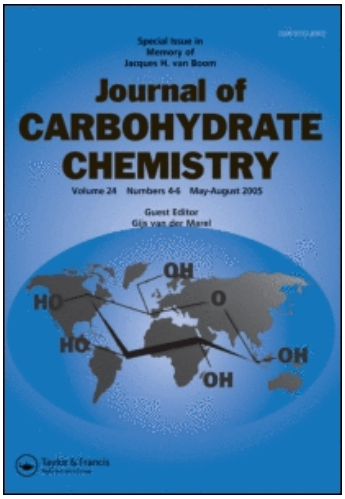

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Synthesis of 1,6-Anhydro-2,3-di-*O*-farnesyl-5-*O*-([{phosphonomethyl}phosphinyl]methyl)-α-d-galactofuranose: A Zaragozic Acid-Presqualene Pyrophosphate Hybrid

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SYNTHESIS OF 1,6-ANHYDRO-2,3-DI-*O*-FARNESYL-5-*O*-([{PHOSPHONOMETHYL}PHOSPHINYL]METHYL)-α-D-GALACTOFURANOSE: A ZARAGOZIC ACID - PRESQUALENE PYROPHOSPHATE HYBRID

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ABSTRACT

A multi-step synthesis of a new potential inhibitor (*i.e.*, compound **2**) of squalene synthase is described, starting from D-galactose. The title compound is a hybrid of presqualene pyrophopshate and the recently discovered zaragozic acids, which are picomolar competitive inhibitors of the enzyme squalene synthase.

INTRODUCTION

Recently, researchers at Glaxo Group Research Ltd elucidated[1] the structure of three novel fungal metabolites, designated the squalestatins 1-3, which were isolated[2] from the culture *Phoma* sp. C2932. It was established that these compounds are potent competitive inhibitors of the enzymes squalene synthase[3a] and protein:farnesyl transferase.[3b] At the same time, researchers at Merck Research Laboratories[4-6] isolated

the structurally related zaragozic acids A-C from the fungal cultures *Sporormiella intermedia* (zaragozic acid A) and *Leptodontium elatius* (zaragozic acids B and C). The common structural element in this new class of fungal metabolites is the polar 2,8-dioxabicyclo[3.2.1]octane-4,6,7-trihydroxy-3,4,5-tricarboxylic acid core. On the other hand, they are distinguished from each other by the presence of different acyl and alkyl side-chains at O-6 and C-1 (see for instance the structures of squalestatin 2 (**1a**) and zaragozic acid B (**1b**) in Fig. 1). The intriguing structural features of this new class of compounds prompted several research groups to embark on the synthesis of the zaragozic acids (squalestatins).[7] Recently, Dufresne *et al.*[6] postulated that the highly effective inhibitory action of the zaragozic acids (squalestatins) on squalene synthase may be ascribed to their topological similarity with the rigid presqualene pyrophosphate (PPP), an intermediate in the squalene synthase-mediated conversion of farnesyl pyrophosphate to squalene. The structural similarity between PPP and zaragozic acids/squalestatins (*i.e.* both molecules possess rigid triacid cores flanked by two lipophilic residues) goaded us to design a hybrid of **1** and PPP embracing a rigid core, two lipophilic farnesyl residues and a triacid phosphinylphosphonate moiety.

As part of an ongoing program[8] to synthesize effective squalene synthase inhibitors we here report the preparation of compound **2**, the 4,6,7-trihydroxy-2,8-dioxabicyclo[3.2.1]octane core which is substituted with two farnesyl moieties at O-6 and O-7 and a phosphonomethylphosphinylmethyl function at O-4.

RESULTS AND DISCUSSION

Retrosynthetic analysis of target compound **2** reveals that D-galactose is an appropriate starting material for the preparation of the bicyclic core. Initially, the synthesis of compound **2** was pursued by the sequence of reactions outlined in Scheme 1, the key step of which entails cyclisation of 1-ethyl 2,3-di-*O*-farnesyl-5-*O*-(diethyl phosphonomethyl)-1-thio-α-D-galactofuranoside **10** to the corresponding 1,6-anhydro derivative **16** (see Scheme 2).

The synthetic pathway to key intermediate **10** commences with the acid mediated ring-closure[9] of readily available[10] D-galactose diethyl dithioacetal **3** to give the known[9] ethyl 1-thio-α-D-galactofuranoside (**4**). Selective acetonation of the 5,6-diol in **4** with

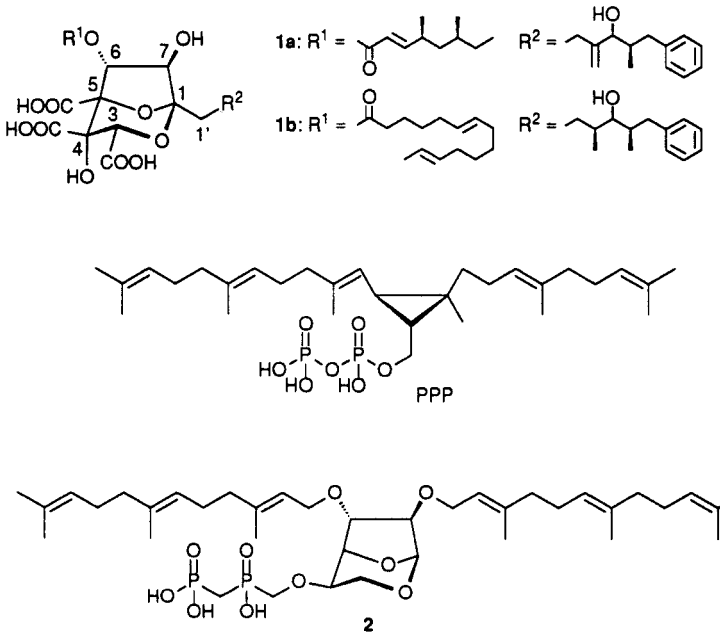
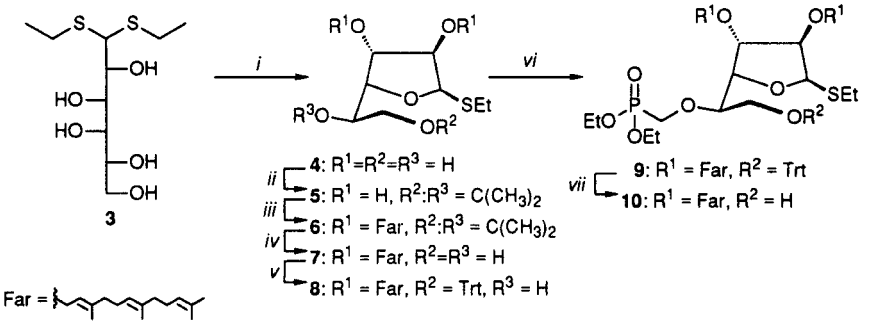


Figure 1



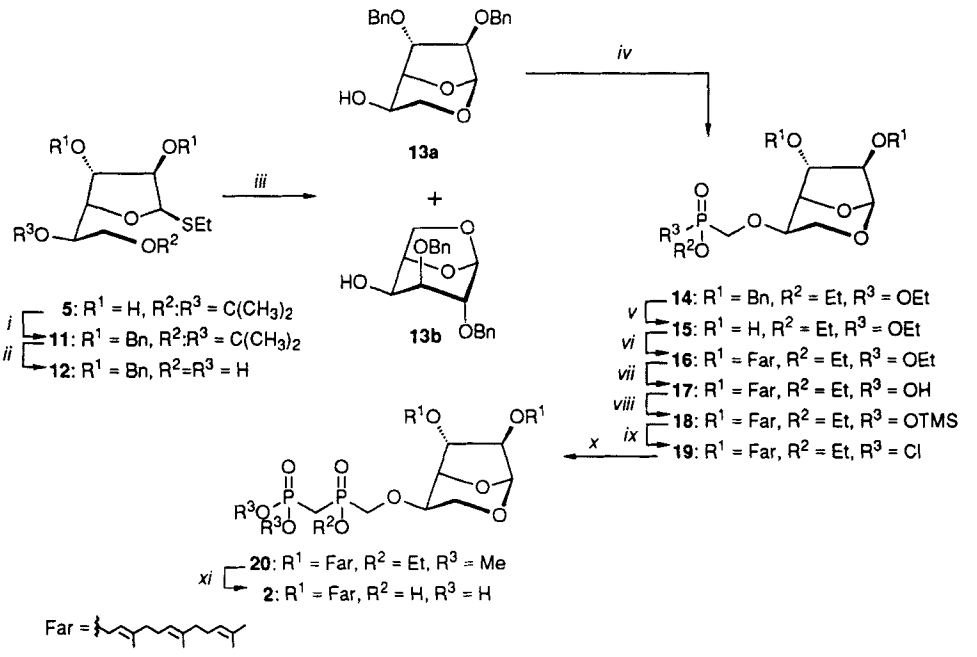
[a]Reagents and conditions

i: 1) dil. HCl, 2) HgO (72%); *ii*: 2,2-dimethoxypropane, DMF, cat. camphorsulfonic acid (70%); *iii*: Farnesyl bromide, NaH, DMF (44%); *iv*: $CuCl_2$, 2-propanol (71%); *v*: Trityl chloride, pyridine, 65 °C (85%); *vi*: KH, THF, $TfOCH_2P(O)(OEt)_2$ (52%); *vii*: HCOOH, MeOH, 0 °C (100%).

Scheme 1[a]

2,2-dimethoxypropane in DMF and a catalytic amount of camphorsulphonic acid gave mono-acetonide derivative **5**. Bis-farnesylation of **5** with farnesyl chloride[11] in DMF and sodium hydride was unfruitful. However, the required di-farnesylated product **6** could be isolated in 44% yield using instead farnesyl bromide[11] as the alkylating agent. Unfortunately, any attempt to improve the effectivity of the farnesylation was abortive. Deacetonation of **6** with $CuCl_2 \cdot 2H_2O$ in 2-propanol[12] proceeded smoothly to give **7** in 71% yield. In this respect it is of interest to note that deblocking of acetonide **6** could not be realized using aqueous acetic acid. However, employment of catalytic pyridinium *p*-toluene sulfonate gave diol **7** in a moderate yield of 50%. Treatment of **7** with trityl chloride in pyridine at elevated temperature, followed by phosphonylation of **8** with diethyl phosphonomethyl triflate[13] and potassium hydride, afforded the fully protected galactofuranoside derivative **9** in an overall yield of 44%. Acidolysis of **9** proceeded quantitatively to give the requisite intermediate **10**. Bromonium-ion (NBS) mediated cyclisation of **10** led to complete recovery of starting material. Moreover, $FeCl_3$-mediated cyclisation of **10** in acetonitrile, as reported earlier by Åberg *et al.*,[14] was not successful either. The unexpected stability of the isopropylidene function as well as the failure to cyclize **10** may be ascribed to the presence of the lipophilic farnesyl residues. Thus, it is not excluded that the farnesyl units may shield the anomeric thioethyl group in the armed[15] galactofuranoside derivative **10** from electrophilic attack.

It occurred to us that introduction of the farnesyl residues at a later stage of the synthesis would have a beneficial effect on the cyclisation process. To this end, we first prepared ethyl 2,3-di-*O*-benzyl-1-thio-α-D-galactofuranoside **12**, as delineated in Scheme 2. Hence, benzylation of **5** and subsequent deacetonation of **11** with aqueous acetic acid furnished the partially benzylated derivative **12**. Cyclisation of **12** according to the procedure of Åberg *et al.*[14] gave the expected 1,6-anhydrofuranoside **13a** and 1,6-anhydropyranoside **13b** in a ratio of 9 to 1. Purification of the reaction mixture by silica gel column chromatography afforded homogeneous **13a** in 64% yield. The ^{13}C and ^{1}H NMR spectroscopic data of **13a** were in complete accordance with those of the same compound prepared starting from ethyl 2,3-di-*O*-benzyl-1-thio-α-D-galactopyranoside. Phosphonylation of **13a** with diethyl phosphonomethyl triflate led to the fully protected derivative **14**. Hydrogenolysis of **14** in the presence of $Pd(OH)_2$ and consecutive



[a]Reagents and conditions

i: Benzyl bromide, NaH, DMF (85%); *ii*: 80% HOAc (91%); *iii*: $FeCl_3$, CH_3CN, reflux (**13a**: 64%, **13b**: 7%); *iv*: KH, THF, $TfOCH_2P(O)(OEt)_2$ (68%); *v*: H_2, $Pd(OH)_2$ (100%); *vi*: Farnesyl bromide, NaH, DMF (40%); *vii*: N KOH/ethanol, reflux; *viii*: *N,N*-Et_2NTMS; *ix*: oxalyl chloride/DMF, 0 °C; *x*: $LiCH_2P(O)(OMe)_2$, THF, -78 °C (41%); *xi*: 1) TMS-Br, *sym*-collidine, 2) N KOH (35%).

Scheme 2[a]

farnesylation of crude diol **15** with farnesyl bromide and sodium hydride furnished farnesylated compound **16** in 40% yield. Saponification of diester **16** and subsequent silylation[16] of the intermediate mono-ester **17** was followed by conversion of resulting **18** into phosphonic chloride **19**. The latter was treated with dimethyl lithiomethylphosphonate, obtained *in situ* by reaction of dimethyl methylphosphonate with *n*-butyllithium at -78 °C, to produce protected phosphinylphosphonate **20** in 41% yield over the four steps. The presence of the phosphinylphosphonate function in **20** was unambiguously ascertained by ^1H, ^{13}C and ^{31}P NMR spectroscopy. Finally, the phosphorus protecting groups were removed by transesterification of **20** with trimethylsilyl bromide and basic hydrolysis of the resulting TMS-esters to furnish target compound **2**, the identity of which was firmly established with ^1H, ^{13}C and ^{31}P NMR spectroscopy.

In conclusion, the results presented in this paper show that compound **2** is readily accessible by a straightforward route, starting from D-galactose diethyl dithioacetal **3**. The inhibitory action of hybrid molecule **2** on the enzyme squalene synthase may open the way to a new series of synthetic squalene synthase inhibitors based on carbohydrates.

Experimental

General methods. (*E,E*)-Farnesol was purchased from Aldrich and distilled. Farnesyl chloride and farnesyl bromide were prepared as described previously.[11] *N,N*-Dimethylformamide was stirred overnight with CaH_2, distilled under reduced pressure and stored over molecular sieves (0.4 nm). Methanol and ethanol were dried by refluxing with magnesium methoxide or magnesium ethoxide, respectively, distilled and stored over molecular sieves (0.3 nm). Toluene, dichloromethane and ether were dried by refluxing with P_2O_5 for 2 h and then distilled. Toluene and ether were stored over sodium wire. Dichloromethane was stored over molecular sieves (0.4 nm). THF and acetonitrile were dried by refluxing with CaH_2 for 16 h, distilled and stored over molecular sieves (0.4 nm). THF and ether were redistilled from $LiAlH_4$ directly before use. TLC-analysis was performed on silica gel (Schleicher & Schull, F 1500 LS 254). Compounds were visualised by spraying the TLC-plates with $KMnO_4$ (1%) in aqueous Na_2CO_3 (2%) or by charring with concd sulfuric acid/methanol (2:8 v/v). Column chromatography was performed on Merck Kieselgel (230-400 Mesh ASTM). Evaporations were carried out below 40 °C under reduced pressure (15 mmHg). ^1H, ^{13}C and ^{31}P NMR spectra were measured at 199.99, 50.1 and 80.7 MHz, respectively, using a JEOL JNM-FX 200 spectrometer on line with a JEC 980 B computer. ^1H and ^{13}C chemical shifts are given in ppm (δ) relative to tetramethylsilane (TMS) as internal standard and ^{31}P chemical shifts are given in ppm (δ) relative to 85% H_3PO_4 as external standard.

Ethyl 5,6-*O*-Isopropylidene-1-thio-α-D-galactofuranoside (5). Ethyl 1-thio-α-D-galactofuranoside (5.6 g, 25 mmol) was dissolved in DMF (10 mL) and treated with 2,2-dimethoxypropane (3.4 mL, 27.5 mmol) and camphorsulfonic acid (50 mg). After stirring overnight at rt, the reaction mixture was quenched with NEt_3 (5 mL) and concentrated under reduced pressure. Column chromatography of the residue (CH_2Cl_2/MeOH 1:0 → 95:5 v/v) afforded pure **5** (4.6 g, 70%). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 15.3

(SCH_2CH_3); 25.4 (SCH_2CH_3); 25.6, 26.1 ($C(CH_3)_2$); 65.3 (C-6); 76.6, 78.1, 78.3, 83.3 (C-2, C-3, C-4, C-5); 88.7 (C-1); 110.0 ($C(CH_3)_2$). ^1H NMR ($CDCl_3$) δ 1.32 (t, 3H, SCH_2CH_3); 1.39, 1.43 (2s, 6H, $C(CH_3)_2$); 2.73 (AB, 2H, SCH_2CH_3); 5.34 (d, 1H, H-1, $J_{1,2}$ = 3.8 Hz).

Ethyl 2,3-Di-*O*-farnesyl-5:6-*O*-isopropylidene-1-thio-α-D-galactofuranoside (6). To a stirred solution of ethyl 5,6-*O*-isopropylidene-1-thio-α-D-galactofuranoside (**5**, 2.64 g, 10 mmol) in DMF was added NaH (720 mg, 30 mmol) at 0 °C. After stirring for 30 min, farnesyl bromide (6.3 g, 22 mmol) was added and stirring was continued at rt for 2 h. The reaction mixture was quenched with dry methanol and concentrated. The residue was dissolved in ether, washed with H_2O, 10% $NaHCO_3$, H_2O and dried over $MgSO_4$. After evaporation of the solvent, the residue was purified by silica gel column chromatography (petroleum ether 40-60/ether 1:0 → 8:2 v/v). Combination of the appropriate fractions gave homogenous **6** (3 g, 44%). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 14.6 (SCH_2CH_3); 15.7, 16.3, 17.4 ($6CH_3$ Farnesyl); 24.0 (SCH_2CH_3); 25.0, 26.4 ($C(CH_3)_2$); 25.4 ($2CH_3$ Farnesyl); 26.0, 26.5 ($4CH_2$ Farnesyl); 39.3, 39.5 ($4CH_2$ Farnesyl); 65.0 (C-6); 66.3, 66.6 ($2OCH_2$ Farnesyl); 77.3, 82.0, 83.0, 84.0 (C-2, C-3, C-4, C-5); 86.2 (C-1); 109.2 ($C(CH_3)_2$); 120.1, 123.5, 124.1 (6CH Farnesyl); 130.8, 135.0, 140.6 ($6C_q$ Farnesyl).

Anal. Calcd for $C_{41}H_{68}O_5S$ (673.06): C, 73.17; H, 10.18. Found: C, 73.04; H, 10.20.

Ethyl 2,3-Di-*O*-farnesyl-1-thio-α-D-galactofuranoside (7). Compound **6** (673 mg, 1 mmol) was dissolved in 2-propanol (10 mL) and $CuCl_2 \cdot 2H_2O$ (900 mg, 5 mmol) was added. After stirring for 16 h at rt the reaction was quenched by addition of 1 M Na_2CO_3 (5 mL) and the mixture was diluted with ethyl acetate. The solids were removed by filtration and the filtrate was concentrated. The residue was purified by silica gel column chromatography (elution: petroleum ether 40-60/ether 1:0 → 1:1 v/v) to give **7** (449 mg, 71%). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 15.0 (SCH_2CH_3); 15.8, 16.4, 17.5 ($6CH_3$ Farnesyl); 24.8 (SCH_2CH_3); 25.5 ($2CH_3$ Farnesyl); 26.1, 26.6 ($4CH_2$ Farnesyl); 39.5 ($4CH_2$ Farnesyl); 64.2 (C-6); 66.7 ($2OCH_2$ Farnesyl); 71.4, 82.0, 83.4 (C-2, C-3, C-4, C-5); 87.2 (C-1); 119.8, 120.0, 123.6, 124.1 (6CH Farnesyl); 131.0, 135.2, 141.0, 141.2 ($6C_q$ Farnesyl).

Ethyl 2,3-Di-*O*-farnesyl-6-*O*-trityl-1-thio-α-D-galactofuranoside (8). Compound **7** (2.33 g, 3.7 mmol) was dissolved in pyridine and trityl chloride (1.3 g, 4.6 mmol) was

added. After stirring for 16 h at 65 °C the reaction mixture was cooled to rt and the excess trityl chloride was destroyed with MeOH. The volatiles were removed by evaporation and the residue was dissolved in CH_2Cl_2, washed with water, 10% $NaHCO_3$, water, dried ($MgSO_4$) and concentrated. Purification was effected by silica gel column chromatography (elution: petroleum ether 40-60/ether 1:0 → 1:1) to give **8** as a colourless oil (2.7 g, 85%). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 14.9 (SCH_2CH_3); 15.8, 16.4, 17.5 ($6CH_3$ Farnesyl); 24.6 (SCH_2CH_3); 25.5 ($2CH_3$ Farnesyl); 26.1, 26.5 ($4CH_2$ Farnesyl); 39.4, 39.5 ($4CH_2$ Farnesyl); 66.3 (C-6); 66.9 ($2OCH_2$ Farnesyl); 69.9, 83.0, 83.5 (C-2, C-3, C-4, C-5); 86.5 (CPh_3); 86.8 (C-1); 120.0-128.5 (6CH Farnesyl, C_{arom} Trityl); 135.0-146.8 ($6C_q$ Farnesyl, C*q* Trityl).

Ethyl 2,3-Di-*O*-farnesyl-5-*O*-(diethyl phosphonomethyl)-6-*O*-trityl-1-thio-α-D-galactofuranoside (9). Compound **8** (874 mg, 1 mmol) was dissolved in DMF (5 mL) and treated with KH (80 mg, 2 mmol). After stirring for 1 h, diethyl phosphonomethyl triflate (330 mg, 1.1 mmol) was added and stirring was continued for 5 h. When TLC-analysis (ether) indicated that no further reaction took place the reaction was quenched with methanol, diluted with ether and washed with 10% $NaHCO_3$. The organic layer was dried over $MgSO_4$ and concentrated. Purification of the residue by silica gel column chromatography (elution: ether) gave compound **9** (532 mg, 52%). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 14.9, 15.9, 16.4, 17.5 (SCH_2CH_3, $2OCH_2CH_3$, $6CH_3$ Farnesyl); 24.6 (SCH_2CH_3); 25.5 ($2CH_3$ Farnesyl); 26.1, 26.6 ($4CH_2$ Farnesyl); 39.4, 39.5 ($4CH_2$ Farnesyl); 61.9-67.3 (C-6, $2OCH_2$ Farnesyl, OCH_2P, $2OCH_2CH_3$); 82.7, 82.9, 83.4, 85.4 (C-2, C-3, C-4, C-5); 86.9 (CPh_3); 87.1 (C-1); 119.9.0-128.8 (6CH Farnesyl, C_{arom} Trityl); 131.0-143.6 ($6C_q$ Farnesyl, C*q* Trityl). $^{31}P\{^1H\}$ NMR ($CDCl_3$) δ 21.9.

Anal. Calcd for $C_{62}H_{89}O_8PS$ (1025.43): C, 72.63; H, 8.75; P, 3.02. Found: C, 72.59; H, 8.80; P, 2.99.

Ethyl 2,3-Di-*O*-farnesyl-5-*O*-(diethyl phosphonomethyl)-1-thio-α-D-galactofuranoside (10). Compound **9** (1 g, 1 mmol) was dissolved in MeOH (2 mL) and cooled to 0 °C. Then formic acid (8 mL) was added and the reaction was stirred for 1 h at 0 °C. The mixture was concentrated and the residue was purified by silica gel column chromatography (elution: ether) to give pure **10** (783 mg, 100%). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 15.0, 15.9, 16.3, 17.6 (SCH_2CH_3, $2OCH_2CH_3$, $6CH_3$ Farnesyl); 24.8 (SCH_2CH_3); 25.6

($2CH_3$ Farnesyl); 26.2, 26.6 ($4CH_2$ Farnesyl); 39.5, 39.6 ($4CH_2$ Farnesyl); 61.9-67.3 (C-6, $2OCH_2$ Farnesyl, OCH_2P, $2OCH_2CH_3$); 82.5, 82.9, 83.6, 85.4 (C-2, C-3, C-4, C-5); 86.9 (C-1); 119.9, 123.6, 124.2, 128.6 (6CH Farnesyl); 135.2, 141.1, 141.2 ($6C_q$ Farnesyl). $^{31}P\{^1H\}$ NMR δ 23.1.

Ethyl 2,3-Di-*O*-benzyl-5:6-*O*-isopropylidene-1-thio-α-D-galactofuranoside (11). To a stirred solution of ethyl 5,6-*O*-isopropylidene-1-thio-α-D-galactofuranoside (**5**, 2.64 g, 10 mmol) in DMF was added NaH (720 mg, 30 mmol) at 0 °C. After stirring for 30 min, benzyl bromide (2.6 mL, 22 mmol) was added and stirring was continued at rt for 2 h. The reaction mixture was quenched with dry methanol and concentrated. The residue was dissolved in ether, washed with H_2O, 10% $NaHCO_3$, H_2O and dried over $MgSO_4$. After evaporation of the solvent, the residue was purified by silica gel column chromatography (petroleum ether 40-60/ether 1:0 → 8:2 v/v). Combination of the appropriate fractions gave homogenous **11** (3.8 g, 85%). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 15.1 (SCH_2CH_3); 24.5 (SCH_2CH_3); 25.2, 26.7 ($C(CH_3)_2$); 65.2 (C-6); 71.9, 72.4 ($2CH_2$ Bn); 77.1, 82.5, 83.4, 84.5 (C-2, C-3, C-4, C-5); 86.5 (C-1); 109.4 ($C(CH_3)_2$); 127.2-128.5 (C_{arom} Bn); 137.5, 137.7 ($2C_q$ Bn).

Ethyl 2,3-Di-*O*-benzyl-1-thio-α-D-galactofuranoside (12). Compound **11** (2.2 g, 5 mmol) was dissolved in 80% acetic acid (20 mL) and stirred for 2 h at 75 °C. When TLC-analysis (ether) showed that the reaction was complete the solvent was removed *in vacuo* and toluene (10 mL) was evaporated from the residue 5 times. Purification of the remaining oil by silica gel column chromatography (elution: ether) gave compound **12** (1.8 g, 91%). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 14.6 (SCH_2CH_3); 24.5 (SCH_2CH_3); 63.2 (C-6); 71.5, 71.7 ($2CH_2$ Bn); 71.2, 81.9, 82.8, 83.4 (C-2, C-3, C-4, C-5); 86.5 (C-1); 127.9-128.4 (C_{arom} Bn); 136.7, 137.1 ($2C_q$ Bn).

1,6-Anhydro-2,3-di-*O*-benzyl-α-D-galactofuranose (13a). Compound **12** (2.02 g, 5 mmol) was dissolved in acetonitrile (375 mL) and anhydrous $FeCl_3$ (227 mg, 1.4 mmol) was added. The reaction mixture was refluxed for 30 min, after which period TLC-analysis (CH_2Cl_2/ether 5:1 v/v) showed complete conversion of the starting material. The solvent was removed and the residue was purified by silica gel column chromatography (elution: CH_2Cl_2/ether 1:0 → 5:1 v/v) to give **13a** (1.1 g, 64%) and **13b** (119 mg, 7%). **13a**: $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 62.3 (C-5); 65.3 (C-6); 71.2, 72.3 ($2CH_2$

Bn); 80.9, 81.6, 85.2 (C-2, C-3, C-4); 96.7 (C-1); 127.8-128.3 (C_{arom} Bn); 137.3, 137.4 ($2C_q$ Bn). ^1H NMR ($CDCl_3$) δ 3.68 (t, 1H, H-6a, $J_{6a,6b}$ = 10 Hz); 4.01 (ddd, 1H, H-6b); 4.03-4.11 (m, 1H, H-5); 4.14 (dq, 1H, H-2); 4.19 (d, 1H, H-3); 4.23 (d(b), 1H, H-4); 5.30 (d, 1H, H-1, $J_{1,2}$ = 3.8 Hz).

Anal. Calcd for $C_{20}H_{22}O_5$ (342.40): C, 70.16; H, 6.48. Found: C, 70.11; H, 6.50.

1,6-Anhydro-2,3-di-*O*-benzyl-5-*O*-(diethyl phosphonomethyl)-α-D-galactofuranose (14). Compound **13a** (342 mg, 1 mmol) was dissolved in DMF (5 mL) and treated with KH (80 mg, 2 mmol). After stirring for 1 h diethyl phosphonomethyl triflate (330 mg, 1.1 mmol) was added and stirring was continued for 5 h. When TLC-analysis (ether) indicated that no further reaction took place the reaction was quenched with methanol, diluted with ether and washed with 10% $NaHCO_3$. The organic layer was dried over $MgSO_4$ and concentrated. Purification of the residue by silica gel column chromatography (elution: ether) gave compound **14** (335 mg, 68%). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 16.2 (OCH_2CH_3); 62.2 (OCH_2CH_3); 63.3 (C-6); 63.5 (d, OCH_2P, J_{C-P} = 167.6 Hz); 71.9 (d, C-5, J_{C-P} = 10.3 Hz); 71.1, 72.1 ($2CH_2$ Bn); 79.0 (C-4); 81.1, 85.2 (C-2, C-3); 96.8 (C-1); 127.5-128.1 (C_{arom} Bn); 137.2, 137.4 ($2C_q$ Bn); ^1H NMR ($CDCl_3$) δ 1.24 (dt, 6H, $2OCH_2CH_3$); 3.66-3.80 (m, 4H, H-6a, H-5, OCH_2P); 3.98-4.10 (m, 7H, H-2, H-3, H-6b, $2OCH_2CH_3$); 4.34 (d(b), 1H, H-4); 4.39-4.55 (2AB, 4H, $2OCH_2$ Bn); 5.25 (d, 1H, H-1, $J_{1,2}$ = 4.36 Hz). $^{31}P\{^1H\}$ NMR ($CDCl_3$) δ 20.6.

Anal. Calcd for $C_{25}H_{33}O_8P$ (492.51): C, 60.97; H, 6.75; P, 6.29. Found: C, 61.01; H, 6.74; P, 6.30.

1,6-Anhydro-5-*O*-(diethyl phosphonomethyl)-α-D-galactofuranose (15). To a solution of compound **14** (492 mg, 1 mmol) in 2-propanol/water (1:1 v/v, 5 mL) was added $Pd(OH)_2$ (25 mg). The mixture was shaken overnight in a Parr-apparatus under an atmosphere of hydrogen. The $Pd(OH)_2$ was filtered off and the solution was concentrated *in vacuo*. The residue was used without purification in the next step. $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 15.8, 15.9 ($2OCH_2CH_3$); 62.1, 62.3, 62.4, 62.5 ($2OCH_2CH_3$); 62.6 (C-6); 62.7 (d, OCH_2P, J_{C-P} = 166.7 Hz); 71.8 (d, C-5, J_{C-P} = 10.2 Hz); 75.0, 80.4, 81.6 (C-2, C-3, C-4); 98.1 (C-1). $^{31}P\{^1H\}$ NMR ($CDCl_3$) δ 20.9.

1,6-Anhydro-2,3-di-*O*-farnesyl-5-*O*-(diethyl phosphonomethyl)-α-D-galactofuranose (16). Crude compound **15** was dissolved in DMF (5 mL) and NaH (96 mg, 4 mmol) was added. After stirring for 1 h, the reaction mixture was treated with

farnesyl bromide (1.2 mL, 4 mmol) and stirring was continued until TLC-analysis ($MeOH/CH_2Cl_2$ 2:8 v/v) showed no further conversion of the starting material. The reaction was quenched with MeOH, the mixture was diluted with ether, washed with water, 10% $NaHCO_3$, water, and dried over $MgSO_4$. After evaporation of the solvent, the residue was purified by silica gel column chromatography (elution: $MeOH/CH_2Cl_2$ 0:1 → 1:9 v/v) to give the title compound as a colourless oil (288 mg, 40% over the two steps). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 15.9 (OCH_2CH_3); 16.4, 16.5, 17.6, 25.7 ($8CH_3$ Farnesyl); 26.3, 26.7, 39.6, 39.7 ($8CH_2$ Farnesyl); 62.5 (OCH_2CH_3); 63.5 (C-6); 63.7 (d, OCH_2P, J_{C-P} = 167.9); 72.3 (d, C-5, J_{C-P} = 9.6 Hz); 66.1, 66.9 ($2OCH_2$ Farnesyl); 79.2, 81.3, 85.3 (C-2, C-3, C-4); 97.3 (C-1); 120.1, 123.7, 124.3 (6CH Farnesyl); 135.3, 140.6, 141.3 ($6C_q$ Farnesyl). $^{31}P\{^1H\}$ NMR ($CDCl_3$) δ 20.7.

Anal. Calcd for $C_{41}H_{69}O_8P$ (720.97): C, 68.30; H, 9.65; P, 4.30. Found: C, 68.24; H, 9.61; P, 4.24.

1,6-Anhydro-2,3-di-*O*-farnesyl-5-*O*-(ethyl {[dimethyl phosphonomethyl]-phosphinyl}-methyl)-α-D-galactofuranose (20). To a solution of **16** (300 mg, 0.42 mmol) in ethanol (4 mL) was added 1 M KOH (4 mL), and the reaction was refluxed for 16 h. After cooling to rt, ethanol was evaporated and the aqueous residue was stirred with dichloromethane and acidified with 10% HCl. The organic layer was washed with water and brine, dried ($MgSO_4$) and concentrated to provide **17**. This was used without further purification in the next step.

To a stirred solution of monoester **17** in CH_2Cl_2 under argon was added *N,N*-diethyl(trimethylsilyl)amine (162 μL, 0.81 mmol). The reaction was allowed to stir for 1.5 h at rt, the solvent was evaporated and the residue was dissolved in toluene (10 mL) and concentrated. The residue was redissolved in CH_2Cl_2 (2 mL) containing one drop of DMF, under argon at 0 °C, and oxalyl chloride (73 μL, 0.84 mmol) was added. After 45 min at 0 °C and 45 min at rt the solution was concentrated and the solution of the residue in toluene (5 mL) concentrated twice to give phosphonic chloride **19**.

To a solution of dimethyl methylphosphonate (100 μL, 0.92 mmol) in THF (2.5 mL) at -78 °C under argon was added *n*-BuLi (560 μL as 1.6 M in hexane, 0.9 mmol). After 40 min, the acid chloride **19** prepared above in THF (2 mL) was added. The reaction was stirred for 1 h at -78 °C before it was quenched with saturated NH_4Cl and diluted with ether. The aqueous layer was made acidic with 10% HCl and the organic

layer was separated and washed with brine. The aqueous layer was re-extracted with CH_2Cl_2, and the CH_2Cl_2 was washed with brine. The combined organic layers were dried ($MgSO_4$) and concentrated. The crude product was purified by silica gel column chromatography (elution: CH_2Cl_2/MeOH 100:0 → 90:10 v/v) to give **20** as a colourless oil (137 mg, 41% yield over the four steps). $^{13}C\{^1H\}$ NMR ($CDCl_3$) δ 15.8 (OCH_2CH_3); 16.4, 17.5, 18.0, 25.5 ($8CH_3$ Farnesyl); 26.1, 26.5, 39.4, 39.5 ($8CH_2$ Farnesyl); 52.9 (OCH_3); 63.3 (OCH_2CH_3); 63.4 (C-6); 63.9 (d, OCH_2P, J_{C-P} = 163.1); 72.4 (d, C-5, J_{C-P} = 14.7 Hz); 66.5, 66.8 ($2OCH_2$ Farnesyl); 79.1, 81.2, 85.1 (C-2, C-3, C-4); 97.2 (C-1); 120.0, 123.6, 124.1 (6CH Farnesyl); 135.2, 140.5, 141.3 ($6C_q$ Farnesyl). ^1H NMR ($CDCl_3$) δ 1.33-1.39 (m, 3H, OCH_2CH_3); 1.59, 1.68, 1.69 (3s, 24H, $8CH_3$ Farnesyl); 1.95-2.12 (m, 16H, $8CH_2$ Farnesyl); 2.40-2.57 (m, 2H, PCH_2P); 3.58-3.86 (m, 8H, $2OCH_3$, H-5, H-6a); 3.88-4.23 (m, 11H, OCH_2P, $2OCH_2$ Farnesyl, OCH_2CH_3, H-2, H-3, H-6b); 4.35 (t(b), 1H, H-4); 5.07-5.11 and 5.36-5.39 (2m, 6H, 6CH Farnesyl); 5.29 and 5.30 (2d, 1H, H-1). $^{31}P\{^1H\}$ NMR ($CDCl_3$) δ 22.7, 40.1, 41.2

Anal. Calcd for $C_{42}H_{72}O_{10}P_2$ (798.98): C, 63.14; H, 9.08; P, 7.75. Found: C, 63.08; H, 9.01; P, 7.72.

1,6-Anhydro-2,3-di-*O*-farnesyl-5-*O*-({[phosphonomethyl]phosphinyl}-methyl)-α-D-galactofuranose (2). To a stirred solution of **20** (100 mg, 0.125 mmol) in CH_2Cl_2 (2 mL) at rt was added *sym*-collidine (50 μL, 0.38 mmol) followed by bromotrimethylsilane (75 μL, 0.42 mmol). The reaction was allowed to stir for 23 h at rt. The volatiles were removed *in vacuo* and the residue was redissolved in toluene (5 mL) and evaporated. The remainder was treated with 1 M KOH (5 mL) and stirred for 30 min at rt, diluted with water and lyophilized. The crude mixture was purified on a CHP20P column that was eluted with a linear gradient of 80% acetonitrile/water in 5% methanol/water. Lyophilization of the appropriate fractions yielded pure **2** (tripotassium salt, 37 mg, 35%). $[\alpha]_D$ +6.2° (*c* = 0.37); $^{13}C\{^1H\}$ NMR (D_2O) δ 16.5, 16.9, 18.1, 26.1 ($8CH_3$ Farnesyl); 27.5, 40.4 ($8CH_2$ Farnesyl); 64.3 (C-6); 64.7 (d, OCH_2P, J_{C-P} = 153.7); 66.5, 66.6 ($2OCH_2$ Farnesyl); 71.7 (d, C-5, J_{C-P} = 11.2 Hz); 80.5, 81.3, 84.8 (C-2, C-3, C-4); 97.6 (C-1); 120.5, 124.8, 125.0 (6CH Farnesyl); 131.2, 135.4, 135.4 ($6C_q$ Farnesyl). ^1H NMR (D_2O) δ 1.58, 1.59, 1.66, 1.70 (4s, 24H, $8CH_3$ Farnesyl); 1.97-2.17 (m, 18H, $8CH_2$ Farnesyl, PCH_2P); 5.30 (d(b), 1H, H-1). $^{31}P\{^1H\}$ NMR (D_2O) δ 32.0 ($J_{P,P}$ = 17.1 Hz); 13.8 ($J_{P,P}$ = 16.9).

Anal. Calcd for $C_{38}H_{61}P_2O_{10}K_3$ (857.14): C, 53.26; H, 7.18; P, 7.24. Found: C, 53.32; H, 7.12; P, 7.19.

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