorporation into PC.

Our data point to a direct incorporation of cytosolic PCho, endogenously produced by the late endosomal/lysosomal breakdown of SM, into PC. This is not compatible with previous reports that concluded that in C6 rat glioma cells, \( C_{6}H_{10}T_{1/2} \) fibroblasts, and rat hepatocytes the intermediates of PC synthesis were channeled to the enzymes involved, defining a so-called PC synthesis metabolon (2, 4). This conclusion was based on experiments in which PCho, which was delivered to the cytosol via partial membrane permeabilization, was not used for PC synthesis. However, our results show that PCho, which is derived from the lysosomal degradation of SM, is directly incorporated into PC via the CDP-choline pathway of PC synthesis. The latter is supported by the fact that HePC, a specific inhibitor of this pathway, inhibited the incorporation of both exogenous choline and endogenous PCho into PC. HePC has been shown by several groups to inhibit PC synthesis in different cell types (12–16), including human breast fibroblasts (30). HePC inhibits CT, the rate-limiting enzyme of PC synthesis, which results in the accumulation of PCho. In those previous studies with HePC, the incorporation of exogenous choline into PC was investigated. In the present study with human skin fibroblasts, the inhibition of PC synthesis from exogenous radiolabeled choline by HePC and the subsequent accumulation of PCho were confirmed. HePC also inhibited PC synthesis from lysosomal SM-derived PCho in these cells, suggesting that this PCho, like exogenous choline, entered the CDP-choline pathway of PC synthesis that is regulated by CT. The fact that the molecular species of PC synthesized from exogenous \( ^{3} \text{H}\)choline and of PC synthesized from late endosomal/lysosomal \( ^{14} \text{C}\text{PCho} \) were nearly identical indicates a common diacylglycerol precursor pool. This supports the idea that \( ^{14} \text{C}\text{SM} \)-derived PCho enters the normal PC biosynthetic pathway, rather than incorporating to PC via another route.

In conclusion, our results clearly indicate that in human skin fibroblasts, PCho that is produced by acid sphingomyelinase activity is transported intact from the late endosomal/lysosomal compartment to the cytoplasm and then enters the CDP-choline pathway of PC synthesis. This means that if the proposed PC synthesis metabolon exists, it is not as impermeable to intermediates of PC synthesis as indicated by previous studies (2, 4).

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REFERENCES
1. Kennedy, E. P., and Weiss, S. B. (1956) \textit{J. Biol. Chem.} 222, 193–214
2. George, T. P., Morash, S. C., Cook, H. W., Byers, D. M., Palmer, F. B. St. C., and Spence, M. W. (1989) \textit{Biochim. Biophys. Acta} 1004, 283–291
3. Zlatkine, P., Leroy, C., Moll, G., and Le Grémellec, C. (1996) \textit{Biochem. J.} 315, 983–987
4. Bladergroen, B. A., Geelen, M. J. H., Reddy, A. C. P., Declerq, P. E., and van Golde, L. M. G. (1998) \textit{Biochem. J.} 334, 511–517
5. Jansen, S. M., Groener, J. E. M., and Pouthuis, B. J. H. M. (1999) \textit{BBA} (Biochim. Biophys. Acta) \textbf{1436}, 363–369
6. Waheed, A., van Etten, R. L., Gieselmann, V., and von Figura, K. (1985) \textit{Biochem. Genet.} \textbf{23}, 309–319
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7. Waheed, A., Gottschalk, S., Hille, A., Krentler, C., Pohlmann, R., Braulke, T., Hauser, H., Geuze, H., and von Figura, K. (1988) EMBO J. 7, 2351–2358
8. Clark, S. A., Ambrose, W. W., Anderson, T. R., Terrell, R. S., and Toverud, S. U. (1989) J. Bone Miner. Res. 4, 399–405
9. Drexler, H. G., and Gignac, S. M. (1994) Leukemia 8, 359–368
10. Fedde, K. N., and Whyte, M. P. (1990) Am. J. Hum. Genet. 47, 767–775
11. Fedde, K. N., Cole, D. E. C., and Whyte, M. P. (1990) Am. J. Hum. Genet. 47, 776–783
12. Geilen, C. C., Wieder, T., and Reutter, W. (1991) J. Biol. Chem. 267, 6719–6724
13. Detmar, M., Geilen, C. C., Wieder, T., Orfanos, C. E., and Reutter, W. (1994) J. Invest. Dermatol. 102, 490–494
14. Geilen, C. C., Haase, A., Wieder, T., Arndt, D., Zeisig, R., and Reutter, W. (1994) J. Lipid Res. 35, 625–632
15. Posse de Chaves, K., Vance, D. E., Campenot, R. B., and Vance, J. E. (1995) Biochem. J. 312, 411–417
16. Boggs, K., Rock, C. O., and Jackowski, S. (1998) BBA (Biochim. Biophys. Acta) Libr. 1389, 1–12
17. Saffig, P., Hartmann, D., Lüllmann-Rauch, R., Wolff, J., Evers, M., Koster, A., Hetman, M., von Figura, K., and Peters, C. (1997) J. Biol. Chem. 272, 18628–18635
18. Hayman, N. R., Jones, S. J., Boyde, A., Foster, D., Colledge, W. H., Carlton, M. B., Evans, M. J., and Cox, T. M. (1996) Development 122, 3151–3162
19. Wanders, R. J. A., Ruiters, J. P. N., and Wijburg, F. A. (1993) BBA (Biochim. Biophys. Acta) Libr. 1181, 219–222
20. O’Brien, J. S., Okada, A., Chen, O. L., and Fillerup, D. L. (1970) N. Engl. J. Med. 283, 15–20
21. Lowry, O. H., Rosebrough, A. L., Farr, A. L., and Randall, R. J. (1951) J. Biol. Chem. 193, 265–275
22. Folch, J., Lees, M., and Sloane Stanley, G. H. (1957) J. Biol. Chem. 226, 497–509
23. Kunze, H., and Bohn, E. (1993) J. Chromatogr. 636, 221–229
24. Kates, M. (1991) Techniques of Lipidology 2nd Ed., pp. 251–253, Elsevier Science Publishers B. V., Amsterdam
25. Patton, G. M., Fasulo, J. M., and Robins, S. J. (1982) J. Lipid Res. 23, 190–196
26. Koivusalo, M., Haimi, P., Heikinheimo, L., Kostiainen, R., and Somerharju, P. (2001) J. Lipid Res. 42, 663–672
27. van Helvoort, A., van’t Hoff, W., Ritsma, T., Sandra, A., and van Meer, G., (1994) J. Biol. Chem. 269, 1763–1769
28. Kunze, H. (1993) BBA (Biochim. Biophys. Acta) Libr. 1169, 273–279
29. Arsenis, C., and Trueter, O. (1988) J. Biol. Chem. 263, 5702–5708
30. Wieder, T., Zhang, Z., Geilen, C. C., Orfanos, C. E., Giuliani, A. E., and Cabot, M. C. (1996) Cancer Lett. 106, 71–79
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