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Synthesis and Evaluation of the In Vivo Tolerance of Amido Fluorocarbon/Fluorocarbon and Fluorocarbon/Hydrocarbon Double-chain Phosphocholines Deriving from Diaminopropanols and Serine.

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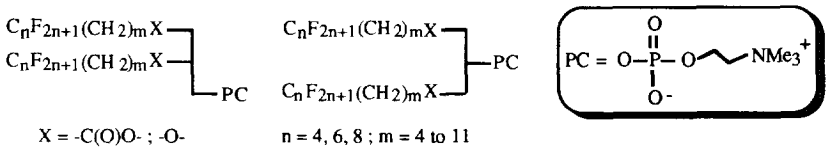
Abstract: The syntheses of various fluorocarbon/fluorocarbon and fluorocarbon/hydrocarbon amido-connected phosphocholines derived from diaminopropanols and serine are described. They were best obtained by phosphorylation of suitable alcohol precursors using 2-chloro-2-oxo-1,3,2-dioxaphospholane and subsequent ring opening with trimethylamine. The di-alkylamidopropanols were prepared by acylation,using perfluoroalkylated acid chlorides, of 1,3-diamino-2-propanol or of 2,3-diamino-1-propionic methylester followed by reduction of the ester bond. The perfluoroalkylated *N*-alkanoyl-serine alkylamides were prepared by condensation of Boc-*O*-Bn-L-serine with an aliphatic or perfluoroalkylated amine, Boc-deprotection, acylation and then hydrogenolysis for benzyl-deprotection. Acute toxicity evaluations indicate a very promising in vivo tolerance for these series of amido-linked compounds.

INTRODUCTION

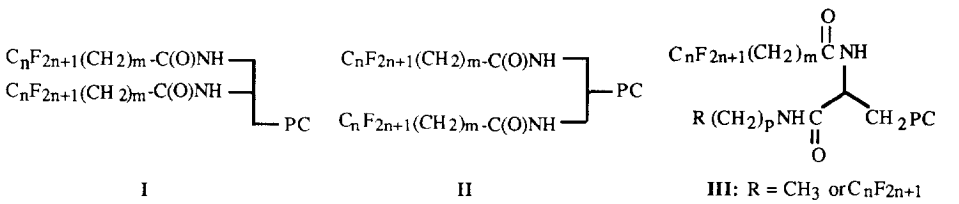
In modern medecine, drug delivery systems are often used in order to improve the pharmacokinetics, pharmacodynamics, and biodistribution of the drug, i.e. to enhance the efficacy of various biologically active materials, and to facilitate their intracellular delivery.[1] Liposomes (vesicles formed from natural or synthetic phospholipids) provide challenging drug delivery systems.[2] However, vesicles made from pure phospholipids have low stability. The development of more stable ones usually requires multicomponent systems and elaborate formulations resulting in increased complexity. Another approach in the elaboration of liposomal devices with new or significantly improved properties, lies in the development of components that are substantially different from those currently utilized. Highly fluorinated amphiphiles are such components: they offer some of the specific features that make up the uniqueness of fluorinated material, e.g. their hydrophobic *and* lipophobic character.[3]

Aiming at this goal, two series of analogs of phosphatidycholines (Scheme 1) having perfluoroalkylated chains connected to glycerol through ester[4] and ether[5] bonds have been synthesized in our laboratory. These fluorinated phospholipids form liposomes,[3,6,7] their fluorinated tails creating inside the liposomal membrane a highly hydrophobic and lipophobic fluorocarbon film. This film was indeed found to induce strong modifications of the physico-chemical and biological properties (membrane permeability, release of encapsulated material in biological media,[8] in vivo blood circulation time[9]) of the membranes and liposomes which these fluorinated phospholipids form.



Scheme 1 : Molecular structure of the perfluoroalkylated ester and ether glycerophosphocholines.

In order to extend the range of fluorinated phospholipids, we have now explored the synthesis of new perfluoroalkylated double-chain amido-connected phosphocholines **I** to **III** (Scheme 2). Their amide bond is intended to confer higher chemical and biological stability (in acidic media and more particularly towards the action of phospholipases)[10a] to these fluorinated phospholipids and to the liposomes that they will form. Furthermore, the amide linkage provides an important inter- and intra-molecular hydrogen bond capability.[10b,c] The formation of a hydrogen bond network within the membrane in proximity to the water interface is expected to enhance the physical and biological stability of the liposomes formed from these amido-phospholipids and to increase their in vivo blood circulation times, as it was found for liposomes formulated with sphingomyelin[11] which is a naturally occuring amido-phospholipid.



Scheme 2 : Molecular structure of the fluorocarbon/fluorocarbon and mixed fluorocarbon/hydrocarbon diamidopropanol- and diamidoserine-based phosphocholines **I** to **III**.

The three series of amide analogs of the fluorinated glycerophosphocholines (compounds **I, II** and **III** in Scheme 2) derive, respectively, from 2,3-diamino-1-propanol, 1,3-diamino-2-propanol and serine. The diaminopropanol skeletons were selected for the, a priori, rapid access they should provide to fluorocarbon/fluorocarbon double-chain amido-amphiphiles. Serine was chosen for its versatility: its different functionalities allows the stepwise connection of two hydrophobic chains which constitutes thus a flexible route to mixed fluorocarbon/fluorocarbon and fluorocarbon/hydrocarbon double-chain diamido-phospholipids. The molecular structures of **I** to **III** follow a modular design, which allows structural variations aimed at the establishment of structure/properties relationships. The structural features (fluorinated tails of various lengths, number of fluorocarbon chains, nature of the connecting unit) are intended to play a role on the hydrophobic-lipophobic/lipophilic/ hydrophilic balance, and consequently on the physico-chemical and biological properties (miscibility with natural phospholipids, permeability, drug release, stability in biological fluids, interactions with bio-compounds, in vivo fate) of the liposomes that these amphiphiles will form.

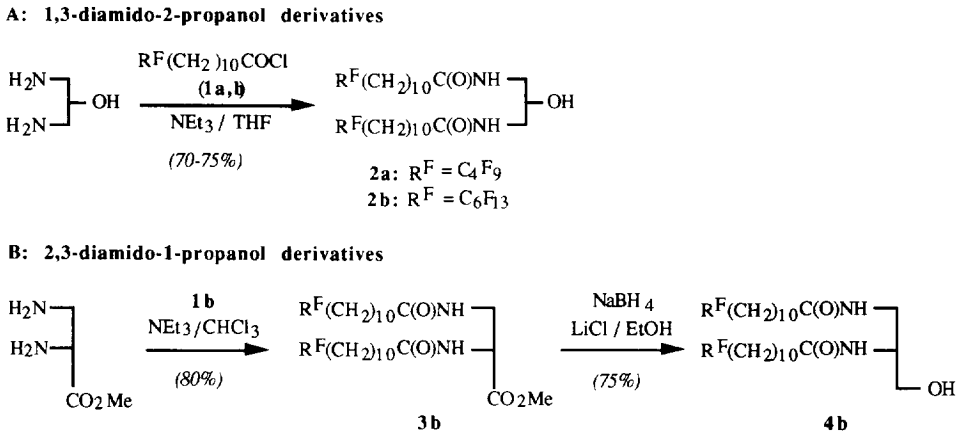
RESULTS AND DISCUSSION

Chemical synthesis of aliphatic long-chain mono- and bis-amido-phosphocholines deriving, respectively, from 2-amino-1,3-propanediol, 3-amino-1,2-propanediol[12] and 2,3-diamino-1-propanol[13] is well documented in the literature. It requires the preparation of suitable acylamino-propanol precursors, the remaining hydroxyl group being phosphorylated in a final step. The 2,3-diacylamino precursors are obtained from commercially available 1,2-diaminopropionic acid or asparagine[13] and aliphatic acid chlorides. Replacing the aliphatic acid chlorides by readily accessible perfluoroalkylated analogs[4] in these synthetic schemes should provide a route to the desired perfluoroalkylated 2,3-diamidopropan-1-phosphocholines.

To our knowledge, the synthesis of long chain diamido-phospholipids based on the commercially available 1,3-diamino-2-propanol and on serine, where serine serves as connecting unit between the hydrophobic part and the phospho-polar head, has not been investigated. Serine has been used as starting material for the synthesis of 2-acylamino-phosphatidylcholines[14,15] and of sphingolipids[16] and is part of the polar head in the well-known phosphatidylserines. We have recently used some of the double-chain bis-amido-serines presented here for the synthesis of perfluoroalkylated β-galactosyl amphiphiles.[17]

Fluorocarbon/fluorocarbon double-chain diamidopropanols.

The synthetic routes to the fluorinated 1,3- and racemic 2,3-diamidopropanols are depicted in Scheme 3. The 1,3-diamido-2-propanol derivatives **2a,b** were obtained in good yields (70-75%) directly from 1,3-diamino-2-propanol and the appropriate perfluoroalkylated acid chlorides **1a,b** without need of protecting the secondary alcohol, owing to its low reactivity.[18]



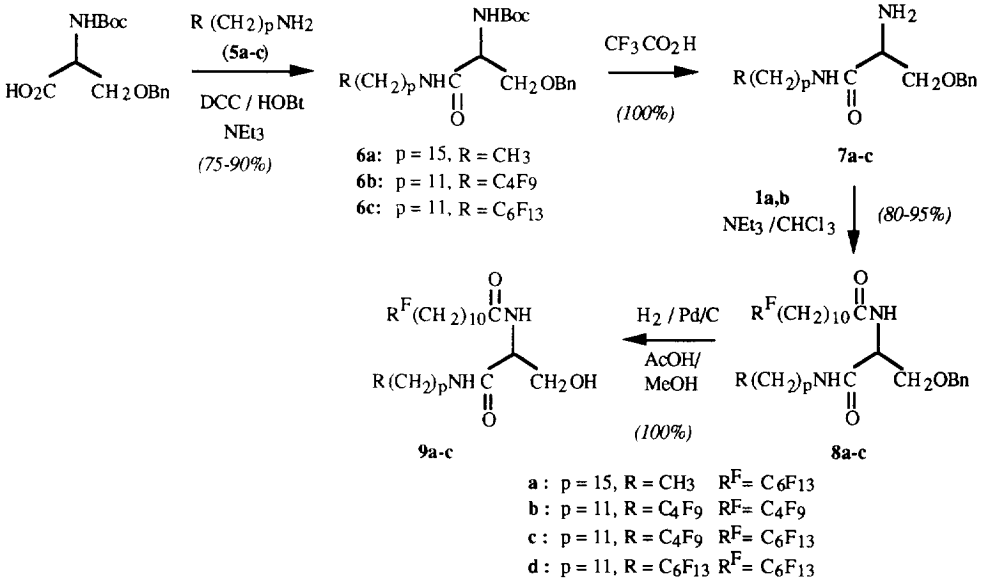
Scheme 3 : Synthetic pathways for the fluorocarbon/fluorocarbon double-chain (A) 1,3-diacylamino-2-propanol and (B) 2,3-diacylamino-1-propanol derivatives.

The synthesis of the fluorinated 2,3-diamido-1-propanol **4b** was performed according to the method described by Sunamoto and coll. in the hydrocarbon series[13] which uses the commercially available racemic 2,3-

diaminopropionic acid (as its methyl ester). This method first implies acylation of methyl-2,3-diaminopropionate using an aliphatic acid chloride, then reduction of the ester group in the resulting diamido compound by $NaBH_4$ in the presence of LiCl. When applied to a perfluoroalkylated acid chloride (e.g. **1b**), this method afforded, after acylation and reduction of **3b**, the corresponding fluorinated 1,2-diamidoalcohol **4b** in good yields (~60%).

Hydrocarbon/fluorocarbon and fluorocarbon/fluorocarbon diamido-serine derivatives

The more sophisticated synthetic route to the mixed hydrocarbon/fluorocarbon **9a** and fluorocarbon/fluorocarbon **9b-d** double-chain serine derivatives starting from protected Boc-*O*-Bn-L-serine is presented in Scheme 4. It consists in a four-step sequence which involves (i) condensation of Boc-*O*-Bn-L-serine with a long-chain (aliphatic or perfluoroalkylated) amine in the presence of DCC and HOBt, (ii) CF_3CO_2H mediated Boc-deprotection, (iii) acylation of **7a-c** with the appropriate perfluoroalkylated acid chloride and then (iv) hydrogenolysis for benzyl-deprotection. This route afforded the diamidoalcohols **9a-d** in almost 60% overall yields. The non-commercially available perfluoroalkylated amines **5b,c** used for the syntheses of **7b,c** were obtained by reduction, using $LiAlH_4$, of their corresponding perfluoroalkylated amides, which were prepared from acid chlorides **1a,b** and NH_3.



Scheme 4 : Synthetic pathway for the fluorocarbon/fluorocarbon and mixed fluorocarbon/hydrocarbon diamido-serine derivatives.

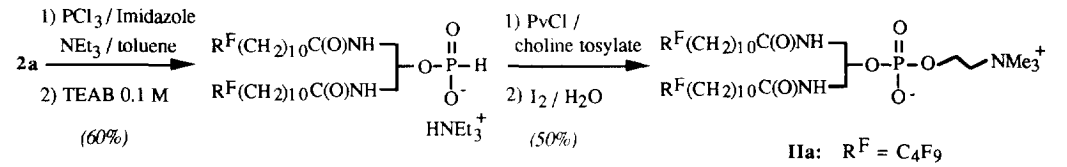
Fluorinated di-alkylamidophosphocholines.

The importance of phospholipids in biological processes has stimulated numerous studies concerning their chemistry and various routes[12,16] were therefore devised for their synthesis. The main difficulty in their

preparation often lies in the phosphorylation step. Furthermore, the presence of amido linkage(s) was shown to complicate substantially the phosphorylation step.[19]

The preparation of the fluorinated glycerophosphocholines displayed in Scheme 1 (60-90% yields) was achieved by phosphorylation of fluorinated di-*O*-acyl or di-*O*-alkylglycerols using either 2-bromoethyl dichlorophosphate and then amination with trimethylamine,[4] or phosphorus oxytrichloride and then condensation with choline tosylate[5] (the main difficulties in these syntheses were due to the low solubility of the fluorinated glycerol precursors). However, previous attempts to phosphorylate amido-propanol intermediates with 2-bromoethyl dichlorophosphate were not very successful[20] (low yields and poor reproducibility due mainly to low solubility of the amido intermediates, although it was reported recently that this method gave 1-alkylamido-2-*O*-alkyl-propan-3-phosphocholines in 23-40% yields[21]). $POCl_3$ proved also unsuitable with the formation of by-products resulting from polyphosphorylation and/or decomposition of the starting material.[21]

We therefore turned to the use of the H-phosphonate method[22] (Scheme 5) and of cyclic phosphorylation reagents, such as 2-chloro-2-oxo-1,2,3-dioxaphospholane[23,24] (Scheme 6), which proved very efficient. The former method consists first in reacting an alcohol with PIm_3 (obtained in situ from phosphorus trichloride and imidazole) and triethylamine, thus giving, after hydrolysis, the corresponding H-phosphonate intermediate. The phosphocholine derivative is then obtained by condensing this H-phosphonate with choline tosylate in the presence of pivaloyl chloride as activating agent, followed by oxidation with iodine. This method when applied to the hindered 1,3-diamidopropanol **2a**, afforded the corresponding phosphocholine **IIa** in an overall yield up to 30%. The major problem of this method lies in its poor reproducibility (mainly due to the low solubility of the diamidoalcohol in toluene which makes it difficult to control the reaction) and/or in the formation of by-products. Thus **IIb** could not be obtained from **2b** which is much less soluble in toluene than **2a**.



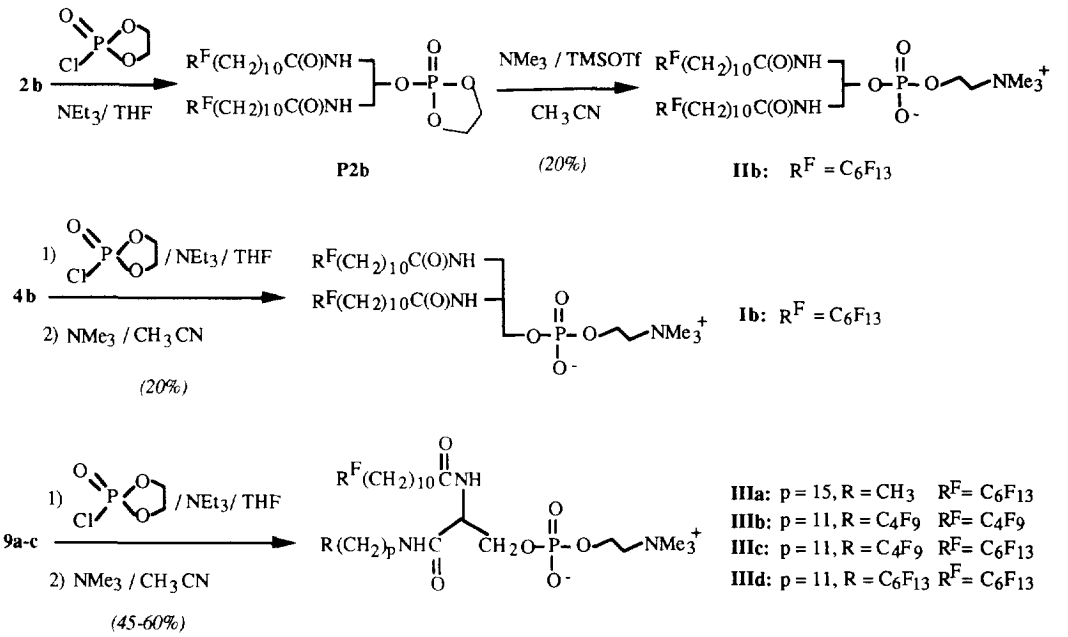
Scheme 5 : Synthetic pathway for the fluorocarbon/fluorocarbon 1,3-diamidopropan-2-phosphocholine **IIa** using the H-phosphonate method.

The phosphorylcholine moiety was also shown to be efficiently introduced onto amidoalcohols when using 2-chloro-2-oxo-1,2,3-dioxaphospholane and subsequent ring opening by NMe_3.[23] This is most probably related to the fact that 2-chloro-2-oxo-1,2,3-dioxaphospholane is a mono-functional reagent which, thus, is much less reactive towards the amido functionality than the above-mentioned phosphorylating agents.

When the amido-alcohols **2b**, **4b** and **9a-d** were phosphorylated with 2-chloro-2-oxo-1,3,2-dioxaphospholane in THF in the presence of triethylamine, and the cyclic phosphotriesters (such as **P2b**) thus obtained reacted with an excess of anhydrous NMe_3 (Scheme 6), yields in the 20-60% range of the diamido-propanol- and serine-based phosphocholines **IIb**, **Ib** and **IIIa-d**,[28] respectively, were obtained. The poor solubility of the fluorinated diamidoalcohols in THF and/or the lower reactivity of the secondary hydroxyl in the 1,3-diamido-2-propanol **2b** were, in part, responsible for the lower yields. We also found out, as it has already

been reported,[25] that phosphorylation and ring opening were improved under strict anhydrous conditions. Even traces of water appear to cause a substantial decrease in yield, most likely due to oxazoline formation. Replacing THF by $CHCl_3$ in which the diamidoalcohols are more soluble, led mainly to the formation of by-products, most probably due to an Arbusov-type reaction on the cyclic phosphotriester intermediate with the chloride anion,[26] owing to the solubility of the triethylammonium chloride salt in $CHCl_3$ (the formation of a chloro-diamido derivative was confirmed by elemental analysis).

By contrast to what has been observed for the synthesis of ether phospholipids,[27] yields in the amido phosphocholines **Ib** and **IIIa-d** were not improved when the ring opening reaction with NMe_3 in the cyclic phosphotriesters was mediated by TMSOTf.



Scheme 6 : Synthetic pathway for the perfluoroalkylated double-chain diamidopropanol- and diamidoserine-based phosphocholines **I** to **III** using the phosphotriester approach.

Biological acceptance

Biocompatibility is a major concern for drug carrier components. We therefore checked the in vivo tolerance of these new fluorinated amido-linked phospholipids. Our preliminary results from acute toxicity evaluations concerning the fluorocarbon/fluorocarbon phospholipids derived from 1,3-diaminopropanol (**IIa**) and serine (**IIIb**) which can be considered as representative, indicate a very promising in vivo tolerance for these series of amido-linked compounds. Acute maximum tolerated dose (MTD) values compatible with the survival of all injected animals.(10 mice) higher than 1050 and 2590 mg/kg body weight were indeed observed respectively for **IIa** and **IIIb**, when injected intraveneously as isotonic liposomal dispersions into the tail vein of the mice.

These two compounds are among the few perfluoroalkylated amphiphiles reported so far that have been found to exhibit such high MTD values, confirming that the presence of highly fluorinated tails does not affect acute toxicity.[3]

In conclusion, these syntheses provide easy and efficient routes to a wide range of perfluoroalkylated diamido-based phosphocholines. Phosphorylation of the diamidoalcohol precursors was best performed using 2-chloro-2-oxo-1,3,2-dioxaphospholane and subsequent ring opening with trimethylamine under strict anhydrous conditions. These perfluoroalkylated diamido-phosphocholines do form long-term shelf stable and heat-sterilizable liposomes. The potential of these liposomes as drug carrier and delivery systems, which include studies concerning membrane permeability and stability (with respect to encapsulated carboxyfluorescein release) in biological fluids, is most promising and will be reported elsewhere. Thus, we found that the release of carboxyfluorescein from liposomes when incubated in human serum is much lower when the liposomes are made from the perfluoroalkylated diamido-phosphocholines rather than from conventional ester-based phospholipids.

EXPERIMENTAL SECTION

General conditions

In most cases, the reactions were performed under anhydrous nitrogen using dry solvents and reagents. Anhydrous solvents were prepared by standard methods. The perfluoroalkylated acid chlorides were synthesized by reacting their corresponding acids (obtained from perfluoroalkyl iodides (Atochem) and commercially available α,ω-alkenoyl acids) with $SOCl_2$ according to reference 4. Choline tosylate was prepared by neutralizing a commercial aqueous 50% choline hydroxyde (Aldrich) with *p*-toluenesulfonic acid, dried then recristallized from acetone and stored under dry nitrogen. The 2-chloro-2-oxo-1,3,2-dioxaphospholane, trimethylsilylmethyl trifluoromethane-sulfonate (TMSOTf) and 1,3-diamino-propanol were purchased from Aldrich and used without further purification. Methyl-2,3-diaminopropionate was prepared from 2,3-diaminopropionic acid (Aldrich) according to the literature.[13] Boc-*O*-Bn-(L)-serine was purchased from Fluka.

Column chromatography purifications were carried out on silica gel 60 (Merck, 70-230 mesh). The purity of all the new compounds was checked by thin layer chromatography (TLC), NMR and/or elemental analysis. TLC analysis was performed on precoated silica gel F_{254} plates (Merck) with detection by UV, charring with $KMnO_4$ in NaOH 1N solution and, for the phospholipids, with Dragendorff's and Molybdenum Blue reagents (Sigma). Typically, *Rf* values of 0.35 ($CHCl_3$/MeOH/H_2O 65/25/4, v/v) were measured by TLC for the phosphocholines **I** to **III**. ^1H, ^{13}C, ^{19}F and ^{31}P NMR spectra were recorded at 200, 50.3, 188.3 and 81 MHz, respectively, on a Bruker AC 200 spectrometer. Chemical shifts (δ) are given in ppm relative to the signal (i) for internal reference Me_4Si or indirectly to $CHCl_3$ (δ 7.27) or CH_3OH (δ 3.35) for ^1H, (ii) for internal reference Me_4Si or indirectly to $CDCl_3$ (δ 76.9) or CD3OD (δ 48.8) for ^{13}C, (iii) for internal reference $CFCl_3$ for ^{19}F and (iv) for external reference H_3PO_4 75% for ^{31}P. Coupling constants are given in Hz. Elemental analysis were performed by the Service Central de Microanalyses of the CNRS.

Synthesis of the 1,3-diamido-2-propanol derivatives 2.

N,N'-di-(11-(F-butyl)-undecanoyl)-1,3-diamino-2-propanol, **2a**, (Procedure A).

A solution of 11-(*F*-butyl)undecanoyl chloride **1a** (4.0 g, 9.4 mmol) in 15 mL of THF was added dropwise, at room temperature, to a solution of 1,3 diaminopropan-2-ol (0.4 g, 4.5 mmol) and triethylamine (1.3 mL, 9.4 mmol) in 90 mL THF. After stirring for 4h at room temperature, the mixture was diluted with 20 mL of water, then poured into 200 mL of ice-cooled water. The crude product was extracted with 300 mL of Et_2O. The organic layer was dried over Na_2SO_4 and the solvent was removed in vacuo. Purification by column chromatography (elution with Et_2O, then with ethyl acetate) afforded compound **2a** (2.7 g, 70%) as a white powder.
^1H NMR ($CDCl_3$): δ 1.20 (br s, 24H, $(CH_2)_6$),1.50 (m, 8H, $C\underline{H}_2CH_2CO$ and $C\underline{H}_2CH_2CF_2$), 1.95 (tt, ^3J = 8.2, $^3J_{HF}$ = 18.0, 4H, CH_2CF_2), 2.15 (t, ^3J = 8.7, 4H, CH_2CO), 3.00-3.45 (m, 4H, CH_2N), 3.70 (quintet, ^3J = 4.9, 1H, CH), 6.70 (t, ^3J = 6.0, 2H, NH). ^{13}C NMR ($CDCl_3$): δ 20.2 (t, $^3J_{CF}$ = 4, $\underline{C}H_2CH_2CF_2$), 25.9 (s, $\underline{C}H_2CH_2CO$), 29.2, 29.3, 29.4, 29.5 (all s, $(CH_2)_6$), 30.9 (t, $^2J_{CF}$ = 22, $\underline{C}H_2CF_2$), 36.8 (s, $\underline{C}H_2CO$), 42.7 (s, CH_2N), 70.4 (s, CH), 175.3 (s, CO). ^{19}F NMR ($CDCl_3$): δ -81.6 (3F, CF_3), -115.1 (2F, CF_2CH_2), -125.0 (2F, $C\underline{F}_2CF_2CH_2$), -126.6 (2F, $CF_3C\underline{F}_2$).

N,N'-di-(11-(F-hexyl)-undecanoyl)-1,3-diamino-2-propanol, **2b**.
Procedure A, when applied to 1,3-diaminopropan-2-ol (0.44 g, 4.9 mmol), triethylamine (1.4 mL, 10.4 mmol) and 11-(*F*-hexyl)-undecanoylchloride (5.4 g, 10.3 mmol) afforded, after chromatography (elution with $CHCl_3$ then $CHCl_3$/MeOH 98/2), compound **2b** (4.0 g, 77%) as a white powder.
^1H and ^{13}C NMR ($CDCl_3$) identical to those of **2a**. ^{19}F NMR ($CDCl_3$): -81.3 (3F, CF_3), -114.9 (2F, CF_2CH_2), -122.4, -123.4, -124.1 (2F, 2F, 2F, $(C\underline{F}_2)_3CF_2CH_2$), -126.6 (2F, $CF_3C\underline{F}_2$).

Synthesis of the 2,3-diamido-1-propanol derivative 4

N,N'-di-(11-(F-hexyl)-undecanoyl) -2,3-diamino-1-propanol, **4b**.
Procedure A when applied to a solution of 11-(*F*-hexyl)-undecanoyl chloride **1b** (9.4 g, 17.9 mmol) in 35 mL of $CHCl_3$ and a solution of methyl-2,3-diaminopropionate (1.5 g, 7.1 mmol), triethylamine (9.9 mL, 7.1 mmol) in 50 mL of $CHCl_3$, afforded, after usual work-up and chromatography (elution with $CHCl_3$) 6.2 g (80%) of **3b** as a white powder. [**3b**: ^1H NMR ($CDCl_3$): δ 130 (br s, 24H, $(CH_2)_6$), 1.55 (m, 8H, $C\underline{H}_2CH_2CO$ and $C\underline{H}_2CH_2CF_2$), 2.05 (tt, ^3J = 8.5, $^3J_{HF}$ = 19.0, 4H, CH_2CF_2), 2.15 and 2.20 (t, t, ^3J = 8.0, 2H, 2H, CH_2CO), 3.60 (m, 2H, CH_2N), 3.75 (s, 3H, OMe), 4.70 (m, 1H, CHN), 6.10 (t, ^3J = 7.0, 1H, $N\underline{H}CH_2$), 6.90 (d, ^3J = 7.0, 1H, $N\underline{H}CH$). ^{13}C NMR ($CDCl_3$): δ 20.1 (t, $^3J_{CF}$ = 4, $\underline{C}H_2CH_2CF_2$), 25.5 and 25.6 (s, s, $\underline{C}H_2CH_2CO$), 29.1, 29.2, 29.3, 29.4 (all s, $(CH_2)_6$), 30.9 (t, $^2J_{CF}$ = 22, $\underline{C}H_2CF_2$), 36.5 and, 36.6 (s, s, $\underline{C}H_2CO$), 41.8 (s, CH_2N), 52.7 and 53.5 (s, s, CHN and OCH_3), 170.8 (s, $\underline{C}O_2CH_3$), 173.8 and 174.6 (s, s, CONH). ^{19}F NMR ($CDCl_3$) identical to that of **2b**]. Reduction of **3b** (3 g, 2.8 mmol) using $NaBH_4$ (0.32 g, 8.5 mmol) and LiCl (0.35 g, 8.3 mmol) in 50 mL of ethanol, under reflux for 6h, yielded a precipitate which was removed by filtration and washed with chloroform. The filtrate was then concentrated under vacuo and the crude product was chromatographied (elution with $CHCl_3$/MeOH 99/1) giving 2 g (70%) of **4b** as a white powder.
^1H NMR ($CDCl_3/CD_3OD$): δ 1.20 (br s, 24H, $(CH_2)_6$), 1.55 (m, 8H, $C\underline{H}_2CH_2CO$ and $C\underline{H}_2CH_2CF_2$), 2.05 (tt, ^3J = 8.5, $^3J_{HF}$ = 19.0, 4H, CH_2CF_2), 2.10 (m, 4H, CH_2CO), 3.10 (m, 2H, CH_2N), 3.25-3.65 (m, 2H, CH_2OH), 3.70 (m, 1H, CHN). ^{13}C NMR ($CDCl_3/CD_3OD$): δ 20.0 (t, $^3J_{CF}$ = 4, $\underline{C}H_2CH_2CF_2$), 25.6 (s, $\underline{C}H_2CH_2CO$), 29.0, 29.1, 29.2, 29.3 (all s, $(CH_2)_6$), 30.7 (t, $^2J_{CF}$ = 22, $\underline{C}H_2CF_2$), 36.3 and 36.4 (s, s, $\underline{C}H_2CO$), 39.4 (s, CH_2N), 51.3 (s, CHN), 61.1 (s, CH_2OH), 174.6 and 175.8 (s, s, CO). ^{19}F NMR ($CDCl_3/CD_3OD$) identical to that of **2b**.

Synthesis of the diamido-L-serine derivatives 9.

11-(F-alkyl)-undecylamine **5a,b.**

11-(*F*-butyl)-undecylamine **5a**. A stream of NH_3 (11.0 g, 0.65 mol) was led through a flask containing a solution of 11-(*F*-butyl)-undecanoyl chloride **1a** (19.4 g, (3.7 mmol) in Et_2O (250 mL). The reaction mixture was stirred at room temperature for 20h. Then, the mixture was poured into 200 mL of water, extracted with $CHCl_3$. The organic layer was then dried over Na_2SO_4 and evaporated. The solid residue was washed with hexane, yielding, after filtration, 10.7 g (72%) of 11-(*F*-butyl)-undecylamide as a white powder [^{1}H NMR ($CDCl_3$) : 1.25 (br s, 12H, $(CH_2)_6$), 1.50 (m, 4H, $CH_2CH_2CF_2$ and CH_2CH_2CO), 1.95 (tt, ^3J = 7.0, $^3J_{HF}$ = 18.0, CH_2CF_2), 2.15 (t, ^3J = 8.0, CH_2CO), 5.50 and 6.05 (m, m, 1H, 1H, NH_2). ^{13}C NMR ($CDCl_3$): δ 20.1 (t, $^3J_{CF}$ = 4, $CH_2CH_2CF_2$), 25.5 (s, CH_2CH_2CO), 29.1, 29.2, 29.3 (all s, $(CH_2)_6$), 30.8 (t, $^2J_{CF}$ = 22, CH_2CF_2), 35.9 (s, CH_2CO), 176.1 (s, CO)]. To a solution of 11-(*F*-butyl)-undecylamide (10.4 g, 25.7 mmol) in 150 ml THF, were added 1.9 g (51 mmol) of $LiAlH_4$. The resulting mixture was stirred at reflux for 24h. After careful hydrolysis, filtration and evaporation, the crude product was chromatographied (elution with $CHCl_3$/MeOH from 95/5 to 7/3). The amine **5a** was precipitated as its hydrochloride salt (7.0 g, 70%) from a $CHCl_3$ solution by adding a few drops of HCl 12N.

^{1}H NMR (CD_3OD) : δ 1.35 (brs, 12H, $(CH_2)_6$), 1.65 (m, 4H, $CH_2CH_2CF_2$ and CH_2CH_2N), 2.15 (tt, ^3J = 7.0, $^3J_{HF}$ = 18.0, CH_2CF_2), 2.95 (t, ^3J = 8.0, CH_2N). ^{13}C NMR (CD_3OD) :δ 21.1 (t, $^3J_{CF}$ = 4, $CH_2CH_2CF_2$), 27.3 (s, CH_2CH_2N), 28.4 (s, CH_2), 29.9, 30.0, 30.2, 30.3, 30.4 (all s, $(CH_2)_6$), 31.6 (t, $^2J_{CF}$ = 22, CH_2CF_2), 40.7 (s, CH_2N) ^{19}F NMR identical to that of **2a**.

11-(*F*-hexyl)undecylamine, **5b**. The same procedure, when applied to 11-(*F*-hexyl)-undecanoyl chloride **1b** (8.3 g (15.8 mmol) and NH_3 afforded 7.5 g (95%) of 11-(*F*-hexyl)-undecylamide as a white powder [^{1}H and ^{13}C NMR ($CDCl_3$) identical to that of 11-(*F*-butyl)-undecyl-amide], and, after reduction, chromatography and precipitation with HCl, **5b** as its hydrochloride salt (75%).

^{1}H and ^{13}C NMR ($CDCl_3$/CD_3OD) identical to those of **5a**. ^{19}F NMR identical to that of **2b**.

N-(11-F-hexyl)-undecanoyl)-(L)-serine hexadecylamide, **9a.**

Synthesis of **6a** (Procedure B). To a solution of Boc-*O*-Bn-L-Ser (3.0 g, 10.2 mmol), triethylamine (1.4 mL, 10.2 mmol), hexadecylamine (2.5 g, 10.2 mmol) and HOBt (1.4 g, 10.4 mmol) in 75 mL of DMF, was added, dropwise and at 0°C, a solution of DCC (2.3 g, 11.2 mmol) in 20 mL DMF. After 30 min at 0°C, the reaction mixture was heated at 50°C for 20h. DMF was then removed in vacuo and the crude product purified by chromatography (elution with $CHCl_3$) to give 4.1 g (78%) of **6a** as a white powder.

^{1}H NMR ($CDCl_3$): δ 0.85 (t, ^3J = 7.0, 3H, Me), 1.20 (br s, 26H, $(CH_2)_{13}$), 1.40 (s and m, 11H, Me_3C and CH_2CH_2N), 3.18 (td, ^3J = 6.7, ^3J = 6.5, 2H, CH_2NH), 3.50 and 3.85 (AB part of an ABX system, $^2J_{AB}$ = 9.2, $^3J_{AX}$ = 6.7, $^3J_{BX}$ = 3.9, 2H, CH_2OBn), 4.20 (m, 1H, CH), 4.43 and 4.52 (AB system, $^2J_{AB}$ = 11.7, 2H, OCH_2Ph), 5.35 and 6.35 (m, m, 1H, 1H, NHBoc and CH_2NHCO), 7.30 (m, 5H, Ph) .^{13}C NMR ($CDCl_3$) :δ 14.1 (s, Me), 22.7 (s, CH_2Me), 26.9 (s, CH_2CH_2N), 28.3 (s, Me_3C), 29.3, 29.4, 29.5, 29.6, 29.7, 29.8 (all s, $(CH_2)_{11}$), 31.9 (s, $CH_2(CH_2)_2N$), 39.6 (s, CH_2N), 54.1 (s, CHN), 70.1 (s, CH_2OBn), 73.5 (s, OCH_2Ph), 80.3 (s, CMe_3), 127.8 and 128.5 (s, Cortho,meta), 127.9 (s, Cpara), 137.5 (s, $CH_2C(Ph)$), 155.6 (s, OCONH), 170.1 (s, CONH).

Synthesis of **7a** (Boc-deprotection, Procedure C). 3.8 g (7.4 mmol) of **6a** were stirred at room temperature in 10 mL of CF_3CO_2H for 1h. The solution was evaporated to dryness. The crude product was then dissolved in $CHCl_3$, washed with a 10% Na_2CO_3 solution and dried. After removal of the solvent, 3.08 g (100%) of **7a** were obtained as a white powder.

^1H NMR ($CDCl_3$): δ 0.85 (t, ^3J = 7.0, 3H, CH_3), 1.20 (br s, 26H, $(CH_2)_{13}$), 1.40 (m, 2H, CH_2CH_2N), 1.65 (m, 2H, NH_2), 3.15 (td, 2H, CH_2NH, ^3J = 6.7, ^3J = 6.8), 3.45-3.75 (m, 3H, CH_2OBn and $CHNH_2$), 4.50 (s, 2H, OCH_2Ph), 7.30 (m, 5H, Ph).^{13}C NMR, ($CDCl_3$): δ 14.1 (s, CH_3), 22.7 (s, CH_2CH_3), 27.0 (s, CH_2CH_2N), 29.3, 29.4, 29.5, 29.6, 29.7, 29.8 (all s, $(CH_2)_{11}$), 32.0 (s, $CH_2(CH_2)_2N$), 39.2 (s, CH_2N), 55.1 (s, CHN), 72.5 (s, CH_2OBn), 73.3 (s, OCH_2Ph), 127.7 and 128.5 (s, C ortho and meta), 127.8 (s, C para), 137.9 (s, $CH_2C(Ph)$), 172.4 (s, CO).

Synthesis of **8a**. Procedure A when applied to **7a** (2.5 g, 6.1 mmol), 11-(*F*-hexyl)-undecanoyl chloride **1b** (3.9 g, 7.5 mmol) and triethylamine (1.0 mL, 7.5 mmol), afforded, after stirring at room temperature for 24h, work-up and chromatography (elution with $CHCl_3$), 5.0 g (92%) of **8a** as a white powder.

^1H NMR ($CDCl_3$): δ 0.85 (t, ^3J = 7.0, 3H, CH_3), 1.30 (br s, 40H, $(CH_2)_7$ and $(CH_2)_{13}$), 1.65 (m, 4H, CH_2CH_2CO and $CH_2CH_2CF_2$), 2.05 (tt, $^3J_{HF}$ = 19.0, ^3J = 8.5, 2H, CH_2CF_2), 2.30 (t, ^3J = 7.0, 2H, CH_2CO), 3.25 (td, ^3J = 6.5, ^3J = 6.1, 2H, CH_2NH), 3.55 and 3.90 (AB part of an ABX system, $^2J_{AB}$ = 9.1, $^3J_{AX}$ = 7.9, $^3J_{BX}$ = 4.3, 2H, CH_2OBn), 4.55 and 4.65 (AB system, $^2J_{AB}$ = 11.8, 2H, OCH_2Ph), 4.60 (m, 1H, CH), 6.50 (d, ^3J = 6.7, 1H, CHNH), 6.55 (t, ^3J = 5.6, 1H, CH_2NH), 7.35 (m, 5H, Ph). ^{13}C NMR ($CDCl_3$): δ 14.1 (s, CH_3), 20.1 (t, $^3J_{CF}$ = 4, $CH_2CH_2CF_2$), 22.7 (s, CH_2CH_3), 25.6 (s, CH_2CH_2CO), 26.9 (s, CH_2CH_2N), 29.1, 29.2, 29.3, 29.4, 29.5, 29.6, 29.7 (all s, $(CH_2)_{11}$ and $(CH_2)_6$), 30.9 (t, CH_2CF_2, $^2J_{CF}$ = 22), 31.9 (s, $CH_2(CH_2)_2N$), 36.6 (s, CH_2CO), 39.7 (s, CH_2N), 52.2 (s, CHN), 69.6 (s, CH_2OBn), 73.5 (s, $PhCH_2O$), 127.8 and 128.6 (s, Cortho and meta), 128.0 (s, Cpara), 137.5 (s, $CH_2C(Ph)$, 170.0 (s, CH_2CO), 173.3 (s, CHCO). ^{19}F NMR ($CDCl_3$) identical to that of **2b.**

Synthesis of **9a** (debenzylation, Procedure D). To a solution of **7a** (5.1 g, 5.6 mmol) in 40 mL methanol and 10 mL acetic acid was added Pd (10%) on charcoal (0.5 g). A slow stream of hydrogen was led through the flask for 2h while the reaction mixture was warmed to 50°C. When the hydrogenolysis was achieved (TLC control), the catalyst was filtered off over celite, washed with $CHCl_3$. The solvents were then evaporated and the residue washed with water and dried under vacuo to give 4.4 g (100%) of **9a**.

^1H NMR ($CDCl_3$/CD_3OD): δ 0.75 (t, ^3J = 7.0, 3H, CH_3), 1.20 (br s, 38H, $(CH_2)_6$ and $(CH_2)_{13}$), 1.50 (m, 6H, CH_2CH_2CO and $CH_2CH_2CF_2$), 1.90 (tt, $^3J_{HF}$ = 19.0, ^3J = 8.5, 2H, CH_2CF_2), 2.10 (t, ^3J = 8.0, 2H, CH_2CO), 3.05 (t, ^3J = 7.1, 2H, CH_2NH), 3.50 and 3.70 (AB part of an ABX system, CH_2OH, $^2J_{AB}$ = 11.2, $^3J_{AX}$ ~ 5, $^3J_{BX}$ = 4.8), 4.25 (t, J ~ 5, 1H, CHN). ^{13}C NMR ($CDCl_3$/CD_3OD): from 14.1 (s, CH_3) to 39.7 (s, CH_2N) identical to that of **8a**, 54.2 (s, CHN), 62.1 (s, CH_2OH), 170.5 (s, CH_2CO), 174.4 (s, CHCO).

N-(11-(F-butyl)-undecanoyl)-L-serine 11-(F-butyl)-undecylamide, **9b**.

Synthesis of **6b**. Procedure B, when applied to Boc-*O*-Bn-L-Ser (2.8 g, 9.4 mmol), triethylamine (2.6 mL, 18.8 mmol), HOBt (1.3 g, 9.6 mmol), 11-(*F*-butyl)-undecylamine (4.0 g, 9.4 mmol) and DCC (2.1 g, 10.3 mmol) in 100 mL $CHCl_3$, afforded, after 20h of reaction at room temperature and purification by chromatography (elution with $CHCl_3$), 4.8 g (77 %) of **6b** as a white powder.

^1H NMR ($CDCl_3$): δ 1.20 (br s, 16H, $(CH_2)_8$), 1.35 (s and m, 11H, Me_3C and CH_2CH_2N), 1.50 (m, 2H, $CH_2CH_2CF_2$), 1.95 (tt, ^3J = 7.5, $^3J_{HF}$ = 18.0, 2H, CH_2CF_2), 3.15 (td, ^3J = 6.5, ^3J = 6.4, 2H, CH_2NH),

3.45 and 3.85 (AB part of an ABX system, $^2J_{AB}$ = 9.2, $^3J_{AX}$ = 6.6, $^3J_{BX}$ = 4.0, 2H, CH_2OBn), 4.2 (m, 1H, CH), 4.40 and 4.48 (AB system, $^2J_{AB}$ = 11.8, 2H, OCH_2Ph), 5.30 (m, 1H, NHBoc), 6.35 (t, ^3J = 7.0, 1H, CH_2NHCO), 7.30 (m, 5H, Ph). ^{13}C NMR ($CDCl_3$): δ 20.1 (t, $CH_2CH_2CF_2$, $^3J_{CF}$ = 4), 26.8 (s, CH_2CH_2N), 28.3 (s, Me_3C), 29.1, 29.2, 29.3, 29.4, 29.5, 29.6 (all s, $(CH_2)_7$), 30.8 (t, CH_2CF_2, $^2J_{CF}$ = 22), 39.6 (s, CH_2N), 54.1 (s, CH), 70.1(s, CH_2OBn), 73.4 (s, OCH_2Ph), 80.2(s, CMe_3), 127.7 and 128.5 (s, Cortho and meta), 127.9 (s, Cpara), 137.6 (s, $CH_2C(Ph)$), 155.5 (s, OC(O)NH), 170.1 (s, C(O)NH). ^{19}F NMR ($CDCl_3$) identical to that of **2a**.

Synthesis of **7b**. Boc-deprotection (Procedure C) of **6b** (2.4 g, 3.6 mmol) in CF_3CO_2H afforded 2.0 g (100%) of **7b** as a white powder.

^1H NMR ($CDCl_3$): δ 1.20 (br s, 16H, $(CH_2)_8$), 1.40 (m, 2H, CH_2CH_2N), 1.65 (s, 2H, NH_2), 2.0 (tt, ^3J = 7.0, $^3J_{HF}$ = 18.0 , 2H, CH_2CF_2,), 3.15 (td, ^3J = 6.7, ^3J = 6.6, 2H, CH_2NH), 3.45-3.75 (m, 3H, CH_2OBn and CH), 4.45 (s, 2H, OCH_2Ph), 7.25 (m, 5H, Ph). ^{13}C NMR ($CDCl_3$): δ 20.1 (t, $CH_2CH_2CF_2$, $^3J_{CF}$ = 4), 26.9 (s, CH_2CH_2N), 29.1, 29.2, 29.3, 29.4, 29.5, 29.6 (all s, $(CH_2)_7$), 30.8 (t, CH_2CF_2, $^2J_{CF}$ = 22), 39.1 (s, CH_2N), 55.1 (s, CHN), 72.5 (s, CH_2OBn), 73.3 (s, OCH_2Ph), 127.7 and 128.4 (s, C ortho and meta), 127.8 (s, C para), 137.9 (s, $CH_2C(Ph)$), 172.5 (s, C(O)NH). ^{19}F NMR ($CDCl_3$) identical to that of **2a**.

Synthesis of **8b**. Procedure A, when applied to **7b** (2.0 g, 3.6 mmol), 11-(*F*-butyl)-undecanoyl chloride (1.8 g, 4.3 mmol) and triethylamine (0.6 mL, 4.3 mmol), afforded after chromatography (elution with $CHCl_3$), 3.2 g (93%) of **7b** as a white powder.

^1H NMR ($CDCl_3$): δ 1.30 (br s, 30H, $(CH_2)_6$ and $(CH_2)_9$), 1.65 (m, 4H, CH_2CH_2CO and $CH_2CH_2CF_2$), 2.05 (tt, $^3J_{HF}$ = 19.0 , ^3J = 8.5, 4H, CH_2CF_2), then identical to that of **8a**. ^{13}C NMR ($CDCl_3$): 20.1 (t, $^3J_{CF}$ = 4, $CH_2CH_2CF_2$), 25.6 (s, CH_2CH_2CO), 26.8 (s, CH_2CH_2N), 29. 0, 29.1, 29.2, 29.3, 29.4, 29.5 (all s, $(CH_2)_6$ and $(CH_2)_7$), 30.8 (t, $^2J_{CF}$ = 23, CH_2CF_2), 36.6 (s, CH_2CO) then identical to that of **7a**. ^{19}F NMR ($CDCl_3$) identical to that of **2a**.

Synthesis of **9b**. Procedure D, when applied to **8b** (2.9 g, 3.0 mmol), 30 mL of methanol, 10 mL of acetic acid and 0.29 g of Pd/C, afforded 2.6 g (100%) of **9b** as a white powder.

^1H NMR ($CDCl_3/CD_3OD$): δ 1.25 (br s, 30H, $(CH_2)_6$ and $(CH_2)_9$), 1.50 (m, 4H CH_2CH_2CO and $CH_2CH_2CF_2$), 1.98 (tt, $^3J_{HF}$ = 19.0, ^3J = 8.5, 4H, CH_2CF_2), 2.18 (t, ^3J = 7.2, 2H, CH_2CO), 3.1 (t, ^3J = 7.4, 2H, CH_2N), 3.42 and 3.80 (AB part of an ABX system, CH_2OH, $^2J_{AB}$ = 11.3, $^3J_{AX}$ = 6.2, $^3J_{BX}$ = 4.4), 4.28 (dd, 1H, CHN, X part of the ABX system), and, in $CDCl_3$, 6.80 (d, ^3J = 7.0, 1H, NHCH), 7.05 (t, ^3J = 6.5, 1H, CH_2NH). ^{13}C NMR ($CDCl_3/CD_3OD$): 20.0 (t, $CH_2CH_2CF_2$, $^3J_{CF}$ = 4), 25.5 (s, CH_2CH_2CO), 26.8 (s, CH_2CH_2N), 29. 0, 29.1, 29.2, 29.3, 29.4, 29.5 (all s, $(CH_2)_6$ and $(CH_2)_7$), 30.7 (t, $^2J_{CF}$ = 22, CH_2CF_2), 36.3 (s, CH_2CO), 39.5 and 39.6 (s, s, CH_2N), 54.1 and 54.2 (s, s, CHN), 62.5 (s, CH_2OH), 170.8 and 170.9 (s, s, CH_2CONH), 174.5 and 174.6 (s, s, $CHCONH$); the splitting of some ^{13}C lines is due cis and trans amide conformers). ^{19}F NMR ($CDCl_3$, CD_3OD) identical to that of **2a**.

N-(11-(F-hexyl)-undecanoyl)-L-serine 11-(F-butyl)-undecylamide , **9c**.

Synthesis of **6c**. Procedure B, when applied to Boc-*O*-Bn-L-Ser (1.1 g, 3.8 mmol), triethylamine (1.1 mL, 7.6 mmol), HOBt (0.52 g, 3.9 mmol), 11-(*F*-hexyl)-undecylamine (2.0 g, 3.8 mmol) and DCC (0.86 g, 4.2 mmol) in 40 mL of $CHCl_3$, afforded after stirring at room temperature for 20h and purification by chromatography (elution with $CHCl_3$), 2.7 g (92%) of **6c** as a white powder.

^1H and ^{13}CNMR ($CDCl_3$) identical to those of **6b**. ^{19}F NMR ($CDCl_3$) identical to that of **2b**.

Synthesis of **7c**. Procedure C, when applied to **6c** (2.6 g, 3.4 mmol) in CF_3CO_2H afforded 2.2 g (100%) of **7c** as a white powder.

^1H and ^{13}C NMR ($CDCl_3$) identical to those of **7b**. ^{19}F NMR ($CDCl_3$) identical to that of **2b.**

Synthesis of **8c**. Procedure A, when applied to **7b** (1.5 g, 2.7 mmol), 11-(*F*-hexyl)-undecanoyl chloride (1.8 g, 3.5 mmol) and triethylamine (0.5 mL, 3.5 mmol), afforded, after chromatography (elution with $CHCl_3$), 2.5 g (87%) of **8c** as a white powder.

^1H NMR ($CDCl_3$) identical to that of **7b**. ^{13}C NMR ($CDCl_3$) identical to that of **8b** . ^{19}F NMR ($CDCl_3$): -81.4, -81.6 (3F, 3F, CF_3), -115.0 (4F, CF_2CH_2), -122.5, -123.4, -124.1, 125.0 (2F, 2F, 2F, 2F ($\underline{C}F_2)_3CF_2CH_2$), -126.6 (4F, $CF_3\underline{C}F_2$).

Synthesis of **9c**. Debenzylation of **8c** (2.16 g, 2.1 mmol) using procedure D (30 mL of methanol, 10 mL of acetic acid and 0.2 g of Pd/C) afforded 2 g (100%) of **9c** as a white powder.

^1H NMR ($CDCl_3/CD_3OD$): δ 1.25 (br s, 30H, $(CH_2)_6$ and $(CH_2)_9$), 1.50 (m, 4H, $C\underline{H}_2CH_2CO$ and $C\underline{H}_2CH_2CF_2$), 1.98 (tt, $^3J_{HF}$ = 19.0, ^3J = 8.5, 4H, CH_2CF_2), 2.18 (t, 2H, CH_2CO, ^3J = 8.0), 3.13 (t, 2H, CH_2N, ^3J = 7.0), 3.58 and 3.75 (AB part of an ABX system, CH_2OH, $^2J_{AB}$ = 11.1, $^3J_{AX}$ = 5.9, $^3J_{BX}$ = 5.0), 4.28 (dd, 1H, CHN, X part of the ABX system). ^{13}C NMR ($CDCl_3/CD_3OD$): 19.7 (t, $^3J_{CF}$ = 4, $\underline{C}H_2CH_2CF_2$), 25.3 (s, $\underline{C}H_2CH_2CO$), 26.5 (s, $\underline{C}H_2CH_2N$), 28.7, 28.8, 28.9, 29.0, 29.1(all s, $(CH_2)_6$ and $(CH_2)_7$), 30.4 (t, $^2J_{CF}$ = 22, $\underline{C}H_2CF_2$), 30.5 (t, $^2J_{CF}$ = 22, $\underline{C}H_2CF_2$), 35.9 (s, $\underline{C}H_2CO$), 39.2 (s, CH_2N), 54.3 (s, CHN), 61.9 (s, CH_2OH), 170.5 (s, $CH_2\underline{C}ONH$), 174.4 (s, $CH\underline{C}ONH$). ^{19}F NMR ($CDCl_3/CD_3OD$) identical to that of **8c**.

N-(11-(F-hexyl)-undecanoyl)-L-serine 11-(F-hexyl)-undecylamide, **9d**.

Synthesis of **8d**. Procedure A, when applied to **7c** (1.9 g, 2.9 mmol), 11-(*F*-hexyl)-undecanoyl chloride (1.9 g, 3.6 mmol) and triethylamine (0.5 mL, 3.6 mmol), afforded, after chromatography ($CHCl_3$), 2.7 g (80%) of **8d** as a white powder.

^1H and ^{13}C NMR ($CDCl_3$) identical to those of **8b** . ^{19}F NMR ($CDCl_3$): identical to that of **2b**.

Synthesis of **9d**. Procedure D, when applied to 2.1 g (1.9 mmol) of **8d,** 30 mL of methanol, 10 mL of acetic acid and 0.2 g of Pd/C afforded 1.9 g (100%) of **9d** as a white powder.

^1H and ^{13}C NMR ($CDCl_3/CD_3OD$) identical to those of **9b**. ^{19}F NMR ($CDCl_3/CD_3OD$): identical to that of **2b**.

Synthesis of phosphocholine derivatives I to III.

N,N'-di-[11-(F-butyl)-undecanoyl]-1,3-diaminopropan-2-phospho-N,N,N-trimethylethanolamine, **IIa**.

To a stirred solution of imidazole (1.66 g, 24.4 mmol) in 20 mL of toluene, was added dropwise and at 0°C, a solution of PCl_3 (0.46 mL, 5.2 mmol) in 6 mL of toluene, then a solution of triethylamine (1.9 mL, 13.7 mmol) in 6 mL toluene. Stirring was continued for 10 min, before the temperature was lowered to -5°C. Then a solution of **2a** (1.5 g, 1.7 mmol) in 25 mL of toluene was added dropwise. The reaction mixture was stirred at 0°C for 3 h. The toluene was then removed under vacuo and $CHCl_3$ was added. The organic layer was washed with a 0.1M solution of triethylammonium bicarbonate (TEAB). Chromatography of the residue obtained after evaporation of $CHCl_3$, (elution with $CHCl_3$/MeOH from 100/0 to 8/1) afforded 1.0 g (62%) of N,N'-di(11-(*F*-butyl)undecanoyl)-1,3-diaminopropan-2-H-phosphonate triethylammonium salt as a waxy solid [^1H NMR (CD_3OD): δ 1.25 (br s, 30H, $(CH_2)_6$ and $(CH_2)_9$), 1.55 (m, 4H, $C\underline{H}_2CH_2CO$ and $C\underline{H}_2CH_2CF_2$), 2.05 (tt, $^3J_{HF}$ = 19.0, ^3J = 8.5, 4H, CH_2CF_2), 2.20 (t, ^3J = 7.2, 2H, CH_2CO), 3.20 (m, 2H, NCH_2), 3.45 (m, 2H, OCH_2), 4.20 (m, 1H, CH), 6.70 (d, $^1J_{PH}$ = 644, PH); ^{31}P NMR (CD_3OD): δ 1.13 (d, $^1J_{PH}$ = 644)]. Pivaloyl chloride

(0.26 mL, 2.1 mmol) was added to a 20 mL pyridine solution of the H-phosphonate (0.64 g, 0.69 mmol) and choline tosylate (0.47 g, 1.7 mmol), which have been dried prealably by evaporation of dry pyridine. The reaction mixture was stirred at room temperature for 17h. Then 0.4 mL water and 0.35 g (1.4 mmol) iodine were added. The mixture was stirred for 2h and evaporated under vacuo. Then, $CHCl_3$ was added and the organic phase was washed with a saturated solution of sodium thiosulfate. The aqueous phase was washed back with $CHCl_3$. The combined organic phases were concentrated under vacuo. The crude product solubilized in a $CHCl_3$/MeOH 1/1 mixture was passed through a mixed ion exchanger resin (Serdolit MB-2). Silica gel chromatography (elution with $CHCl_3$/MeOH from 1/0 to 1/1), afforded 0.35 g (50%) of **IIa** as a white powder.
Anal. Calcd. for $C_{38}H_{60}F_{18}N_3O_6P,3H_2O$: C, 42.19; H, 6.15; N, 3.88 P, 2.86; Found: C, 42.19; H, 6.09; N, 3.84; P, 2.78. ^1H NMR ($CDCl_3/CD_3OD$): δ 1.25 (br s, 24H, $(CH_2)_6$), 1.52 (m, 8H, $C\underline{H}_2CH_2CO$ and $C\underline{H}_2CH_2CF_2$), 2.00 (tt, ^3J = 8.2, $^3J_{HF}$ = 18.0, 4H, CH_2CF_2), 2.15 (t, ^3J = 8.0, 4H, CH_2CO), 3.07 and 3.45 (AB part of an ABX system, $^2J_{AB}$ = 14.0, $^3J_{AX}$ = 6.3, $^3J_{BX}$ = 4.2, 4H, $C\underline{H}_2CHOP$), 3.17 (s, 9H, NMe_3^+), 3.60 (m, 2H, $C\underline{H}_2NMe_3^+$), 4.03 (m, 1H, CH), 4.23 (m, 2H, $POCH_2$). ^{13}C NMR ($CDCl_3/CD_3OD$): δ 19.8 (t, $^3J_{CF}$ = 4, $\underline{C}H_2CH_2CF_2$), 25.6 (s, $\underline{C}H_2CH_2CO$), 28.8, 28.9, 29.0, 29.1 (all s, $(CH_2)_6$), 30.0 (t, $^2J_{CF}$ = 22, $\underline{C}H_2CF_2$), 36.1 (s $\underline{C}H_2CO$), 40.8 (d, $^3J_{CP}$ = 5, $\underline{C}H_2CHOP$), 53.7 (three lines due to $^1J_{CN}$ = 4, NMe_3^+), 59.0 (d, $^2J_{CP}$ = 5, $POCH_2$), 66.5 (m, $\underline{C}H_2NMe_3^+$), 72.5 (d, $^2J_{CP}$ = 6, CHOP), 175.2 (s, CO). ^{19}F NMR ($CDCl_3/CD_3OD$): identical to that of **2a**. ^{31}P {^1H} NMR ($CDCl_3/CD_3OD$): δ -1.6 (s).

N,N'-di-[11-(F-hexyl)-undecanoyl]-1,3-diaminopropan-2-phospho-N,N,N-trimethylethanolamine, **IIb**.
(Procedure E). To a solution of **2b** (1.5 g, 1.4 mmol) and triethylamine (0.50 mL, 3.5 mmol) in 40 mL of THF, was added dropwise, at room temperature, a solution of 2-chloro-2-oxo-1,3,2-dioxaphospholane (0.33 mL, 3.5 mmol) in 15 mL of THF. The mixture was stirred at room temperature for 24-48h. The precipitate of triethylammonium salt was filtered off under nitrogen and washed with THF. The solvent was then evaporated under reduced pressure and the residue was transferred to a dry pressure flask containing 30 mL of acetonitrile and 1.36 mL (7.1 mmol) of TMSOTf. Anhydrous trimethylamine (2.0 g, 34 mmol) was introduced and the bottle was closed and kept at 65°C for 16h. After cooling, 2.5 mL of water were added to the reaction mixture and stirring was continued for 15 min. The solvents were then removed under vacuo. The crude product solubilized in a $CHCl_3$/MeOH 1/1 mixture was passed through a mixed ion exchanger resin (Serdolit MB-2). Silica gel chromatography (elution with $CHCl_3$/MeOH from 1/0 to 6/4), afforded 0.29 g (20%) of **IIb** as a white powder.
Anal. Calcd. for $C_{42}H_{60}F_{26}N_3O_6P,3H_2O$: C, 39.35; H, 5.19; N, 3.28 P, 2.42; Found: C, 39.13; H, 5.01; N, 3.30; P, 2.25. ^1H and ^{13}CNMR ($CDCl_3/CD_3OD$): identical to those of **IIa**. ^{19}F NMR ($CDCl_3/CD_3OD$): identical to that of **2b**. ^{31}P {^1H} NMR ($CDCl_3/CD_3OD$): δ -1.9 (s).

N,N'-di-[11-(F-hexyl)-undecanoyl]-2,3-diaminopropan-1-phospho-N,N,N-trimethylethanolamine, **Ib**.
Procedure E, when applied to alcohol **5** (0.5 g, 0.5 mmol), 2-chloro-2-oxo-1,3,2-dioxaphospholane (0.09 mL, 1.0 mmol) and triethylamine (0.13 mL, 1.0 mmol), then to anhydrous trimethylamine (0.3 g), gave, after ion exchange and column chromatography (elution with $CHCl_3$ until $CHCl_3$/MeOH 2/8), 0.12 g (21%) of **Ib** as a white powder.
Anal. Calcd. for $C_{42}H_{60}F_{26}N_3O_6P,3H_2O$: C, 39.35; H,5.19; N, 3.28 P, 2.42; Found: C, 39.00; H, 4.98; N, 3.33; P, 2.19. ^1H NMR ($CDCl_3/CD_3OD$): δ 1.25 (br s, 24H, $(CH_2)_6$), 1.50 (m, 8H, $C\underline{H}_2CH_2CO$ and $C\underline{H}_2CH_2CF_2$), 2.00 (tt, ^3J = 8.5, $^3J_{HF}$ = 19.0, 4H, CH_2CF_2), 2.10 (m, 4H, CH_2CO), 3.00-3.50 (m, 2H,

CH_2N), 3.15 (s, 9H, NMe_3^+), 3.60 (m, 2H, $C\underline{H}_2NMe_3$), 3.70-4.00 (m, 3H, $CHC\underline{H}_2OP$ and CHN), 4.20 (m, 2H, $POCH_2$), 7.65 (t, ^3J = 7.0, 1H, $N\underline{H}CH_2$), 7.75 (d, ^3J = 7.0, 1H, $N\underline{H}CH$). ^{13}C NMR ($CDCl_3/CD_3OD$): δ 20.0 (t, $^3J_{CF}$ = 4, $\underline{C}H_2CH_2CF_2$), 25.6 and 25.7 (s, s, $\underline{C}H_2CH_2CO$), 29.0, 29.1, 29.2, 29.3 (all s, $(CH_2)_6$), 30.8 (t, $^2J_{CF}$ = 22, $\underline{C}H_2CF_2$), 36.3 and 36.4 (s, s, $\underline{C}H_2CO$), 40.3 (s, CH_2N), 50.5 (d, $^3J_{CP}$ = 7, $\underline{C}HCH_2OP$), 54.3 (three lines due to $^1J_{CN}$ = 4, NMe_3^+), 59.0 (d, $^2J_{CP}$ = 5, $POCH_2$), 63.8 (d, $^2J_{CP}$ = 5, $CH\underline{C}H_2OP$), 66.5 (m, $\underline{C}H_2NMe_3$), 174.7 and 175.6 (s, s, CONH). ^{19}F NMR ($CDCl_3/CD_3OD$): identical to that of **2b**. ^{31}P {^1H} NMR ($CDCl_3/CD_3OD$): δ 0.2 (s).

N-[11-F-hexyl)-undecanoyl]-O-phospho-N,N,N-trimethylethanolamine-L-serine hexadecylamide, **IIIa**.
Procedure E, when applied to alcohol **9a** (0.50 g, 0.61 mmol), 2-chloro-2-oxo-1,3,2-dioxaphospholane (0.11 mL, 1.2 mmol) and triethylamine (0.17 mL, 1.2 mmol), then to anhydrous trimethylamine (1.9 g), gave, after ion exchange and column chromatography (elution with $CHCl_3$ until $CHCl_3$/MeOH 3/7), 0.25 g (42%) of **IIIa** as a white powder.
Anal. Calcd. for $C_{41}H_{71}F_{13}N_3O_6P,3H_2O$: C, 47.62; H, 7.50; N, 4.06; P, 2.99; Found: C, 47.65; H, 7.41; N, 4.06; P, 2.91. ^1H NMR ($CDCl_3/CD_3OD$): δ 0.85 (t, 3H, CH_3), 1.10-1.70 (m, 44H, $(CH_2)_8$ and $(CH_2)_{14}$), 2.05 (tt, $^3J_{HF}$ = 19.0, ^3J = 8.5, 2H, CH_2CF_2), 2.25 (t, ^3J = 6.7, 2H, CH_2CO), 3.15 (m, 2H, CH_2N), 3.20 (s, 9H, NMe_3^+), 3.65 (m, 2H, $CH_2NMe_3^+$), 3.85-4.20 (m, 2H, $CHC\underline{H}_2OP$), 4.25 (m, 2H, $POCH_2$), 4.55 (m, 1H, CH). ^{13}C NMR ($CDCl_3/CD_3OD$): δ 14.0 (s, CH_3), 20.1 (t, $^3J_{CF}$ = 4, $\underline{C}H_2CH_2CF_2$), 22.6 (s, $\underline{C}H_2CH_3$), 25.6 (s, $\underline{C}H_2CH_2CO$), 26.9 (s, $\underline{C}H_2CH_2N$), 29.1, 29.2, 29.3, 29.4, 29.6, 29.7 (all s, $(CH_2)_{11}$ and $(CH_2)_6$), 30.9 (t, $^2J_{CF}$ = 22, $\underline{C}H_2CF_2$), 31.9 (s, $\underline{C}H_2(CH_2)_2N$), 36.2 (s, $\underline{C}H_2CO$), 39.6 (s, CH_2N), 53.8 (d, $^3J_{CP}$ = 6, $\underline{C}HCH_2OP$), 54.4 (m, NMe_3^+), 59.1 (d, $^2J_{CP}$ = 5, $POCH_2$), 64.8 (d, $^2J_{CP}$ = 7, $CH\underline{C}H_2OP$), 66.6 (m,$\underline{C}H_2NMe_3^+$), 169.8 (s, $CH_2\underline{C}ONH$), 174.3 (s, $CH\underline{C}ONH$). ^{19}F NMR ($CDCl_3/CD_3OD$): identical to that of **2b**. ^{31}P {^1H} NMR ($CDCl_3/CD_3OD$): δ 0.2 (s).

N-[11-(F-butyl)-undecanoyl]-O-phospho-N,N,N-trimethylethanolamine-L-serine 11-(F-butyl)-undecylamide, **IIIb.** Procedure E, when applied to alcohol **9b** (1.6 g, 1.9 mmol), 2-chloro-2-oxo-1,3,2-dioxaphospholane (0.34 mL, 3.7 mmol) and triethylamine (0.52 mL, 3.7 mmol), then to anhydrous trimethylamine (4.0 g), gave, after ion exchange and column chromatography (elution with $CHCl_3$ until $CHCl_3$/MeOH 3/7), 1.2 g (60%) of **IIIb** as a white powder.
Anal. Calcd. for $C_{38}H_{60}F_{18}N_3O_6P,2H_2O$: C, 42.90; H, 6.06; N, 3.95; P, 2.91; Found: C, 42.68; H, 6.15; N, 3.96; P, 2.87. ^1H NMR ($CDCl_3/CD_3OD$): 1.20-1.70 (m, 34H, $(CH_2)_8$ and $(CH_2)_9$), 2.00 (tt, $^3J_{HF}$ = 19.0, ^3J = 7.6, 4H, CH_2CF_2), 2.20 (t, ^3J = 7.2, 2H, CH_2CO), 3.10 (m, 2H, CH_2N), 3.25 (s, 9H, NMe_3^+), 3.60 (m, 2H, $C\underline{H}_2NMe_3^+$), 3.80-4.20 (m, 2H, $CHC\underline{H}_2OP$), 4.25 (m, 2H, $POCH_2$), 4.50 (t, ^3J = 4.6, 1H, CHN). ^{13}C NMR ($CDCl_3/CD_3OD$): 20.0 (t, $^3J_{CF}$ = 4, $\underline{C}H_2CH_2CF_2$), 25.6 (s, $\underline{C}H_2CH_2CO$), 26.9 (s, $\underline{C}H_2CH_2N$), 29.0, 29.2, 29.3, 29.4, 29.5 (all s, $(CH_2)_6$ and $(CH_2)_7$), 30.7 (t, $^2J_{CF}$ = 22, $\underline{C}H_2CF_2$), 36.2 (s, $\underline{C}H_2CO$), 39.6 and 39.7 (s, s, CH_2N), 53.8 (d, $^3J_{CP}$ = 6, $\underline{C}HCH_2OP$), 54.3 (br s, NMe_3^+), 59.2 (d, $^2J_{CP}$ = 5, $POCH_2$), 64.8 (d, $^2J_{CP}$ = 5, $CH\underline{C}H_2OP$), 66.5 (d, $^2J_{CP}$ = 5, $\underline{C}H_2NMe_3^+$), 169.9 and 170.0 (s, s, $CH_2\underline{C}ONH$), 174.3 and 174.4 (s, s, $CH\underline{C}ONH$). ^{19}F NMR identical to that of **2a**. ^{31}P {^1H} NMR ($CDCl_3/CD_3OD$): -0.02 (s).

N-[11-(F-hexyl)-undecanoyl]-O-phospho-N,N,N-trimethylethanolamine-L-serine 11-(F-butyl)-undecylamide, **IIIc**. Procedure E, when applied to alcohol **9c** (0.96 g, 1.0 mmol), 2-chloro-2-oxo-1,3,2-dioxaphospholane (0.18 mL, 2.0 mmol) and triethylamine (0.28 mL, 2.0 mmol), then to anhydrous trimethylamine (2.3 g, mmol),

gave, after ion exchange and column chromatography (elution with $CHCl_3$ until $CHCl_3$/MeOH 3/7), 0.68 g (61%) of **IIIc** as a white powder.

Anal. Calcd. for $C_{40}H_{60}F_{22}N_3O_6P,3H_2O$: C, 40.65; H, 5.63; N, 3.56; P, 2.62; Found: C, 41.17; H, 5.34; N, 3.58; P, 2.59. ^1H and ^{13}C NMR ($CDCl_3/CD_3OD$): identical to that of **IIIb** apart the triplet at 30.7 in the ^{13}C NMR spectrum which is replaced by two triplets at, respectively, 30.7 ($^2J_{CF}$ = 22) and 30.8 ($^2J_{CF}$ = 22) for the two $\underline{C}H_2CF_2$. ^{19}F NMR identical to that of **8c**. ^{31}P {^1H} NMR ($CDCl_3/CD_3OD$): -0.36 (s).

N-[11-(F-hexyl)-undecanoyl]-O-phospho-N,N,N-trimethylethanolamine-L-serine 11-(F-hexyl)-undecylamide, **IIId**. Procedure E, when applied to alcohol **9d** (1.1 g, 1.0 mmol), 2-chloro-2-oxo-1,3,2-dioxaphospholane (0.19 mL, 2.1 mmol) and triethylamine (0.29 mL, 2.1 mmol), then to anhydrous trimethylamine (2.5 g), gave, after ion exchange and column chromatography (elution with $CHCl_3$ until $CHCl_3$/MeOH 2/8), 0.64 g (50%) of **IIId** as a white powder.

Anal. Calcd. for $C_{42}H_{60}F_{26}N_3O_6P,H_2O$: C, 40.50; H, 5.02; N, 3.37; P, 2.49; Found: C, 40.81; H, 5.22; N, 3.57; P, 2.42. ^1H and ^{13}C NMR ($CDCl_3/CD_3OD$): identical to those of **IIIb**. ^{19}F NMR ($CDCl_3/CD_3OD$) identical to that of **2b**. ^{31}P {^1H} NMR ($CDCl_3/CD_3OD$): δ 0.2 (s).

Biological tests

The biological tests were performed on heat-sterilized (121°C, 15 min) liposomal dispersions of the phospholipid to be tested in a phosphate-buffered saline (Biomérieux). These dispersions (average particle size of ~ 100 nm measured by light scattering spectroscopy using a Coulter Model N4 MD sub-micron particle analyzer) were prepared according to the general procedure described in reference 8a,b. The in vivo test consisted of injecting 500 µL of the isotonic heat-sterilized liposomal dispersion into the tail vein of 10 Dawley mice of 20 g weight at a dose of 1050 and 2590 mg/kg body weight of compound **IIa** and **IIIb**, respectively. Growth and any symptoms of intoxication of the animals were monitored over a period of 21 days and compared with those of a control group. All the animals displayed regular growth and no death nor symptoms were recorded over this observation period.

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