Chem. Phys. Lipids **2** (1968) 415–418 © North-Holland Publ. Co., Amsterdam

SYNTHESIS OF L-α-PHOSPHATIDYLETHANOLAMINES FROM DIACYL-L-α-GLYCEROLBROMOHYDRINS

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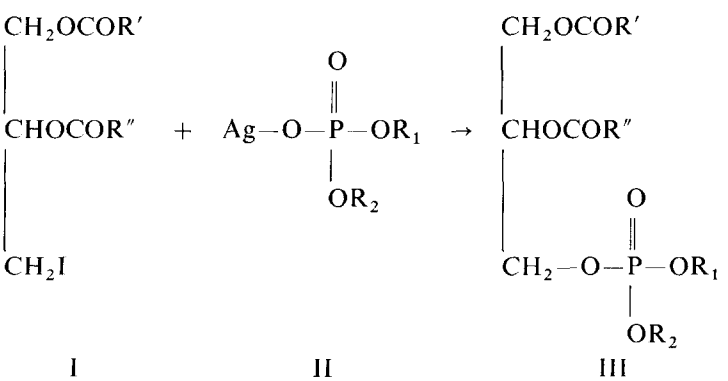
Received 17 May 1968

Diacyl-L-α-glycerolbromohydrins condense with the silver salt of monophenylphosphoryl-N-carbobenzoxyethanolamine in acetonitrile solution. The diacyl-L-α-glycerylphenylphosphoryl-(N-carbobenzoxy)-ethanolamines thus obtained are readily purified by chromatography on silicic acid (SilicAR CC-7) using benzene–ether mixtures for elution. Pure cephalins are obtained from these intermediates by hydrogenolysis in the usual way. L-α-dipalmitoyl cephalin and L-α-distearoyl cephalin were prepared by this procedure.

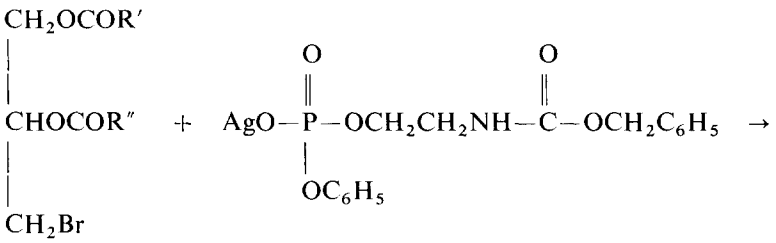
The condensation of diacylglyceroliodohydrins (I) and the silver salt of phosphate diesters (II) is a well-known route to the synthesis of several phosphoglycerides, including phosphatidic acids (PA), phosphatidylethanolamines (PE), phosphatidylcholines (PC) and phosphatidylserines (PS)[1]).



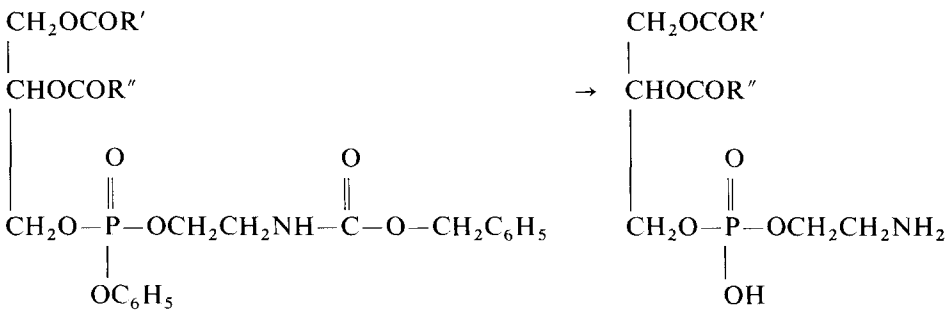
I II III

We have earlier[2]) commented on the drawbacks of the procedure connected with the light sensitivity of diacylglyceroliodohydrins (I) and all their iodine-containing precursors and demonstrated the advantages of using the bromohydrins for the synthesis of phosphatidic acids. We now report the

successful utilisation of the procedure for the synthesis of PE, involving a condensation of diacyl-L-α-glycerolbromohydrins (IV) and the silver salt of phenylphosphoryl-N-carbobenzoxyethanolamine (V).



IV V



VI VII

The bromohydrins (IV) are, as may be anticipated, less reactive and do not condense with the silver salt (V) under the reaction conditions known for the reaction of iodocompounds (I)[3,4]. Thus no reaction was detected when IV and V were refluxed in benzene solution. The reaction, however, proceeds favourably in acetonitrile medium and gives reasonable yields of the product (VI), comparable with those recorded from the corresponding diacylglyceroliodohydrins.

Experimental

Dipalmitoyl-L-α-glycerolbromohydrin

This compound was prepared by the acylation of L-α-glycerol-bromohydrin in 90% yield, as described earlier[2]. It was crystallised from methanol, dried over P_2O_5 under vacuum m.p. 74°, $[\alpha]_D^{22} = +3.31°$ (C, 10) in chloroform (ethanol-free and dry).

Found	C66.28	H10.80	Br13.18%
Calcd. for $C_{35}H_{67}O_4Br$ (M = 631.98)	C66.54	H10.68	Br12.64%

Distearoyl-L-α-glycerolbromohydrin was prepared as already described [2]. *Monophenylphosphoryl-N-carbobenzoxyethanolamine mono-silver salt* was prepared by the method of Baylis et al.[3] and was purified as described by Maurukas et al.[4].

Dipalmitoyl-L-α-glycerylphenylphosphoryl-(N-carbobenzoxy) ethanolamine

Dipalmitoyl-L-α-glycerolbromohydrin (3.4 g, 5.4 mmole) and phenylphosphoryl N-carbobenzoxyethanolamine silver salt (2.0 g, 4.4 mmole) were taken in dry acetonitrile (50 ml) and heated under reflux for 6 hr. Hyflo (0.5 g) was added to the reaction mixture and it was filtered hot. The ppt. was washed with chloroform and the washings bulked with the rest of the filtrate. The solvent was evaporated on a rotary evaporator and the residue chromatographed on a silica column (SilicAR CC-7 ex. Mallinkrodt, activated by heating at 110° for several hours) using a benzene–ether system (continuous gradient, increasing ether) and the fractions were monitored by T.L.C. in hexane–ether (3:1, v/v).

Two by-products (Rf's very close to each other, about 0.8–0.9) were first eluted and then the required compound (Rf 0.4, 3.0 g, 3.3 mmoles).

The compound was crystallised from hexane and dried in vacuum over phosphorous oxide, $[\alpha]_D^{22} = +2.05$ (C, 9.1) in ethanol-free and dry chloroform, m.p. 52–54°. Lit. values[5], $[\alpha]_D^{26} = +2.3°$ (C, 9.2) in chloroform (ethanol-free and dry), m.p. 43–44° (shrinks 42°).

Found	C 67.58	H 9.47	N 1.66%
Calcd. for $C_{51}H_{84}O_{10}NP$ (M = 902.5)	C 67.89	H 9.38	N 1.55%

Dipalmitoyl-L-α-glycerylphosphorylethanolamine

Dipalmitoyl-L-α-glycerylphenylphosphoryl-(N-carbobenzoxy)-ethanolamine (1.0 g, 1.1 mmole) was dissolved in glacial acetic acid (30 ml) and to this a mixture of Adams Catalyst (0.5 g) and palladium black (5% on carbon, 0.5 g) was added. It was hydrogenated at slightly more than one atmosphere pressure, while being stirred by a magnetic stirrer. The hydrogen absorption started after 2–3 min. and was complete in 1½ hr. The cephalin separated from the reaction mixture. The reaction mixture was heated until all the separated cephalin dissolved and the solution was filtered hot. On cooling the filtrate, cephalin crystallised out. It was filtered and washed with anhydrous ether. The yield was (0.7 g, 1.0 mmole, 91%) and the compound was pure by T.L.C. It was recrystallised from hot dry dioxane, filtered and washed with anhydrous ether. The compound was dried in vacuum over phosphorous

oxide. M.p. 190–91°, $[\alpha]22 = +6.4°$ (C, 3.1), in $CHCl_3$ (EtOH-free and dry): AcOH (9:1; v/v). Lit. values, m.p.[7]) 186–987° and specific rotation [6]) $[\alpha]\,D^{24}$ +6.3° in $CHCl_3$:AcOH, 9:1; v/v (C, 3).

Found	C 63.19	H 10.53	N 2.20%
Calcd. for $C_{37}H_{74}O_8NP\frac{1}{2}H_2O$ (M = 700.9)	C 63.39	H 10.78	N 1.99%

Distearoyl-L-α-glycerylphenylphosphoryl-(N-carbobenzoxy)-ethanolamine

The compound was prepared from distearoyl-L-α-glycerolbromohydrin in the same way as the dipalmitoyl compound. It was purified by chromatography on silica (SiliCAR CC-7, ex. Mallinkrodt) using benzene–ether (continuous gradient, increasing amount of ether). The compound was finally crystallised from hexane, m.p. 63–64° (Lit.[5]) sinters 48°, m.p. 51–52°) $[\alpha]_D^{23} = +2.0°$ (C, 10) in $CHCl_3$ (EtOH-free and dry). Lit.[5]) value $[\alpha]_D + 2.1°$ (C, 10) in the same solvent.

Found	C 69.08	H 9.87	N 1.40%
Calcd. for $C_{55}H_{92}O_{10}NP$ (M = 958.3)	C 68.93	H 9.68	N 1.46%

Distearoyl-L-α-glycerylphosphorylethanolamine

It was prepared by the hydrogenation of the above O-phenyl-N-carbobenzoxy derivative in glacial acetic acid and using a mixed Adams and palladium black catalyst in exactly the same manner as in the preparation of Dipalmitoyl-L-α-cephalin. The pure and dry compound had m.p. 188–890°, $[\alpha]_D^{23} = = +6.2°$ in $CHCl_3$ (EtOH-free and dry):AcOH (9:1 v/v) (C, 3.4). Lit. values, m.p.[7]) 180–82° and specific rotation [6]) $[\alpha]_D^{24} = +6.0°$ in the same solvent (C, 4.4)

Found	C 65.75	H 11.07	N 1.64%
Calcd. for $C_{41}H_{82}O_8NP$ (M = 748.1)	C 65.82	H 11.04	N 1.87%

Acknowledgment

The author is grateful to Mr. J. A. Henrys for technical assistance.

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