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TITLE: SYNTHESIS OF PHOSPHATES AND PHOSPHONATES OF 1',2'-SECO-NUCLEOSIDES AND OTHER ANTIVIRALS

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19. ABSTRACT (Continue on reverse if necessary and identify by block number)

Synthetic methodologies for the preparation of 1',2'-seco-nucleoside phosphates and phosphonates have been developed. 5'-Phosphates as well as 3', 5-cyclic phosphates have been synthesized.

A unique reaction was discovered that allows the preparation of phosphonate chirons. The latter have been successfully converted to 1', 2'-seco-nucleoside phosphonates and homophosphonates.

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FOREWORD

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PI - Signature DATE

A. Objective: The synthesis of 1',2'-*seco*-nucleoside phosphates and phosphonates.

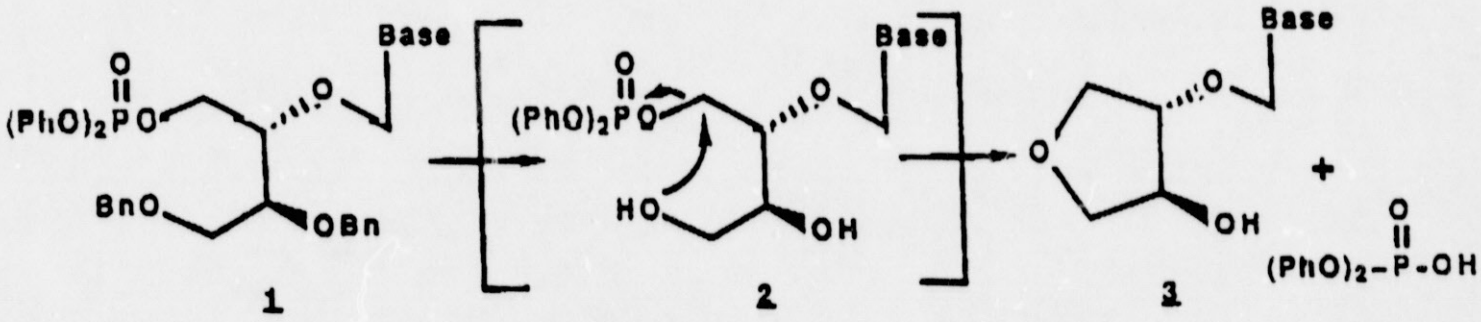
B. Summary: Synthetic methodologies for the preparation of 1',2'-*seco*-nucleoside phosphates and phosphonates have been developed. 5'-Phosphates as well as 3',5'-cyclic phosphates have been synthesized.

A unique reaction was discovered that allows the preparation of phosphonate chirons. The latter have been successfully converted to 1',2'-*seco*-nucleoside phosphonates and homophosphonates.

C. Discussion:

1. Phosphate esters:

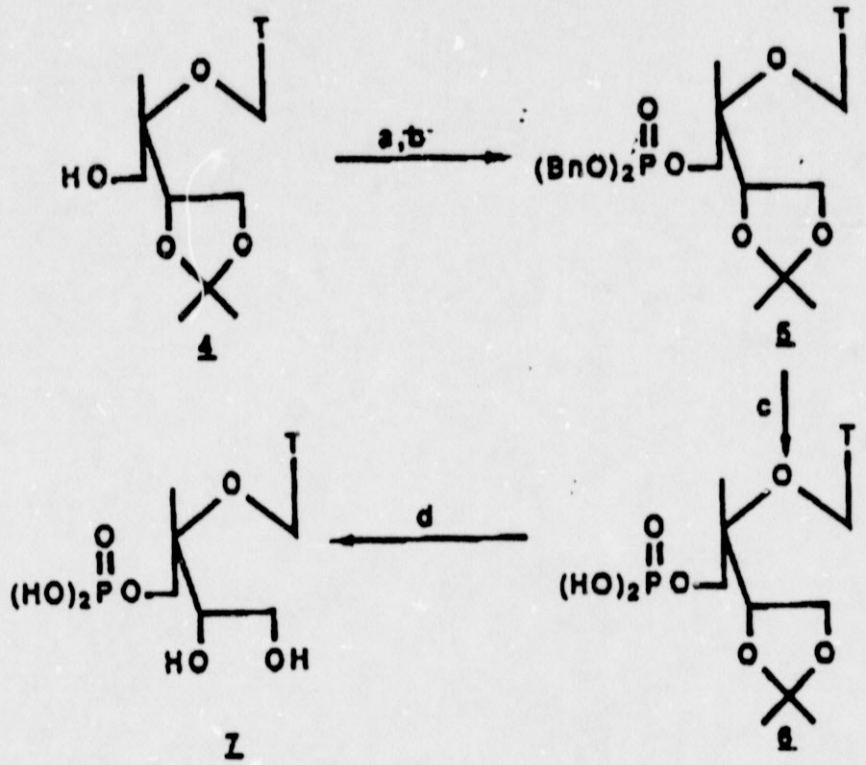
a. 5'-*seco*-nucleotides: Previous efforts in this area indicated that while such compounds are routinely made in the 2-deoxy series, such was not the case in the corresponding 2'-oxyanalogs. Deblocking of the 2'-hydroxyl groups resulted interestingly in a spontaneous dephosphorylation reaction to give 3.



This reaction was shown to proceed through intermediate 2, and the structure of product 3 was proved by an independent synthesis.[1]

Alternate routes for the preparation of these nucleotides were therefore explored. A successful attempt in the 4'*S* series is illustrated in Scheme 1.

SCHEME 1

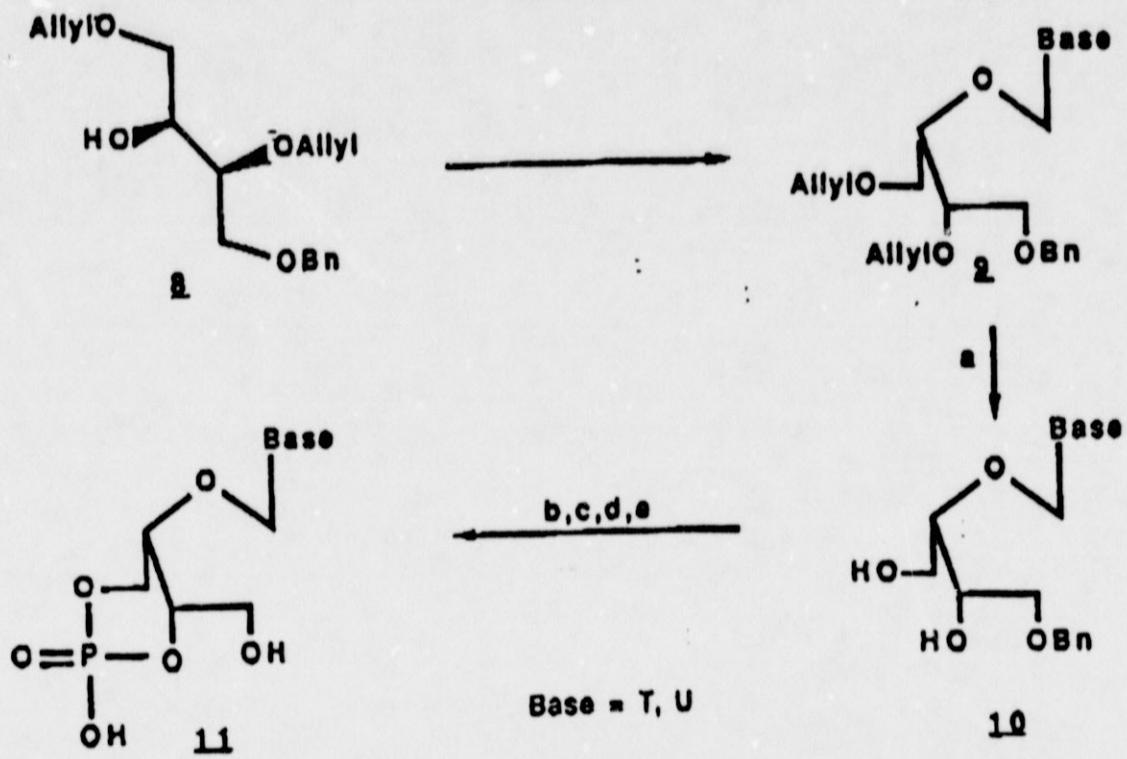


a = $(\quad)_2$N–P–$(OBn)_2$, tetrazole; b = mCPBA; c = H_2, 10% Pd / C; d = HCl, MeOH

Phosphorylation of 4 by the phosphoramidate method[2] followed by oxidation furnished 5 which was deblocked by hydrogenolysis. Methanolic HCl cleavage of the isopropylidene group gave nucleotide 7. Thus, dephosphorylation was avoided by sequential deblocking of the phsophate ester followed by the hydroxyl groups. Having developed this pathway, it is now feasible to apply the same methodology in the 4'*R* series to prepare target compounds.

b. 1',2'-*seco*-nucleosides 3',5'-cyclic phosphates: A versatile method for the preparation of these compounds has been developed and is outlined in Scheme 2.

SCHEME 2



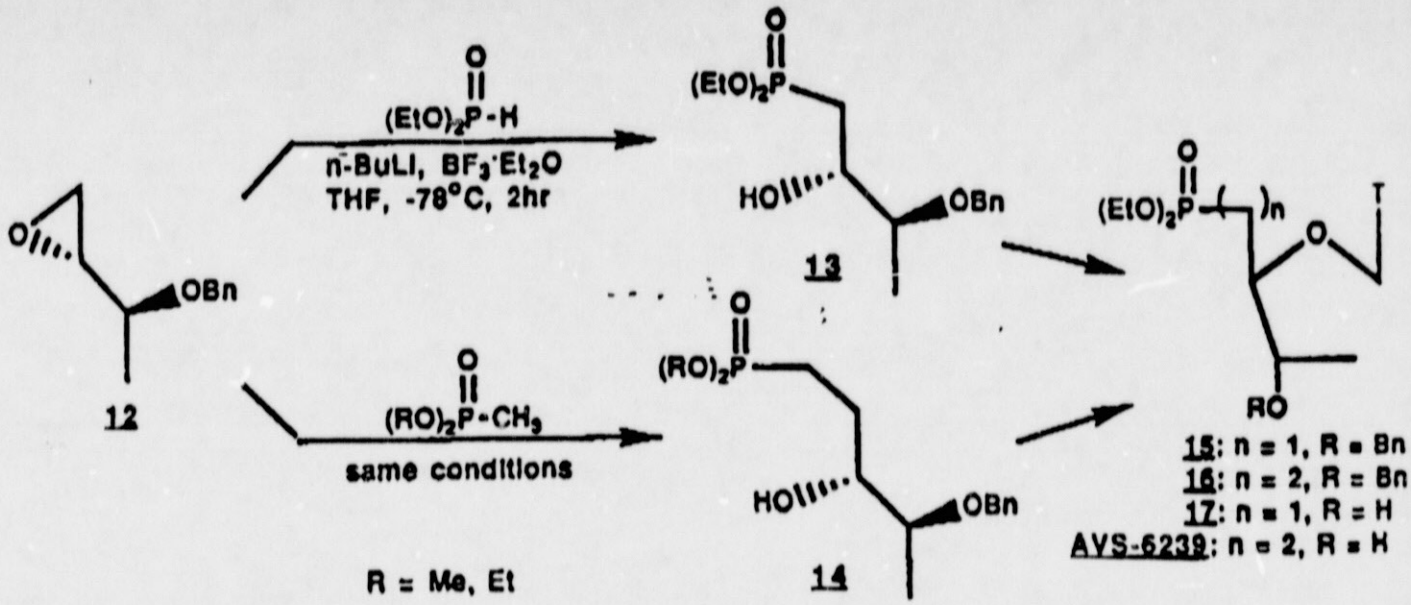
a = 10% Pd / C, p-TSOH; b = $Cl_2P(O)O$-C_6H_4-NO_2, pyridine; c = NH_4OH, dioxane;

d = H^+ neutralization; e = 10% Pd / C, cyclohexene

Chloromethylation of 8 followed by treatment with a persilylated base furnished 9 which was deallylated to give 10. Cyclic phosphorylation with p-nitrophenyl-phosphorodichloridate, hydrolysis of the p-nitrophenyl ester with ammonium hydroxide, neutralization, and deblocking furnished the cyclic nucleotide 11. Here again, expertise developed in the 4'*S* series can be applied to prepare the target 4'*R* diastereomers.

2. Phosphonate and homophosphonate esters: A novel and unique reaction involving $BF_3 \cdot Et_2O$ catalyzed opening of epoxides with anions of diethylphosphite and dialkylmethanephosphonates has been developed. Thus phosphonate chirons containing four and five carbon atoms have been prepared as shown in Scheme 3.

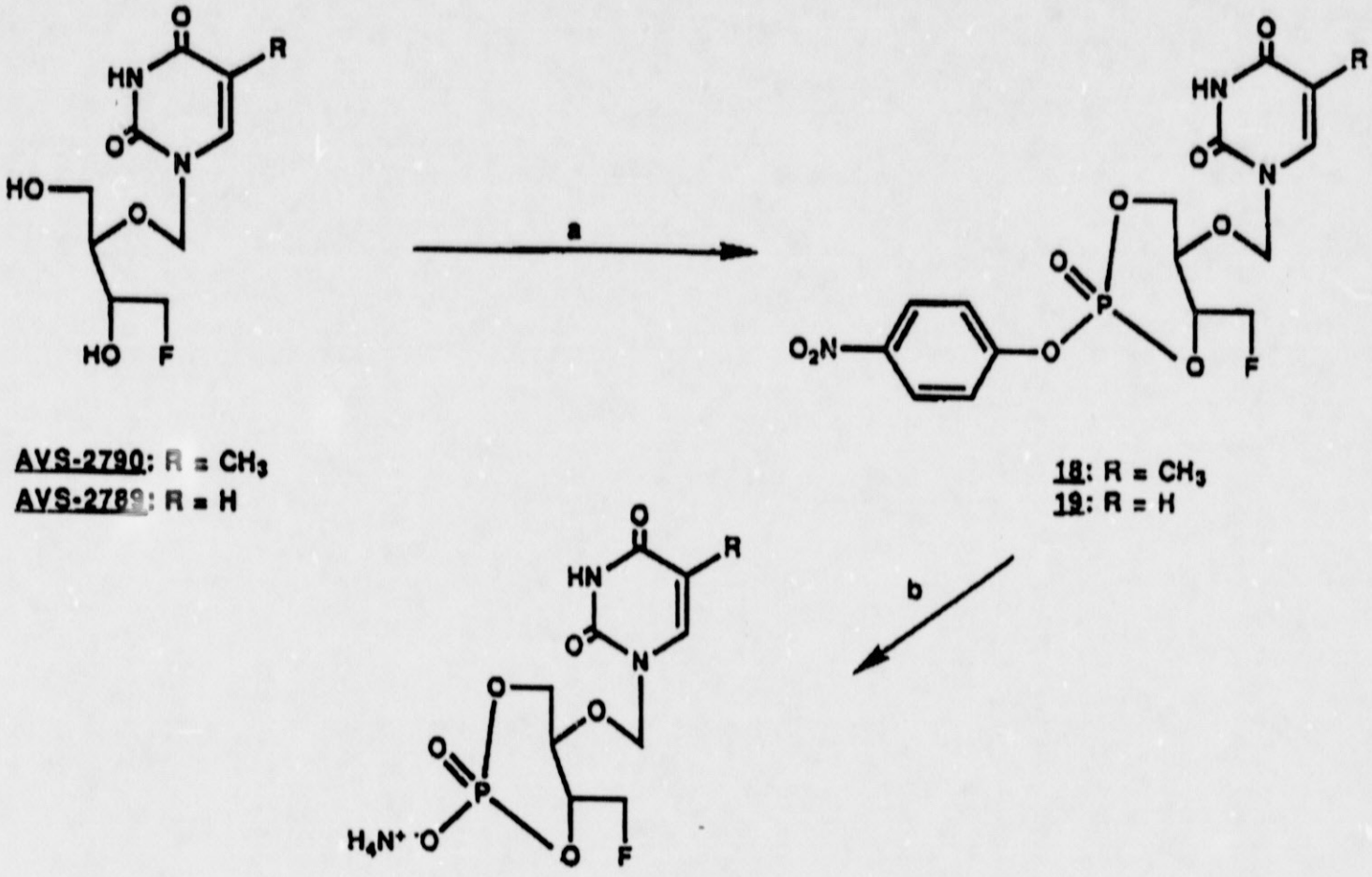
SCHEME 3



After several attempts which were outlined in the third annual report of contract DAMD17-86-C-6012, successful reaction conditions were discovered that allowed the preparation of chirons 13 and 14.

In a recent publication[3] Tanaka et. al., reported a regiospecific ring opening of an oxetane using diethylmethanephosphonate, n-BuLi, and five equivalents of $BF_3 \cdot Et_2O$. Application of these conditions to compound 12 using diethylphosphite and dialkylmethanephosphonate resulted in a quantitative formation of compounds 13 and 14, respectively. These chirons have been smoothly converted to the target thymidine phosphonate and homophosphonate 17and AVS-6239.

SCHEME 4



AVS-2790: R = CH_3
AVS-2789: R = H

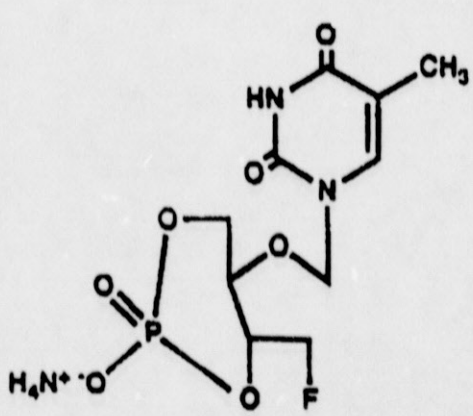
18: R = CH_3
19: R = H

b

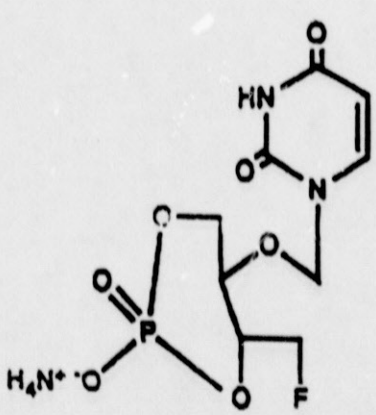
AVS-6237: R = CH_3
AVS-6238: R = H

a = O_2N-Ph-O-P(O)Cl_2, Pyr., CH_3CN; b = Conc. NH_4OH, p- Dioxane

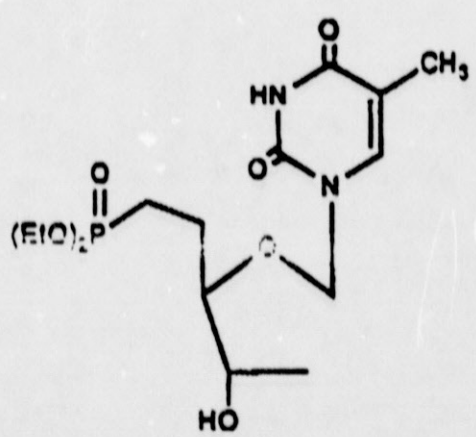
COMPOUNDS SUBMITTED



AVS-6237
Amount: 146mg



AVS-6238
Amount: 100mg



AVS-6239
Amount: 142mg

Experimental:

1- [[(4-Hydroxy)-3-pentoxy]methyl] thymine-1'-diethyl phosphonate (AVS-6239)

A mixture of the benzyl ether (16) (0.396 g, 0.846 mmol), 20% $Pd(OH)_2/C$ (0.310g) and cyclohexene (4.6 ml) in absolute ethanol (9ml) was stirred and heated at reflux for 2 h. The reaction mixture was then filtered and the catalyst was washed with warm ethanol. The filtrate and the wash were combined, concentrated under reduced pressure and chromatographed over silica gel column (ethyl acetate) to furnish the alcohol (AVS-6239) (0.276g, 86%) as a gum: ^1H NMR ($CDCl_3$): δ 0.96-1.42 (overlapping d and t, 9H), 1.44-2.12 (overlapping s and m, 7H), 3.2-4.24 (m, 7H), 5.18 (bs, 2H), 7.16 (bs, 1H) and 10.05 (bs, 1H, D_2O exchangeable).

Anal. Calcd. for $C_{15}H_{27}N_2O_7P$: C, 47.62; H, 7.19; N, 7.40
Found C, 47.54; H, 7.27; N, 7.47

1-[(1,3S-Dihydroxy-4-fluoro-2R-butoxy)methyl]thymine-1',3'-cyclic-*p*-nitrophenyl phosphate(18)

To a stirred solution of diol (AVS-2790) (0.814g. 3.1 mmol) in dry acetonitrile (25ml) was added dry pyridine (3 ml) and *p*-nitrophenyl phosphorodichloridate (1.2g, 4.65 mmol). This mixture was stirred at room temperature for 24 h with the exclusion of moisture. The excess solvents were removed in <u>vacuo</u>, the resulting residue was dissolved in CH_2Cl_2 (75 ml), and the organic layer was washed successively with water (25 ml), saturated sodium chloride solution (25 ml), and water (25 ml). After drying over anhydrous $MgSO_4$, the organic layer was evaporated under diminished pressure and the residue was dissolved in a minimal amount of ethyl acetate and applied to a silica gel column. Elution of the column with ethyl acetate provided the title compound (0.910g, 66%) as a white crystalline solid: m.p. 179-181°C (dec) ^1H NMR ($CDCl_3$): δ 1.9 (s, 3H, CH_3), 4.06-5.3 (m, 8H), 9.56 (br s, 1H).

Anal. calcd. for $C_{16}H_{17}FN_3O_9$: C, 43.16; H, 3.85; F, 4.26; N, 9.44
Found C, 43.43; H, 3.99; F, 4.43; N, 9.17.

1-[(1,3S-Dihydroxy-4-fluoro-2R-butoxy)methyl]thymine-1',3'-cyclic phosphate ammonium salt (AVS-6237)

The product obtained from the previous experiment (0.665 g, 1.5 mmol) was dissolved in *p*-dioxane (30ml) and to this solution was added concentrated ammonium hydroxide (2.6 ml, 3 mmol.). After stirring overnight at room temperature, the excess solvents were removed in vacuo and the resulting residue was purified by silica gel column chromatography. The column was eluted with ethyl acetate-methanol (7:3) to give AVS-6237 (0.25g, 49%) as a white solid: m.p. 150-155°C (dec); ^{1}H NMR (D_2O): δ 1.84 (s, 3H, CH_3), 3.9-5.36 (m, 13H), 7.48 (s, 1H).

Anal. Calcd. for $C_{10}H_{17}FN_3O_7$: C, 35.20; H, 5.02; N, 12.32.
Found: C, 35.02; H, 5.28; N, 12.07.

1-[(1,3S-Dihydroxy-4-fluoro-2R-butoxy)methyl]uracil-1',3'-cyclic-*p*-nitrophenyl phosphate (19).

Similar experimental conditions as described for 18 were used to prepare 19. Thus, diol (AVS-2789) (1.2g, 4.84 mmol), dry pyridine (5ml), p-nitrophenyl phosphorodichloridate (1.86 g, 7.26 mmol) in dry acetonitrile (20mL) afforded, after work-up, 19 (1.45g, 69.5%) as a crystalline solid: m.p. 147-152°(dec); ^{1}H NMR (DMSO-d_6): δ 4.12-5.4 (m, 8H), 5.68 (d, J=7Hz, 1H), 7.4-7.84 (m, 3H), 8.3 (d, J=9Hz, 2H), 11.4 (br s, 1H, NH).

Anal. Calcd. for $C_{15}H_{15}FN_3O_9$: C, 41.78; H, 3.50; N, 9.74.
Found: C, 41.92; H, 3.50; N, 9.60.

1- [(1,3S-Dihydroxy-4-fluoro-2R-butoxy)methyl] uracil- 1',3'- cyclic phosphate ammonium salt (AVS-6238)

Similar experimental conditions as described for AVS-6237 were used to prepare AVS-6238. A solution of the triester (19) (1.18g, 2.7 mmol) in p-dioxane (50 ml) was treated with concentrated ammonium hydroxide (6.4 ml, 5.5 mmol) and after work-up furnished AVS-6238 (0.72g, 80%) as a white solid: m.p. 180-186° (dec); ^{1}H NMR (D_2O): 3.6-5.4 (m, 13H) , 5.82 (d, 1H, J= 8Hz), 7.68 (d, 1H, J=8Hz).

Anal. Cald. for $C_9H_{15}FN_3O_7$: C, 33.04; H, 4.62; N, 12.32
Found: C, 33.23; H, 4.94; N, 12.51.

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