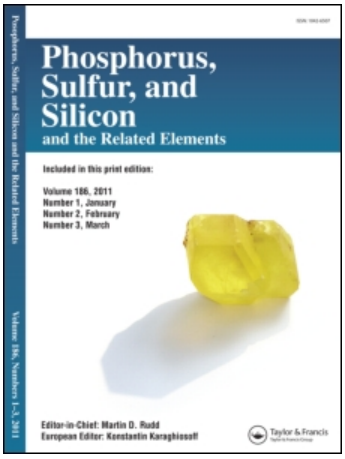

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SYNTHESIS OF PHENYL OR 4-NITROPHENYL METHYL β-KETOPHOSPHONATE ESTERS

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Seven new aryl methyl β-ketophosphonate esters were synthesized. The hydrolytic rates of the compounds under physiological conditions were studied. Most of the compounds are effective inhibitors of acetylcholinesterase. The enzyme recovers on the 10–50 h time scale from its adducts with two of the inhibitors.

Keywords: Aryl alkylphosphonate esters; ketoalkyl aryl phosphonate esters; reversible inhibitors; serine hydrolase inhibitors

INTRODUCTION

4-Nitrophenyl 4-substituted-phenacyl methylphosphonate (PMN) esters were synthesized[1–2] and studied in this laboratory for their propensity to reversibly inhibit a wide range of serine hydrolase enzymes.[2–8] The compounds inhibit cholinesterases,[8] and a broad range of serine proteases[2–7] with good to modest stereoselectivity and enzyme activity can be recovered in several hours. The para substituents in the phenacyl group modulate the rate of recovery. These studies have now been extended to analogues of the PMN type of compounds; seven new compounds were synthesized.

METHODS AND RESULTS

The Synthesis of Intermediates

General Procedure

Equimolar quantities of phenol/4-nitrophenol and TEA were dissolved in toluene and added dropwise to methylphosphonic dichloride

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in toluene at 0°C. After the addition, the temperature of the reaction mixture was slowly raised to r.t. and refluxed for 8 h. The precipitated triethylamine hydrochloride was filtered off and the solvent was evaporated under reduced pressure. The crude product was distilled under vacuum to get pure compound.

Phenyl Methylphosphonochloridate

Phenol (3.54 g, 0.0376 mol) and TEA (5.24 mL, 0.0376 mol) in 20 mL of toluene were added to methylphosphonic dichloride (5.0 g, 0.0376 mol) in 20 mL of toluene. Liquid at r.t., distilled at 89–90°C/0.05 mm Hg, yield 5.2 g (72%). ^{1}H NMR ($CDCl_3$): δ 2.09 (d, J 17, 3H, P—CH_3) and 7.2–7.4 (m, 5H, Ar—H).

4-Nitrophenyl Methylphosphonochloridate

4-Nitrophenol (5.23 g, 0.0376 mol) and TEA (5.24 mL, 0.0376 mol) in 20 mL of toluene were added to methylphosphonic dichloride (5.0 g, 0.0376 mol) in 20 mL of toluene; distilled at 120–121°C/0.04 mm Hg; yield 5.4 g (76%); solid at r.t., mp: 75–76°C. ^{1}H NMR ($CDCl_3$): δ 2.29 (d, J 17, 3H, P—CH_3), 7.39 (d, J 9, 2H, Ar—H) and 7.52 (d, J 9, 2H, Ar—H).

Phenyl/4-Nitrophenyl methyl β-ketophosphonate Esters

R= H, NO_2; R_1 = CH_3, C_6H_5; R_2 = H, CH_3, C_6H_5 and R_3 = H, CH_3.

General Procedure

Equimolar quantities of α-keto alcohol (0.005 mol) and TEA (0.7 mL, 0.005 mol) in 10 mL of benzene were added to phenyl/4-nitrophenyl methyl-phosphonochloridate (0.005 mol) in 10 mL of benzene at 0°C. After the addition, the temperature was slowly raised to 50°C and stirring was continued for 24 h. The precipitated triethylamine hydrochloride was filtered off and solvent was evaporated under reduced pressure. The crude product was purified on a silica gel column using a chloroform-acetone 95:5 mixture as eluent.

***Phenacyl methyl phenylphosphonate* (PMP);** yield 66%. ^{1}H NMR ($CDCl_3$): δ 1.95 (d, J 17, 3H, P—CH_3), 5.28 (q, J 17, 1H, CH_2) 5.52 (q, J 17, 1H, CH_2) and 7.15–7.89 (m, 10H, Ar—H).

***Acetol methyl phenylphosphonate* (AMP);** yield 63%. ^{1}H NMR ($CDCl_3$): δ 1.76 (d, J 18, 3H, P—CH_3), 2.10 (s, 3H, CH_3) 4.52

(q, *J* 17, 1H, CH_2) 4.73 (q, *J* 17, 1H, CH_2) and 7.13–7.34 (m, 5H, Ar—H).

***Methylacetoin methyl phenylphosphonate* (MAMP);** yield 67%. ^{1}H NMR ($CDCl_3$): δ 1.58 (d, *J* 8, 6H, $(CH_3)_2$), 1.72 (d, *J* 18, 3H, P—CH_3), 2.24 (s, 3H, CH_3) and 7.16–7.37 (m, 5H, Ar—H); ^{13}C NMR: 13.3 (d, J_{P-C} 147, *P—CH*$_3$), 24.5 (s, CH_3), 25.2 (d, J_{P-C} 3, CH_3), 25.6 (d, J_{P-C} 5, *C*H_3), 87.4 (d, J_{P-C} 8, *C*$(CH_3)_2$), 120.7 (d, *J* 4, 2C, Ar—C2&6), 125.0 (s, 1C, Ar—C4), 129.6 (s, 2C, Ar—C3&5) and 150.2 (d, *J* 8, 1C, Ar—C1).

***Benzoin methyl phenylphosphonate* (BMP);** yield 63%. ^{1}H NMR ($CDCl_3$): δ 1.96 (d, *J* 17, 3H, P—CH_3), 6.89 (d, *J* 7, 1H, CH) and 7.24–8.19 (m, 15H, Ar—H).

***Acetol methyl 4-nitrophenylphosphonate* (AMN);** yield 63%. ^{1}H NMR ($CDCl_3$): δ 1.78 (d, *J* 19, 3H, P—CH_3), 2.12 (s, 3H, CH_3), 4.58 (q, *J* 18, 1H, CH_2) 4.76 (q, *J* 18, 1H, CH_2), 7.42 (d, *J* 8, 2H, Ar—H) and 8.34 (d, *J* 8, 2H, Ar—H).

***Methylacetoin methyl 4-nitrophenylphosphonate* (MAMN);** yield 67%. ^{1}H NMR ($CDCl_3$): δ 1.61 (d, *J* 14, 6H, $(CH_3)_2$), 1.86 (d, *J* 18, 3H, P—CH_3), 2.26 (s, 3H, CH_3), 7.41 (d, *J* 9, 2H, Ar—H) and 8.27 (d, *J* 9, 2H, Ar—H).

***Benzoin methyl 4-nitrophenylphosphonate* (BMN).** The reaction was successful, but the compound was very unstable and decomposed during purification on column.

Determination of Pseudo-First-Order Hydrolysis Rate Constants

(Table I) Hydrolytic rates were measured for phosphonate/thiophosphonate esters at buffer concentrations 0.03–0.1 M phosphate, or 0.01 M sodium acetate. Five μL of $1 - 3 \times 10^{-3}$ M acetonitrile stock solutions of the compounds were mixed in 1000 μL of buffer that had been preequilibrated at 25.0 ± 0.1°C in a quartz cell in the

TABLE I First-Order Hydrolysis Rate Constants of the Phosphonates in Phosphate Buffers at 25.0 ± 0.1°C

Phosphonate	Buffer	M	pH	10^4 k_h, s^{-1}
PMN	Phosphate	0.10	7.7	6.4 ± 0.6
PMN-S	Phosphate	0.045	7.4	14.1 ± 0.1
PMP	Tris	0.02	7.0	11.43 ± 0.05
	Phosphate	0.037	7.6	34.4 ± 0.3
	Tris	0.02	8.3	90.0 ± 0.7
MAMP	Phosphate	0.045	7.4	5.26 ± 0.05
MAMN	Phosphate	0.05	7.2	7.10 ± 0.10
BMP	Phosphate	0.045	7.4	6.18 ± 0.13

cell compartment of a Perkin Elmer Lambda-6 Spectrophotometer interfaced to a PC. The release of phenol/4-nitrophenol was monitored at 275 and 400 nm, respectively, for four half-lives.

CONCLUSIONS

The seven new compounds were successfully synthesized and characterized. They hydrolyze rapidly at pH 7.0, most probably, with intramolecular catalysis, by the hydrated ketone. Nevertheless, MAMP and BMP inhibit eel actylcholinesterase, and the enzyme recovers in 10–50 h at pH 7.0 and 25.0°C.

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REFERENCES

[1] C. N. Lieske, J. W. Howanec, G. M. Steinberg, J. N. Piculin, W. J. Lennox, A. B. Ash, and P. Blumbergs, *J. Agric. Food Chem.*, **17,** 255–258 (1969).
[2] I. M. Kovach and L. McKay, *Biomed. Chem. Lett.*, **2,** 1735 (1992).
[3] I. M. Kovach, Q. Zhao, M. Keane, and R. Reyes, *J. Amer. Chem. Soc.*, **115,** 10471–10476 (1993).
[4] Q. Zhao, I. M. Kovach, A. Bencsura, and A. Papathanassiu, *Biochemistry*, **33,** 8128–8138 (1994).
[5] Q. Zhao and I. M. Kovach, *Bioorg. Med. Chem.*, **4,** 523–529 (1996).
[6] E. J. Enyedy and I. M. Kovach, *Bioorg. Med. Chem.*, **5,** 1531–1541 (1997).
[7] E. J. Enyedy and I. M. Kovach, *Bioorg. Med. Chem.*, **8,** 549–556 (2000).
[8] C. Viragh, L. Zaslavskaia, I. M. Kovach, and C. Broomfield, *Bioscience Review* (2000).