Phenytoin-Lipid Conjugates as Potential Prodrugs of Phenytoin

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Phenytoin-1-triglycerides and phenytoin-2-triglycerides were synthesized as potential prodrugs of phenytoin by covalent binding of 3-hydroxymethylphenytoin by succinic acid to the positions 1 and 2 of diglycerides, respectively. The corresponding 1- and 2-monoglycerides were prepared. In addition, replacement of glycerol by 3-hydroxy-2-hydroxymethylpropionic acid furnished lipids that allowed direct coupling of 3-hydroxymethylphenytoin. The lipid conjugates proved to be substrates for pancreatic lipase *in vitro*.

Phenytoin-Lipid-Konjugate als potentielle Prodrugs des Phenytoins

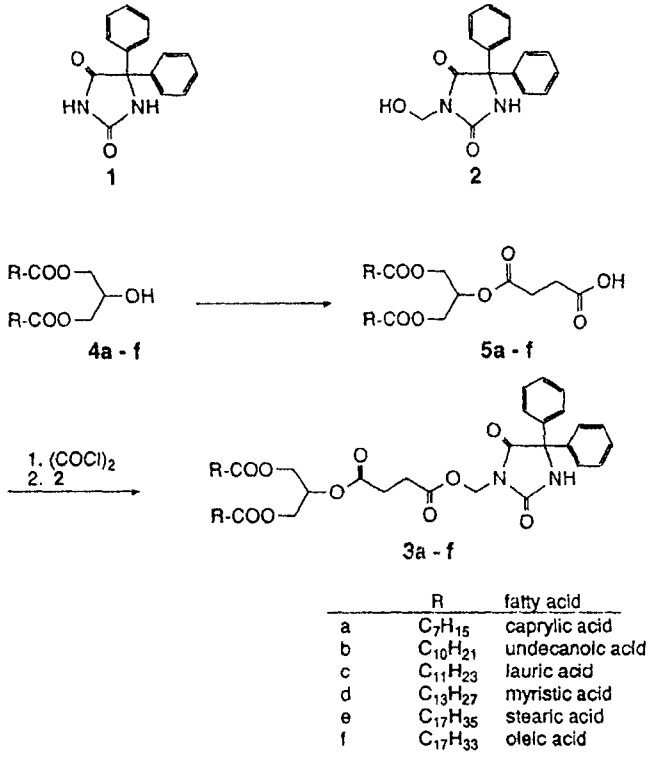
Als potentielle Prodrugs des Phenytoins wurden Phenytoin-1-triglyceride und Phenytoin-2-triglyceride durch kovalente Bindung von 3-Hydroxymethylphenytoin mittels Bernsteinsäure an die Positionen 1 bzw. 2 von Diglyceriden synthetisiert. Die entspr. 1- bzw. 2-Monoglyceride wurden hergestellt. Der Austausch von Glycerin gegen 3-Hydroxy-2-hydroxymethylpropionsäure führte zu Lipiden, die eine direkte Esterbindung mit 3-Hydroxymethylphenytoin erlaubten. Pankreaslipase hydrolysierte *in vitro* die Lipid-Konjugate in Analogie zu natürlichen Triglyceriden.

Mixed triglycerides formed by coupling of drugs to diglycerides exhibit physicochemical properties[1,2] and absorption characteristics[3,4] similar to those of natural triglycerides. Drugs which lack carboxyl groups allowing the direct formation of an ester bond with a diglyceride have to be bound to the glycerides by spacers such as succinic acid. Phenytoin (**1**) has shown erratic bioavailability when administered orally[5]. Superior availability was obtained by co-administration of lipids[6] or by ester prodrugs of 3-hydroxymethylphenytoin (**2**)[7,8]. At physiological pH, **2** undergoes fast and spontaneous decomposition to give **1**[8].

The present study describes the synthesis of "phenytoin-triglycerides" and "phenytoin-monoglycerides" in which **2** has been attached by succinic acid to the position 1 or 2 of glycerides and glycerol, respectively. Moreover, 3-acyloxy-2-acyloxymethylpropionic acids were designed as "glyceride mimics" allowing direct esterification of **2**.

The phenytoin-2-triglycerides **3a-f** were obtained by reaction of 1,3-diacylglycerols **4a-f**[9] with succinic anhydride to yield the monoesters **5a-f** followed by condensation with **2** (Scheme 1).

The synthesis of the monoglycerides **6** and **7** (Scheme 2) required special precautions during the removal of the protecting groups and further purification steps in order to avoid the easily occurring intramolecular acyl migration of 1- and 2-monoglycerides[10]. The isomers can be differentiated by their NMR spectra[10]. Treatment of benzylidene glycerol (**8**)[11] with succinic anhydride yielded the monoester **9**. Bis(2-oxo-3-oxazolidinyl)phosphinic acid chloride(BOP-Cl)-mediated esterification with **2** afforded **10**. Cleavage of the benzylidene acetal with H_3BO_3 in triethyl borate[12] and chromatographic purification on H_3BO_3-treated silica gel[13], both methods known to avoid acyl migration in monoglycerides, gave the 2-monoglyceride **6**. NMR analysis did not reveal an impurity of the 1-isomer in freshly prepared samples of **6**. However, **6** proved to be unstable. Even at +4°C considerable isomerization occurred. Thus, about 10% of the 1-isomer **7** was detected in samples of **6** by NMR spectroscopy after 4 weeks at +4°C. Pure samples of **7** were synthesized starting with the hemisuccinate **11**



	R	fatty acid
a	C_7H_{15}	caprylic acid
b	$C_{10}H_{21}$	undecanoic acid
c	$C_{11}H_{23}$	lauric acid
d	$C_{13}H_{27}$	myristic acid
e	$C_{17}H_{35}$	stearic acid
f	$C_{17}H_{33}$	oleic acid

Scheme 1

followed by esterification with **2** to yield **12**. H_3BO_3-catalyzed cleavage of the protecting group and chromatographic purification on H_3BO_3-treated silica gel afforded **7**. In contrast to compound **6**, the 1-monoglyceride **7** was stable for at least 6 months at +4°C. Esterification with myristic acid chloride yielded the phenytoin-1-triglyceride **13**.

The starting material of the synthesis of the "inverse" lipids **14a-f** was the dioxane derivative **15** (Scheme 3). Saponification and decarboxylation yielded 2-phenyl-1,3-dioxan-

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5-yl-carboxylic acid (**16**). *trans* Conformation of **16** with the carboxyl- and phenyl-group in equatorial positions of the dioxane ring can be derived from comparison of the NMR spectrum of **16** with lit. data of other 2,5-substituted dioxane and cyclohexane derivatives[13,14]. Subsequent formation of the ester bond to give **17** was mediated by BOP-Cl. Pd-catalyzed hydrogenolysis afforded the 3-hydroxy-2-hydroxymethylpropionic acid ester **18** which was converted to the lipids **14a-f** by treatment with the respective fatty acid chlorides.

Preliminary results showed that the lipid prodrugs are hydrolyzed by pancreatic lipase with the same positional specificity as observed for natural triglycerides[15]. Thus, the 2-monoglyceride **6** and the dihydroxypropionic acid ester **18** were the primary products of the lipase-mediated hydrolysis of the prodrugs **3d** and **14d**, respectively, whereas 3-hydroxymethylphenytoin succinic acid mono ester[8] arising from the cleavage of the ester bond between succinic acid and glycerol in position 1 was found in incubations of the phenytoin-1-triglyceride **13**. Lipid **3d** was the best substrate for the enzyme. From time-dependent hydrolysis plots monitoring the deacyl derivatives **6**, **7**, **18** and the 3-hydroxymethylphenytoin succinic acid mono ester half-lives of 1.1, 5.7, and 9.8 min were estimated for **3d**, **13**, and **14d**, respectively, under the conditions applied. Further degradation of the primary products of the lipase-catalyzed hydrolysis eventually gave phenytoin (**1**) in quantitative yield.

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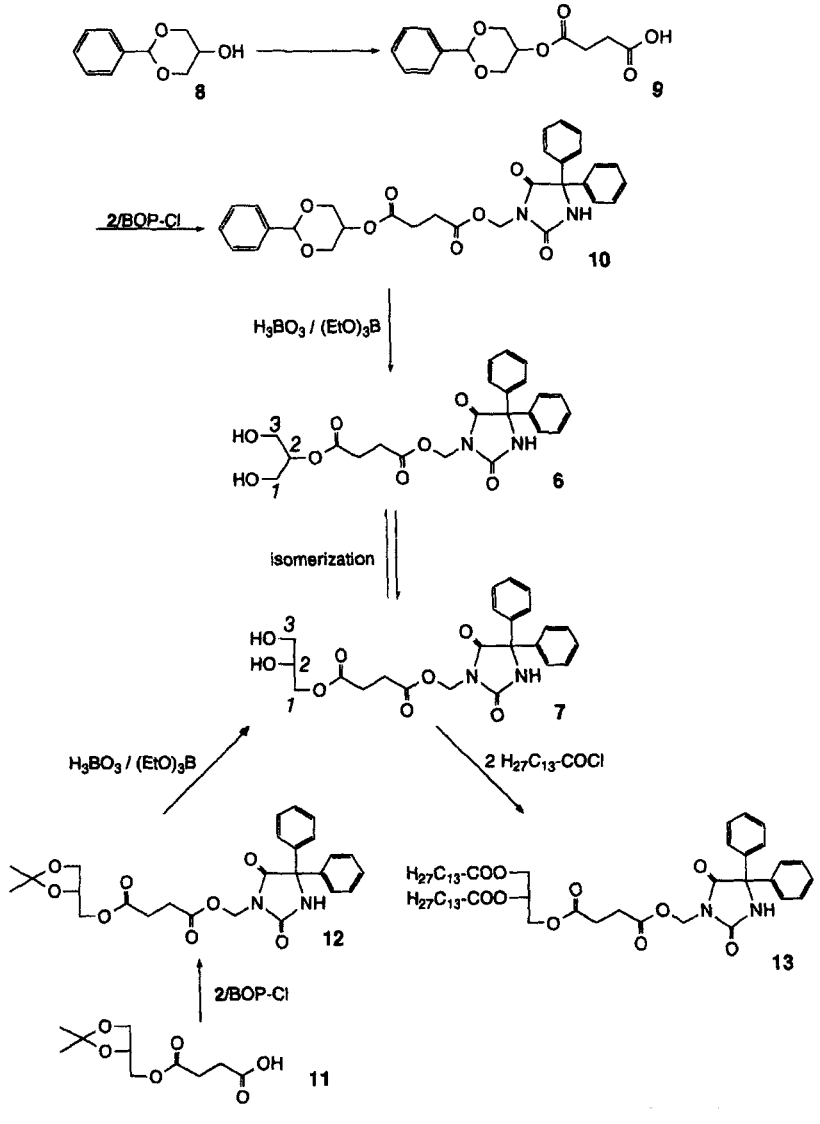
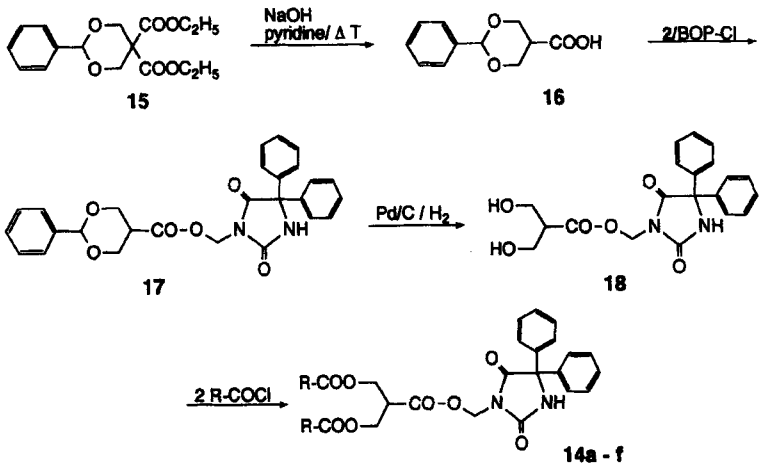
Experimental Part

Mp.: Kofler melting point apparatus, uncorrected.- NMR: Varian Gemini 200 (TMS), ^{1}H: 200 MHz, ^{13}C: 50 MHz.- MS: Varian MAT 44S, EI: 70 eV, source temp. 200°C; DCI: source temp. 100°C, ammonia.- Column



Scheme 2



Scheme 3

chromatography: Silica gel Si-60, 0.063-0.200 mm (70-230 mesh) (Merck).- 3-Hydroxymethylphenytoin (**2**)[8] and 1,3-benzylidene glycerol (**8**)[10] were synthesized as described. 1,3-diacylglycerols (**4a-f**) were prepared according to *Bentley* and *McCrae*[9].

2-(1,3-Diacylglycerol)succinic acid mono esters (**5a-f**)

10 mmol 1,3-diacylglycerol **4a-f**, 11 mmol pyridine, 0.5 mmol 4-dimethylaminopyridine (DMAP), and 15 mmol succinic anhydride were refluxed for 15 h in CH_2Cl_2. The org. phase was washed with 0.1 N HCl and water and evaporated. The waxy solids were crystallized from ethanol. **5a**: mp. 34-35°C, yield 3.8 g (85%).- **5b**: mp. 51-53°C, yield 3.4 g (64%).- **5c**: mp. 62-63°C, yield 4.4 g (79%).- **5d**: mp. 65-66°C, yield 5.0 g (82%).- **5e**: mp. 69-71°C, yield 5.3 g (74%).- **5f**: oil, yield 4.1 g (57%).

2-(1,3-Diacylglycerol)-3-[(2,4-dioxo-5,5-diphenyl-3-imidazolidinyl)-methoxycarbonyl]propionic acid ester (**3a-f**)

5 mmol **5a-f** were converted into the pertinent acid chloride by treatment with oxalyl chloride. The resulting oil was added at 0°C to a mixture of 1.3 g (4.6 mmol) **2** and 0.8 g (10 mmol) pyridine in THF. Stirring was continued for 2 h. The mixture was poured into a mixture of ice and N HCl and extracted with CH_2Cl_2. The org. phase was washed with water, dried (Na_2SO_4) and evaporated i.vac. Column chromatography (CH_2Cl_2/methanol 98:2) afforded waxy solids or oils.

3a: Oil, yield 2.7 g (77%).- $C_{39}H_{52}N_2O_{10}$ (708.8) Calcd. C 66.1 H 7.40 N 4.0 Found C 66.4 H 7.52 N 4.0.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 8.11 (s; 1H, NH), 7.36 (m; 10 H, Ar), 5.55 (s; 2H, O-CH_2-N), 5.23 (m; 1H, CH), 4.27 and 4.12 (dd each; J = 4.3/12.0 Hz and J = 5.8/12.0 Hz, 2H each, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.61 (s; 4H, CO-CH_2-CH_2-CO), 2.30 (t; J = 7.4 Hz, 4H, 2 x CO-CH_2), 1.56 (m; 4H, 2 x CO-CH_2-C$\underline{H}_2$), 1.27 (m; 16 H, fatty acid CH_2), 0.87 (t; J = 6.2 Hz, 6H, 2 x CH_3).- DCI-MS m/z (%) = 726 (100, [M + NH_4]$^+$).

3b: Mp. 34-36°C, yield 2.6 g (67%).- $C_{45}H_{64}N_2O_{10}$ (793.0) Calcd. C 68.2 H 8.14 N 3.5 Found C 68.5 H 8.41 N 3.2.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.59 (s; 1H, NH), 7.38 (m; 10 H, Ar), 5.59 (s; 2H, O-CH_2-N), 5.23 (m; 1H, CH), 4.27 and 4.12 (dd each; J = 4.4/12.0 Hz and J = 5.8/12.0 Hz, 2H each, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.62 (s; 4H, CO-CH_2-CH_2-CO), 2.30 (t; J = 7.3 Hz, 4H, 2 x CO-CH_2), 1.59 (m; 4H, 2 x CO-CH_2-C$\underline{H}_2$), 1.26 (m; 28 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 811 (100, [M + NH_4]$^+$).

3c: Mp. 46-48°C, yield 3.3 g (80%).- $C_{47}H_{68}N_2O_{10}$ (821.0) Calcd. C 68.7 H 8.35 N 3.4 Found C 68.3 H 8.51 N 3.4.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.68 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.58 (s; 2H, O-CH_2-N), 5.22 (m; 1H, CH), 4.26 and 4.12 (dd each; J = 4.3/11.9 Hz and J = 5.8/11.9 Hz, 2H each, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.63 (s; 4H, CO-CH_2-CH_2-CO), 2.30 (t; J = 7.3 Hz, 4H, 2 x CO-CH_2), 1.59 (m; 4H, 2 x CO-CH_2-C$\underline{H}_2$), 1.26 (m; 32 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 839 (100, [M + NH_4]$^+$).

3d: Mp. 52-54°C, yield 3.2 g (73%).- $C_{51}H_{76}N_2O_{10}$ (877.1) Calcd. C 69.8 H 8.73 N 3.2 Found C 70.0 H 8.99 N 3.1.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.87 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.57 (s; 2H, O-CH_2-N), 5.22 (m; 1H, CH), 4.26 and 4.12 (dd each; J = 4.3/11.9 Hz and J = 5.8/11.9 Hz, 2H each, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.63 (s; 4H, CO-CH_2-CH_2-CO), 2.30 (t; J = 7.3 Hz, 4H, 2 x CO-CH_2), 1.59 (m; 4H, 2 x CO-CH_2-C$\underline{H}_2$), 1.26 (m; 40 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 894 (100, [M + NH_4]$^+$).

3e: Mp. 44-45°C, yield 3.2 g (65%).- $C_{59}H_{92}N_2O_{10}$ (989.3) Calcd. C 71.6 H 9.37 N 2.8 Found C 71.8 H 9.60 N 3.0.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.52 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.58 (s; 2H, O-CH_2-N), 5.22 (m; 1H, CH), 4.26 and 4.11 (dd each; J = 4.5/12.0 Hz and J = 5.8/12.0 Hz, 2H each, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.62 (s; 4H, CO-CH_2-CH_2-CO), 2.30 (t; J = 7.3 Hz, 4H, 2 x CO-CH_2), 1.59 (m; 4H, 2 x CO-CH_2-C$\underline{H}_2$), 1.26 (m; 56 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 1007 (100, [M + NH_4]$^+$).

3f: Oil, yield 2.8 g (63%).- $C_{59}H_{88}N_2O_{10}$ (985.3) Calcd. C 71.9 H 9.00 N 2.8 Found C 72.5 H 9.22 N 2.6.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.87 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.57 (s; 2H, O-CH_2-N), 5.35 (m; 4H, 2 x CH=CH), 5.22 (m; 1H, CH), 4.26 and 4.11 (dd each; J = 4.3/12.0 Hz and J = 5.7/12.0 Hz, 2H each, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.62 (s; 4H, CO-CH_2-CH_2-CO), 2.30 (t; J = 7.3 Hz, 4H, 2 x CO-CH_2), 1.98 (m; 8H, 2 x C$\underline{H}_2$-CH=CH-C$\underline{H}_2$), 1.59 (m; 4H, 2 x CO-CH_2-C$\underline{H}_2$), 1.26 (m; 40 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 1003 (100, [M + NH_4]$^+$).

2-(1,3-Benzylidene glycerol)succinic acid mono ester (**9**)

12.5 g (70 mmol) 1,3-benzylidene glycerol (**8**)[10], 9 g (90 mmol) succinic anhydride, 9 g (90 mmol) triethylamine and 1 g (8 mmol) DMAP were refluxed in 100 ml CH_2Cl_2 for 15 h. The org. phase was washed with ice-cold 0.1 N HCl and water, dried (Na_2SO_4) and evaporated. Colorless crystals, mp. 118-119°C (diethylether/n-hexane), yield 14.1 g (72%).- $C_{14}H_{16}O_6$ (280.3) Calcd. C 60.0 H 5.75 Found C 60.0 H 5.72.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.53-7.33 (m; 5H, Ar), 5.55 (s; 1H, Ph-CH), 4.74 (m; 1H, CH-O), 4.21 (m; 4H, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.72 (m; 4H, CO-CH_2-CH_2-CO).- MS: m/z (%) = 280 (9, M$^{+\cdot}$), 279 (8), 162 (7), 158 (9), 140 (89), 107 (51), 105 (82), 101 (100), 91 (22), 79 (28), 77 (37).

2-(1,3-Benzylidene glycerol)-3-[(2,4-dioxo-5,5-diphenyl-3-imidazolidinyl)methoxycarbonyl]propionic acid ester (**10**)

1.4 g (5 mmol) **9**, 1.5 g (5.3 mmol) **2**, 1 g (10 mmol) triethylamine and 1.3 g (5.1 mmol) BOP-Cl were stirred at room temp. for 1 h. The solvent was washed with 10% $NaHCO_3$ and water, dried (Na_2SO_4) and evaporated. Colorless crystals, mp. 73-75°C (diethylether/n-hexane), yield 2.5 g (91%).- $C_{30}H_{28}N_2O_8$ (544.5) Calcd. C 66.2 H 5.18 N 5.1 Found C 66.3 H 5.25 N 5.0.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.42 (m; 15 H, Ar), 5.58 (s; 2H, O-CH_2-N), 5.53 (s; 1H, Ph-CH), 4.67 (m; 1H, CH-O), 4.19 (m; 4H, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.69 (m; 4H, CO-CH_2-CH_2-CO).- DCI-MS: m/z (%) = 562 (100, $[M + NH_4]^+$).

2-Glycerol-3-[(2,4-dioxo-5,5-diphenyl-3-imidazolidinyl)methoxycarbonyl]propionic acid ester (**6**)

2.18 g (4 mmol) **10** were dissolved in 15 ml 1,4-dioxane and 15 ml triethyl borate and 1 g (16 mmol) finely powdered boric acid was added. The mixture was heated at 100°C for 20 min. The solvents were evaporated while heating under vacuum and the heating under vacuum was continued for another 15 min. The cooled mixture was dissolved in ethyl acetate, washed with water, dried (Na_2SO_4) and evaporated. Column chromatography on boric acid-treated silica gel[13] afforded 1.23 g (67%) of a hygroscopic foam which could not be crystallized.- $C_{23}H_{24}N_2O_8$ (456.4) Calcd. C 60.5 H 5.30 N 6.1 Found C 60.3 H 5.51 N 5.9.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.71 (s; 1H, NH), 7.34 (m; 10 H, Ar), 5.53 (s; 2H, O-CH_2-N), 4.91 (m; 1H, CH), 3.78 (m; 4H, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.59 (s; 4H, CO-CH_2-CH_2-CO).- ^{13}C-NMR: δ (ppm) = 172.4 (CO), 171.7 (CO), 171.4 (CO), 154.7 (C-$\underline{C}$O), 138.6 (Ar-C), 128.9, 128.8 and 126.9 (Ar-CH), 75.7 (C-2), 70.5 ($\underline{C}$-CO), 62.2 (O-CH_2-N), 61.8 (C-1 and C-3), 29.3 and 29.1 (CH_2-CH_2).- DCI-MS: m/z (%) = 474 (100, $[M + NH_4]^+$).

rac-1-(2,3-O-Isopropylidene glycerol)succinic acid mono ester (**11**)

6.6 g (50 mmol) *rac*-2,3-*O*-isopropylidene glycerol, 6.5 g (65 mmol) succinic anhydride, 4.3 g (55 mmol) pyridine and 0.3 g (2.5 mmol) DMAP were reacted as described for **5a-f**. Colorless crystals, mp. 59-60°C (diethylether), yield 10.1 g (87%).- $C_{10}H_{16}O_6$ (232.2) Calcd. C 51.7 H 6.94 Found C 51.7 H 7.18.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 10.24 (s; 1H, COOH), 4.33 (m; 1H, CH), 4.39-4.05 and 3.79-3.72 (complex multiplets, 4H, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.68 (s; 4H, CO-CH_2-CH_2-CO), 1.44 (s; 3H, CH_3), 1.37 (s; 3H, CH_3).- MS: m/z (%) = 232 (0.1, $M^{+\cdot}$), 217 (47), 117 (24), 101 (100), 83 (11), 73 (47), 72 (36).

rac-1-(2,3-O-Isopropylidene glycerol)-3-[(2,4-dioxo-5,5-diphenyl-3-imidazolidinyl)methoxycarbonyl]propionic acid ester (**12**)

3.0 g (13 mmol) **11**, 3.7 g (13 mmol) **2**, 2.6 g (26 mmol) triethylamine, and 3.3 g (14 mmol) BOP-Cl were treated as described for **9**. Column chromatography (CH_2Cl_2/Methanol, 99:1) afforded colorless crystals, mp. 123-125°C (diethylether/n-hexane), yield 5.4 g (84%).- $C_{26}H_{28}N_2O_8$ (496.5) Calcd. C 62.9 H 5.68 N 5.6 Found C 62.8 H 5.83 N 5.5.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.79 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.57 (s; 2H, O-CH_2-N), 4.34-4.00 and 3.74-3.67 (complex multiplets, 5H, CH_2-CH-CH_2), 2.63 (s; 4H, CO-CH_2-CH_2-CO), 1.41 (s; 3H, CH_3), 1.35 (s; 3H, CH_3).- DCI-MS: m/z (%) = 514 (100, $[M + NH_4]^+$).

rac-1-Glycerol-3-[(2,4-dioxo-5,5-diphenyl-3-imidazolidinyl)methoxycarbonyl]propionic acid ester (**7**)

5.35 g (11 mmol) **12** were dissolved in 30 ml 1,4-dioxane and 30 ml triethyl borate. 2.75 g (44 mmol) finely powdered H_3BO_3 were added and the mixture was treated as described for **10**. Yield 4.1 g (83%) of a foam that could not be crystallized.- $C_{23}H_{24}N_2O_8$ (456.4) Calcd. C 60.5 H 5.30 N 6.1 Found C 60.9 H 5.42 N 5.9.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 8.04 (s; 1H, NH), 7.33 (m; 10 H, Ar), 5.51 (s; 2H, O-CH_2-N), 4.09-3.79 and 3.61-3.46 (complex multiplets, 5H, CH_2-CH-CH_2), 2.56 (s; 4H, CO-CH_2-CH_2-CO).- ^{13}C-NMR: δ (ppm) = 172.4 (CO), 172.1 (CO), 171.2 (CO), 154.8 (C-$\underline{C}$O), 138.5 (Ar-C), 128.9, 128.8 and 126.8 (Ar-CH), 70.4 ($\underline{C}$-CO), 70.0 (C-2), 65.5 (C-1), 63.3 (C-3), 62.1 (O-CH_2-N), 28.9 (CH_2-CH_2).- DCI-MS: m/z (%) = 474 (100, $[M + NH_4]^+$).

rac-1-(2,3-Dimyristoylglycerol)-3-[(2,4-dioxo-5,5-diphenyl-3-imidazolidinyl)-methoxycarbonyl]propionic acid ester (**13**)

1.8 g (4 mmol) **7** were esterified with 2 g (8.2 mmol) myristic acid chloride. Column chromatography (CH_2Cl_2/methanol, 99:1) yielded 2.6 g (74%) of a waxy solid, mp. 36-37°C.- $C_{51}H_{76}N_2O_{10}$ (877.1) Calcd. C 69.8 H 8.73 N 3.2 Found C 70.0 H 8.84 N 3.1.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.96 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.52 (s; 2H, O-CH_2-N), 5.22 (m; 1H, CH), 4.20 (m; 4H, C$\underline{H}_2$-CH-C$\underline{H}_2$), 2.61 (s; 4H, CO-CH_2-CH_2-CO), 2.31 (t; J = 7.4 Hz, 2H, CH_2-CO), 2.29 (t; J = 7.5 Hz, CH_2-CO), 1.59 (m; 4H, 2 x C$\underline{H}_2$-CH_2-CO), 1.26 (m; 56 H, fatty acid CH_2), 0.88 (t; J = 6.6 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 895 (100, $[M + NH_4]^+$).

5,5-Dicarboxyethyl-2-phenyl-1,3-dioxane (**15**)

22 g (0.1 mol) bis-(hydroxymethyl)malonic acid ethyl ester, 12 g (0.11 mol) benzaldehyde and 0.2 g *p*-toluene sulfonic acid were refluxed in 250 ml toluene using a *Dean-Stark* trap. The cooled mixture was washed with saturated $NaHCO_3$ and water. Evaporation of the solvent yielded an oil which was purified by column chromatography (diethylether/n-hexane 9:1). Oil, yield 23.4 g (76%).- $C_{16}H_{20}O_6$ (308.3) Calcd. C 62.3 H 6.54 Found C 62.3 H 6.47.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.38 (m; 5H, Ar), 5.47 (s; 1H, Ph-CH), 4.85 (d; J = 11.5 Hz, 2H, H-4_{eq} and H-6_{eq}), 4.32 (q; J = 7.2 Hz, 2H, C$\underline{H}_2$-CH_3), 4.18 (q; J = 7.2 Hz, 2H, C$\underline{H}_2$-CH_3), 4.15 (d; J = 11.5 Hz, 2H, H-4_{ax} and H-6_{ax}), 1.30 (t; J = 7.2 Hz, CH_3), 1.24 (t; J = 7.2 Hz, CH_3).- MS: m/z (%) = 308 (3, $M^{+\cdot}$), 307 (13), 279 (11), 263 (8), 235 (7), 186 (11), 173 (56), 127 (65), 105 (100), 99 (23), 77 (20).

2-Phenyl-1,3-dioxane-5-yl-carbonic acid (**16**)

23 g (74.5 mmol) **15** and 8.5 g KOH (0.15 mol) were refluxed in 300 ml ethanol for 2 h. After removal of the solvent the residue was refluxed in 200 ml pyridine for 3 h until all gas evolution had settled. Pyridine was evaporated and the residue dissolved in ethyl acetate. The org. phase was washed with ice-cold N HCl and water, dried (Na_2SO_4) and evaporated. Colorless needles, mp. 165-166°C (diethylether), yield 14.6 g (94%).- $C_{11}H_{12}O_4$ (208.2) Calcd. C 63.5 H 5.81 Found C 63.6 H 5.85.- ^{1}H-NMR ([D_6]acetone): δ (ppm) = 7.40 (m; 5H, Ar), 5.50 (s; 1H, Ph-CH), 4.41 (m; 2H, H-4_{eq} and H-6_{eq}), 4.01 (m; 2H, H-4_{ax} and H-6_{ax}), 3.08 (m; 1H, CH-CO).- MS: m/z (%) = 208 (11, $M^{+\cdot}$), 207 (32), 131 (13), 123 (22), 106 (39), 105 (100), 85 (19), 78 (29), 77 (72).

2-Phenyl-1,3-dioxane-5-yl-[(2,4-dioxo-5,5-diphenyl-3-imidazolidinyl)-methoxy]-carbonic acid ester (**17**)

14 g (67 mmol) **16**, 19.8 g (70 mmol) **2**, 14 g (0.14 mol) triethylamine, and 17.8 g (70 mmol) BOP-Cl in 100 ml CH_2Cl_2 were treated as described for **9**. Column chromatography (CH_2Cl_2/methanol 98:2) yielded 28.8 g (91%) colorless crystals, mp. 80-81°C (CH_2Cl_2/*n*-hexane).- $C_{27}H_{24}N_2O_6$ (472.5) Calcd. C 68.6 H 5.12 N 5.9 Found C 68.9 H 5.20 N 5.9.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.74 (s; 1H, NH), 7.41 (m; 15 H, Ar), 5.58 (s; 2H, O-CH_2-N), 5.37 (s; 1H, Ph-CH), 4.37 (m; 2H, H-4_{eq} and H-6_{eq}), 3.92 (m; 2H, H-4_{ax} and H-6_{ax}), 3.11 (m; 1H, CH-CO).- MS: m/z (%) = 472 (15, $M^{+\cdot}$), 471 (9), 265 (10), 237 (21), 209 (21), 208 (31), 207 (100), 194 (14), 180 (38), 165 (12), 105 (59), 104 (31), 91 (21), 77 (23).

3-Hydroxy-2-hydroxymethyl-[(2,4-dioxo-5,5-diphenyl-3-imidazolidinyl)-methoxy]-propionic acid ester (**18**)

28.4 g (60 mmol) **17** were hydrogenated in 200 ml THF over 4 g Pd/C (10%). Colorless crystals, mp. 115-116°C (toluene/isopropanol), yield 17.8 g (77%).- $C_{20}H_{20}N_2O_6$ (384.4) Calcd. C 62.5 H 5.24 N 7.3 Found C 62.6 H 5.34 N 7.2.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 8.01 (s; 1H, NH), 7.30 (m; 10 H, Ar), 5.48 (s; 2H, O-CH_2-N), 3.76 (m; 4H, $C\underline{H}_2$-CH-$C\underline{H}_2$), 3.58 (m; 2H, 2 x OH), 2.62 (m; 1H, CH).- MS: m/z (%) = 384 (0.5, $M^{+\cdot}$), 354 (15), 266 (12), 265 (12), 252 (26), 236 (18), 223 (21), 209 (49), 208 (63), 194 (16), 181 (30), 180 (100), 165 (24), 104 (42), 91 (16), 77 (29).

3-Acyloxy-2-acyloxymethyl-[(2,4-dioxo-5,5-diphenyl-3-imidazolidinyl)-methoxy]-propionic acid esters (**14a-f**)

1.15 g (3 mmol) **18** in 15 ml THF were reacted with 6.3 mmol of the respective fatty acid chloride in the presence of 6 mmol pyridine. Work-up and column chromatography (diethylether/n-hexane 1:1) yielded oils or waxy solids.

14a: Mp. 35-37°C, yield 1.3 g (68%).- $C_{36}H_{48}N_2O_8$ (636.8) Calcd. C 67.9 H 7.60 N 4.4 Found C 67.5 H 7.55 N 4.4.- ^{1}H-NMR: ($CDCl_3$): δ (ppm) = 7.83 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.61 (s; 2H, O-CH_2-N), 4.32 and 4.25 (dd each; J = 6.1/11.2 Hz, 2H each, $C\underline{H}_2$-CH-$C\underline{H}_2$), 3.03 (m; 1H, CH), 2.22 (t; J = 7.4 Hz, 4H, 2 x CH_2-CO), 1.54 (m; 4H, 2 x $C\underline{H}_2$-CH_2-CO), 1.25 (m; 16 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 654 (100, $[M + NH_4]^+$).

14b: Oil, yield 1.7 g (79%).- $C_{42}H_{60}N_2O_8$ (720.9) Calcd. C 70.0 H 8.39 N 3.9 Found C 70.2 H 8.25 N 3.8.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.76 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.62 (s; 2H, O-CH_2-N), 4.33 and 4.25 (dd each; J = 6.1/11.3 Hz, 2H each, $C\underline{H}_2$-CH-$C\underline{H}_2$), 3.04 (m; 1H, CH), 2.23 (t; J = 7.3 Hz, 4H, 2 x CH_2-CO), 1.55 (m; 4H, 2 x $C\underline{H}_2$-CH_2-CO), 1.25 (m; 28 H, fatty acid CH_2), 0.88 (t; J = 6.5 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 739 (100, $[M + NH_4]^+$).

14c: Mp. 34-37°C, yield 1.8 g (81%).- $C_{44}H_{64}N_2O_8$ (749.0) Calcd. C 70.6 H 8.61 N 3.7 Found C 71.0 H 8.83 N 3.6.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.95 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.61 (s; 2H, O-CH_2-N), 4.32 and 4.25 (dd each; J = 6.0/11.3 Hz, 2H each, $C\underline{H}_2$-CH-$C\underline{H}_2$), 3.03 (m; 1H, CH), 2.22 (t; J = 7.5 Hz, 4H, 2 x CH_2-CO), 1.54 (m; 4H, 2 x $C\underline{H}_2$-CH_2-CO), 1.25 (m; 32 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 767 (100, $[M + NH_4]^+$).

14d: Mp. 38-41°C, yield 1.7 g (70%).- $C_{48}H_{72}N_2O_8$ (805.1) Calcd. C 71.6 H 9.02 N 3.5 Found C 71.6 H 9.29 N 3.4.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.72 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.62 (s; 2H, O-CH_2-N), 4.32 and 4.25 (dd each; J = 6.0/11.3 Hz, 2H each, $C\underline{H}_2$-CH-$C\underline{H}_2$), 3.03 (m; 1H, CH), 2.22 (t; J = 7.4 Hz, 4H, 2 x CH_2-CO), 1.54 (m; 4H, 2 x $C\underline{H}_2$-CH_2-CO), 1.25 (m; 40 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 823 (100, $[M + NH_4]^+$).

14e: Mp. 37-40°C, yield 2.0 g (74%).- $C_{56}H_{88}N_2O_8$ (917.3) Calcd. C 73.3 H 9.67 N 3.1 Found C 73.6 H 9.99 N 3.2.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.64 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.63 (s; 2H, O-CH_2-N), 4.33 and 4.26 (dd each; J = 6.0/11.2 Hz, 2H each, $C\underline{H}_2$-CH-$C\underline{H}_2$), 3.04 (m; 1H, CH), 2.23 (t; J = 7.4 Hz, 4H, 2 x CH_2-CO), 1.55 (m; 4H, 2 x $C\underline{H}_2$-CH_2-CO), 1.25 (m; 56 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 935 (100, $[M + NH_4]^+$).

14f: Oil, yield 1.6 g (58%).- $C_{56}H_{84}N_2O_8$ (913.3) Calcd. C 73.6 H 9.27 N 3.1 Found C 74.2 H 9.79 N 2.9.- ^{1}H-NMR ($CDCl_3$): δ (ppm) = 7.95 (s; 1H, NH), 7.37 (m; 10 H, Ar), 5.62 (s; 2H, O-CH_2-N), 5.35 (m; 4H, 2 x CH=CH), 4.32 and 4.25 (dd each; J = 6.0/11.3 Hz, 2H each, $C\underline{H}_2$-CH-$C\underline{H}_2$), 3.03 (m; 1H, CH), 2.23 (t; J = 7.4 Hz, 4H, 2 x CH_2-CO), 2.00 (m; 8H, 2 x $C\underline{H}_2$-CH=CH-$C\underline{H}_2$), 1.55 (m; 4H, 2 x $C\underline{H}_2$-CH_2-CO), 1.25 (m; 40 H, fatty acid CH_2), 0.88 (t; J = 6.4 Hz, 6H, 2 x CH_3).- DCI-MS: m/z (%) = 931 (100, $[M + NH_4]^+$).

In vitro incubations

The incubations of the glycerides with lipase were performed at 37 ± 0.2°C with an activity of 375 U/ml of porcine pancreatic lipase (assayed by Sigma Chem. Co. using triacetin as substrate, pH 7.4) in 0.1 M PIPES buffer[+], pH 6.5, containing 25 mM $CaCl_2$. Dispersions of the lipids prepared by the addition of 1 part of a solution of the lipids in ethanol to 4 parts of a 25 mM solution of sodium taurodeoxycholate in water followed by brief sonication were added to the prewarmed incubation mixtures to give a final concentration of 1 mM. The reaction mixtures were vigorously stirred during the kinetic run. At selected time intervals 100 µl aliquots were quenched by the addition to 50 µl ice-cold 0.5 M $HClO_4$ followed by dilution with 850 µl water/acetonitrile (7:3, v/v) and centrifugation at 2500 g for 10 min. Aliquots of the clear supernatant were assayed by HPLC on a LiChrospher 100 RP-18 column (125 x 4.6 mm, 5 µm particle size). The mobile phase consisted of phosphate buffer, pH 5.8, containing 25% acetonitrile at a flow rate of 1.5 ml/min. The compounds were detected at 254 nm. The retention times of **1**, **6**, **7**, and **18** were 9.9 min, 13.5 min, 14.0 min, and 7.3 min, respectively, the retention time of the hydroxymethylphenytoin succinic acid mono ester was 4.7 min. The concentrations of the compounds were calculated from calibration curves obtained by analysis of the pure compounds under identical chromatographic conditions. The recovery of each compound was at least 96%.

[+] PIPES: 1.4-Piperazinediethanesulfonic acid

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