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Syntheses, Characterizations, and Crystal Structures of Phosphonopeptides

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ABSTRACT: *α-Aminophosphonic acids and their derivatives, as phosphorus analogs of amino acids, have attracted much attention as they show a range of biological activities. In this paper, dialkyl phenyl(4-pyridylcarbonylamino)methylphosphonates were synthesized via the Mannich reaction (Yuan et al., Synthesis 1990, 3, 256) and peptide coupling. Their structures were confirmed by elemental analysis, IR, ^{1}H NMR, ^{13}C NMR, and MS. X-ray diffraction data of compounds (***2a, 2b, 2c****) were reported respectively. The antibacterial and antitumor activities of these compounds are first reported in this paper.*
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INTRODUCTION

The importance of α-aminophosphonic acids and their derivatives is due to their diverse bioactivities including herbicide [1], antibacterial [2], antiviral and antitumor [3], and their utilizations in a variety of pesticide and therapeutic areas [4]. The study is focused on the linkage of α-aminophosphonic acids and bioactive structure units in order to find novel phosphonopeptides and their derivatives with higher bioactivity and low toxicity. Numerous methods have been reported for the preparation of amides. The most typical procedures are the Yasutsugu Shimonishi method, the *N*-carboxyl anhydride (NCA) method, and the DCC-HOSu method [5–8]. Organic phosphorus coupling reagents [9–11] also have been applied for the synthesis of amides, such as the Appel [12] coupling reagents (triphenylphosphine and hexachloroethane). Hence, it is an easy, facile synthetic route for the coupling of amides. X-ray diffraction structures of compounds **2a, 2b, 2c** were determined. In the solid state, for each **2a, 2b, 2c** there is an intermolecular hydrogen bond (N—2···O—2) with lengths 2.949(3) Å, 2.891(3) Å, and 2.919(5) Å, respectively [13]. Bioassay showed that most of them are bioactive. For example, **2a, 2b, 2c** exhibited moderate cytotoxicity toward the KB cell line (IC_{50} values are 114.1 μg/mL, 68.5 μg/mL, and 51.8 μg/mL, respectively). Bioactivities increased with the bulk of the alkyl group.

EXPERIMENTAL

Materials and Methods

Triphenylphosphine, hexachloroethane, isonicotic acid, and phosphorus trichloride were commercially

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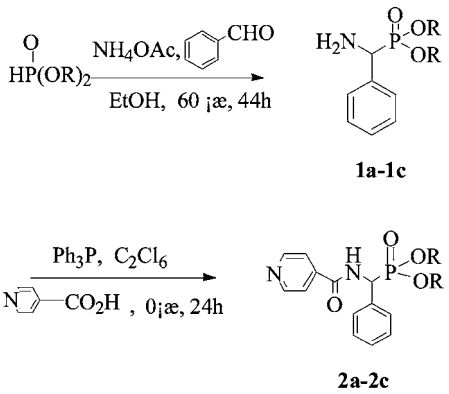
available (they were obtained from Aldrich and were used without further purification). The melting points were obtained with Yanaco micromelting point apparatus and are uncorrected. Infrared-spectra were recorded on a Nicolet AVATAR 360 FT-IR spectrophotometer using KBr disks. ^{1}H, ^{13}C, and ^{31}P NMR spectra were recorded on a Varian 500 MHz spectrometer operating on 500, 125, 202 MHz, respectively. The chemical shifts were reported in ppm with respect to the references and were stated relative to external tetramethylsilane (TMS) for ^{1}H and ^{13}C NMR, and to 85% phosphoric acid for ^{31}P NMR. Elemental analyses were performed with a Flash EA 1112. All mass spectra were acquired with a Bruker ESQUIRE-3000 plus ion trap spectrometer equipped with a gas nebulizer probe in the positive ion mode, microplate reader (M-3550, Bio-Rad) at 595 to 655 nm as reference. A Bruker SMART CCD X-ray diffractometer was used.

Syntheses

*General Procedure for the Preparation of the Hydrochloride of Dialkyl α-Aminobenzylphosphonate (***1a–1c***).* To the EtOH solution of ammonium acetate (7.70 g, 0.10 mol) was added actively molecular sieves (4 Å) (2.0 g), benzaldehyde (10.61 g, 0.10 mol), and dialkyl phosphite (0.10 mol) at room temperature. The reaction mixture was stirred at 60°C for 44 h and cooled to room temperature. The reaction mixture was acidified to pH 1 with HCl, and the solution was washed with Et_2O to remove neutral materials. The aqueous phase was then adjusted to pH 11 with aq. NaOH, and the product was extracted with CH_2Cl_2. The solvent was removed to give the crude product as pale yellow oil, which was further treated with HCl (gas) in EtOH (10 mL)–Et_2O (10 mL) to afford the hydrochloride of dialkyl α-aminobenzylphosphonate (**1a–1c**) as a white crystalline material [14–16].



Where: R = Me (a); Et (b); iPr (c)

SCHEME 1

Dimethyl α-Aminobenzylphosphonate Hydrochloride **1a**: White crystals, yield 37.4%, mp 220.6–223.5°C; ^{1}H NMR (D_2O) δ: The NH_2 signal disappeared with D_2O exchange, 7.44–7.64 (m, 5H, ArH), 5.08 (d, 1H, $J_{P\text{-}CH} = 18$ Hz, CH), 3.78 (s, 6H, $2OCH_3$); IR (KBr) ν: 3425 (N-H), 1245 (P=O), 1033 (P—O—C), 1604, 1521, 1494, 1456 (aromatic vibrations) (cm^{-1}); MS *m/z* (%): 106.1 (10.64), 215.9 ($[M + H]^+$, 21.6), 430.7 ($[2M + H]^+$, 100). Anal. Calcd for $C_9H_{14}NO_3P{\cdot}HCl$: C 42.96, H 6.01, N 5.57; Found C 42.81, H 6.05, N 5.48.

Diethyl α-Aminobenzylphosphonate Hydrochloride **1b**: White crystals, yield 36.8%, mp 176.3–177.6°C; ^{1}H NMR (D_2O) δ: The NH_2 signal disappeared with D_2O exchange, 7.44–7.56 (m, 5H, ArH), 4.96 (d, 1H, $J_{(P\text{-}CH)} = 18$ Hz, CH), 4.16 (d, $J_{(CH_2\text{-}CH_3)} = 8$ Hz, 4H, $2C\mathbf{H}_2CH_3$), 1.26 (d, $J_{(CH_2\text{-}CH_3)} = 8$ Hz, 6H, $2CH_2C\mathbf{H}_3$); IR (KBr) ν: 3438 (N-H), 1239 (P=O), 1024 (P—O—C), 1605, 1520, 1498, 1455 (aromatic vibrations) (cm^{-1}); MS *m/z* (%): 106.1 (44.21), 243.9 ($[M + H]^+$ 95.44), 486.7 ($[2M + H]^+$, 100). Anal. Calcd for $C_{11}H_{18}NO_3P{\cdot}HCl$: C 47.24, H 6.85, N 5.01; found C 47.10, H 6.77, N 4.76.

Diisopropyl α-Aminobenzylphosphonate Hydrochloride **1c**: White crystals, yield 33.7%, mp 171.7–173.2°C ^{1}H NMR (D_2O) δ: The NH_2 signal disappeared with D_2O exchange, 7.44–7.64 (m, 5H, ArH), 4.92 (d, 1H, $J_{(P\text{-}CH)} = 18$ Hz, CH), 4.68 (d, $J_{CH\text{-}CH_3} = 6$ Hz, 2H, $2C\mathbf{H}(CH_3)_2$), 1.26 (d, $J_{(CH\text{-}CH_3)} = 6$ Hz, 12H, $2CH(C\mathbf{H}_3)_2$); IR (KBr) ν: 3431 (N-H), 1246 (P=O), 1019 (P—O—C), 1595, 1562, 1516, 1457 (aromatic vibrations) (cm^{-1}); MS *m/z* (%): 106.1 (17.16), 271.9 ($[M + H]^+$, 66.53), 544.5 ($[2M + H]^+$, 100). Anal. Calcd for $C_{13}H_{22}NO_3P{\cdot}HCl$: C 50.74, H 7.53, N 4.55; found C 50.51, H 7.56, N 4.39.

*General Procedure for the Preparation of Dialkyl Phenyl(4-pyridylcarbonylamino)methylphosphonate (***2a–2c***).* Triphenylphosphine (3.93 g, 15 mmol) and hexachloroethane (3.58 g, 15 mmol) were dissolved in 1,2-dichloroethane (20 mL) under nitrogen atmosphere for 1 h. The reacted solution was added dropwise to a mixture of the dialkyl α-aminobenzylphosphonate hydrochloride (2.79 g, 10 mmol) and isonicotinic acid (1.23 g, 10 mmol) in 1,2-dichloroethane (90 mL) and 4 mL of triethylamine. After 24 h the reaction was completed. The reaction mixture was acidified to pH 1 with HCl, and the solution was washed with Et_2O to remove neutral materials. The aqueous phase was then adjusted to pH 11 with aq. NaOH, and the product was extracted with CH_2Cl_2; the solvent was removed

to give the crude product, which was purified by recrystallization [17,18].

Dimethyl Phenyl(4-pyridylcarbonylamino)methylphosphonate **2a**: It was recrystallized from hot ethanol to give white crystals, yield 76.5%, mp 134.1–135.5°C ^{1}H NMR ($CDCl_3$) δ: 9.72 (dd, $J_{(NH\text{-}CH)} = 9$ Hz, NH,), 8.38–8.68 (m, 4H, Py), 7.20–7.60 (m, 5H, ArH), 5.42 (dd, 1H, $J_{(P\text{-}CH)} = 21$ Hz, $J_{(NH\text{-}CH)} = 9$ Hz, CH), 4.02 (s, 6H, $2OCH_3$); ^{13}C NMR ($CDCl_3$) δ: 149.44, 123.94, 145.22 (Py), 133.69, 128.90, 127.25, 128.73 (Ar), 168.28 (C═O), 52.39 (CH), 55.55, 53.93 ($2OCH_3$); IR (KBr) ν: 3402 (N-H), 1662 (C═O), 1225 (P═O), 1074 (P─O─C), 1557, 1511, 1495, 1453 (the vibration of aromatic) (cm^{-1}); MS *m/z* (%): 210.9 (19.48), 321 ($[M + H]^+$, 100), 641.0 ($[2M + H]^+$, 1.01); ^{31}P ($CDCl_3$) δ: 12.8 ppm. Anal. Calcd for $C_{15}H_{17}N_2O_4P{\cdot}H_2O$: C 53.26, H 5.66, N 8.28; found C 53.55, H 5.54, N 8.09.

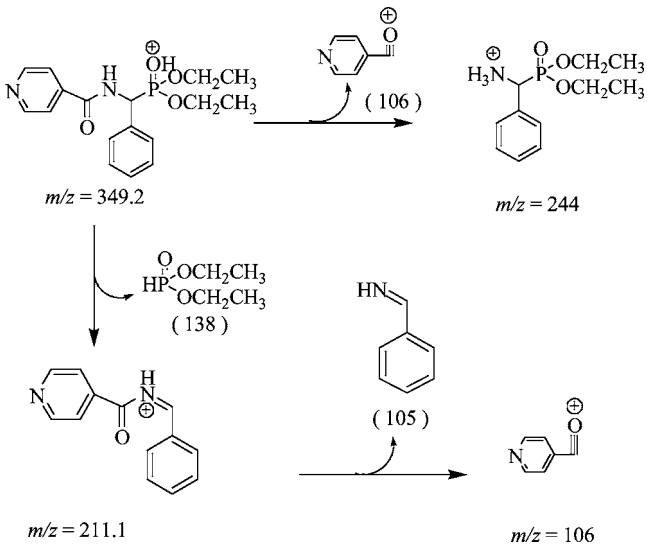
Diethyl Phenyl(4-pyridylcarbonylamino)methylphosphonate **2b**: It was recrystallized from a 1:1 mixture of hexane and dichloroethane to give white crystals, yield 82.0%, mp 107.0–108.1°C; ^{1}H NMR ($CDCl_3$) δ: 8.23 (dd, $J_{(NH\text{-}CH)} = 9$ Hz, NH), 8.08–8.28 (m, 4H, Py), 7.23–7.62 (m, 5H, ArH), 5.76 (dd, 1H, $J_{(P\text{-}CH)} = 21$ Hz, $J_{(CH\text{-}NH)} = 9$ Hz CH), 3.98 (t, $J_{(CH_2\text{-}CH_3)} = 8$ Hz, 4H, $2C\mathbf{H}_2CH_3$), 1.22 (d, $J_{(CH_2\text{-}CH_3)} = 8$ Hz, 6H, $2CH_2C\mathbf{H}_3$); ^{13}C NMR ($CDCl_3$) δ: 150.51, 121.19, 140.84 (Py), 134.64, 128.47, 128.17, 128.78 (Ar), 165.09 (C═O), 51.40 (CH), 63.72, 63.20 ($2\mathbf{C}H_2CH_3$), 16.44, 16.10 ($2CH_2\mathbf{C}H_3$); IR (KBr) ν: 3440 (N-H), 1664 (C═O), 1244 (P═O), 1032 (P─O─C), 1596, 1546, 1500, 1456 (aromatic vibrations) (cm^{-1}); MS *m/z* (%): 211.0 (17.77), 348.9 ($[M + H]^+$, 100), 696.7 ($[2M + H]^+$, 10.16); ^{31}P ($CDCl_3$) δ: 19.5 ppm. Anal. Calcd for $C_{17}H_{21}N_2O_4P$: C 58.62, H 6.08, N 8.04; found C 58.44, H 6.10, N 7.95.

Diisopropyl Phenyl(4-pyridylcarbonylamino)-methylphosphonate **2c**: It was recrystallized from a 1:2 mixture of acetic ether and petroleum ether to give white crystals, yield 85.1%, mp 134.3–135.7°C; ^{1}H NMR ($CDCl_3$) δ: 7.94 (dd, $J_{(NH\text{-}CH)} = 9$ Hz, NH), 8.05–8.29 (m, 4H, Py), 7.18–7.72 (m, 5H, ArH), 5.72 (dd, 1H, $J_{(P\text{-}CH)} = 21$ Hz, $J_{(NH\text{-}CH)} = 9$Hz, CH), 4.50 (d, $J_{(CH\text{-}CH_3)} = 6$ Hz, 2H, $2C\mathbf{H}(CH_3)_2$), 1.20 (d, $J_{(CH\text{-}CH_3)} = 6$ Hz, 12H, $2CH(C\mathbf{H}_3)_2$); ^{13}C NMR ($CDCl_3$) δ: 150.26, 121.42, 141.15 (Py), 135.13, 128.61, 128.15, 128.65 (Ar), 165.39 (C═O), 51.27 (CH), 72.48, 71.96 ($2\mathbf{C}H(CH_3)_2$), 24.12, 24.01, 23.86, 23.81 ($2CH(\mathbf{C}H_3)_2$); IR (KBr) ν: 3448 (N-H), 1676 (C═O), 1231 (P═O), 1009 (P─O─C), 1661, 1600, 1537, 1491 (aromatic vibrations) (cm^{-1}); MS *m/z* (%): 211.0 (40.38), 377.0 ($[M + H]^+$, 100), 752.8 ($[2M + H]^+$, 22.63); ^{31}P ($CDCl_3$) δ: 19.9 ppm. Anal. Calcd for $C_{19}H_{25}N_2O_4P$: C 60.63, H 6.69, N 7.44; found C 60.49, H 6.74, N 7.33.

RESULTS AND DISUSSION

Mass Spectroscopy

The main mass spectrometric data of compounds in Table 1 showed $[M + H]^+$ and $[2M + H]^+$ signals. Mass spectrometry of the **2a, 2b, 2c** produced two kinds of fragment ions at *m/z* 211 and 106. Scheme 2 shows fragmentation pathways of diethyl phenyl(4-pyridylcarbonylamino)methylphosphonate (**2b**).



SCHEME 2

TABLE 1 The Main Mass Spectra Data of **1a–1c, 2a–2c**

Compounds	*m/z (%)*			
1a	106.1 (10.64)	215.9 ($M + H^+$, 21.60)	430.7 ($2M + H^+$,100)	237.7 (87.23)
1b	106.1 (44.21)	243.9 ($M + H^+$, 95.44)	486.7 ($2M + H^+$,100)	267.0 (90.47)
1c	106.1 (17.16)	271.9 ($M + H^+$, 66.53)	544.5 ($2M + H^+$, 100)	294.1 (98.25)
2a	210.9 (19.48)	321.0 ($M + H^+$, 100)	641.0 ($2M + H^+$, 1.01)	215.6 (30.48)
2b	211.0 (17.77)	348.9 ($M + H^+$, 100)	696.7 ($2M + H^+$, 10.16)	244.0 (44.57)
2c	211.0 (40.38)	377.0 ($M + H^+$, 100)	752.8 ($2M + H^+$, 22.63)	271.8 (27.83)

TABLE 2 Crystal data and Refinement Details for compounds **2a, 2b**, and **2c**

	2a	**2b**	**2c**
CCDC number	248275	248276	248277
Empirical formula	$C_{15}H_{17}N_2O_4P$	$C_{17}H_{21}N_2O_4P$	$C_{19}H_{25}N_2O_4P{\cdot}H_2O$
Formula weight	320.28	348.33	394.38
Temperature (K)	293(2)	293(2)	293(2)
Crystal size	$0.21 \times 0.19 \times 0.15$	$0.35 \times 0.27 \times 0.26$	$0.10 \times 0.09 \times 0.20$
Crystal system	Monoclinic	Monoclinic	Triclinic
Space group	*P*(2)1/*n*	*C*2/*c*	*P*-1
Unit cell dimensions			
a (Å)	10.748(3)	23.714(5)	10.438(2)
b (Å)	14.417(4)	8.093(5)	13.738(3)
c (Å)	10.920(3)	20.019(5)	16.398(3)
α (°)	90.0	90.0	102.579(3)
β (°)	111.28(4)	110.325(5)°	105.028
γ (°)	90.0	90.0	93.650
Volume (Å^3)	1576.7(7)	3602(3)	2198.5(7)
Z	4	8	2
d (Mg/m^3)	1.349	1.285	1.185
μ(mm^{-1})	0.193	0.175	0.154
F(000)	672	1472	832
Diffractometor	Bruker APEX area-detector	Bruker APEX area-detector	Bruker APEX area-detector
Radiation	Monochromatize Mo Kα (0.7103)	Monochromatize Mo Kα (0.7103)	Monochromatize Mo Kα (0.7103p
θ range (deg)	2.28–25.0	1.83–25.0	1.53–25
Limiting indices	$-11 = h = 12$ $-17 = k = 16$ $-12 = l = 12$	$-28 = h = 28$ $-9 = k = 9$ $-23 = l = 23$	$-12 = h = 12$ $-16 = k = 16$ $-19 = l = 19$
Reflections collections	7844	12050	15980
Independent reflections	2757 [R(int) = 0.0234]	3162 [R(int) = 0.0240]	7695 [R(int) = 0.0269]
Data/restraints/ parameters	2757 / 0 / 199	3162/ 0 / 219	7985/ 0 / 478
Goodness-of-fit on F^2	1.182	1.05	1.070
R indices [$I > 2\sigma(I)$]	$R_1 = 0.0627$, $wR_2 = 0.1609$	$R_1 = 0.0770$, $wR_2 = 0.2403$	$R_1 = 0.0900$, $wR_2 = 0.2429$
R indices (all data)	$R_1 = 0.0581$, $wR_2 = 0.1648$	$R_1 = 0.0836$, $wR_2 = 0.2478$	$R_1 = 0.1079$, $wR_2 = 0.2593$
Refinement methods	Full-matrix least-squares	Full-matrix least-squares	Full-matrix least-squares
Largest diff. Peak/hole (e Å^{-3})	0.34/−0.22	0.99/−0.36	1.35/−0.52

TABLE 3 Selected Bond Lengths (Å) and Angles (°) for $C_{15}H_{17}N_2O_4P$ (**2a**)

Bond Lengths	*(Å)*	*Angles*	*(°)*
P(1)–O(2)	1.459(2)	O(2)–P(1)–O(4)	117.18(14)
P(1)–O(3)	1.568(2)	O(3)–P(1)–O(2)	113.83(13)
P(1)–O(4)	1.553(2)	O(4)–P(1)–O(3)	102.78(13)
P(1)–C(13)	1.814(3)	O(3)–P(1)–C(13)	105.52(14)
N(1)–C(8)	1.329(5)	O(4)–P(1)–C(13)	102.32(13)
N(1)–C(9)	1.320(5)	O(2)–P(1)–C(13)	113.83(14)
N(2)–C(12)	1.342(4)	C(9)–N(1)–C(8)	115.6(3)
N(2)–C(13)	1.458(3)	C(12)–N(2)–C(13)	121.3(2)
O(1)–C(12)	1.215(3)	C(14)–O(3)–P(1)	123.1(2)
O(3)–C(14)	1.434(4)	C(15)–O(4)–P(1)	123.3(2)
O(4)–C(15)	1.434(4)		

TABLE 4 Selected Bond Lengths (Å) and Angles (°) for $C_{17}H_{21}N_2O_4P$ (**2b**)

Bond Lengths	*(Å)*	*Angles*	*(°)*
P(1)–O(2)	1.449 (2)	O(3)–P(1)–O(4)	107.27 (15)
P(1)–O(3)	1.560 (3)	O(3)–P(1)–C(13)	102.96 (14)
P(1)–O(4)	1.568 (2)	O(4)–P(1)–C(13)	108.49 (14)
P(1)–C(13)	1.822 (3)	O(2)–P(1)–O(3)	116.08 (15)
N(1)–C(8)	1.305 (6)	O(2)–P(1)–O(4)	108.49 (14)
N(1)–C(9)	1.316 (6)	O(2)–P(1)–C(13)	113.15 (13)
N(2)–C(12)	1.334 (4)	C(16)–O(4)–P(1)	125.0 (3)
N(2)–C(13)	1.443 (3)	C(14)–O(3)–P(1)	126.3 (2)
O(1)–C(12)	1.206 (4)	C(8)–N(1)–C(9)	116.0 (4)
O(3)–C(14)	1.433 (5)	C(12)–N(2)–C(13)	121.4 (2)
O(4)-C(16)	1.415 (5)		

TABLE 5 Selected Bond Lengths (Å) and Angles (°) for $C_{19}H_{25}N_2O_4P$ (**2c**)

Bond Lengths	*(Å)*	*Angles*	*(°)*
P(1)–O(2)	1.453(3)	O(3)–P(1)–O(4)	103.05(16)
P(1)–O(3)	1.570(3)	O(2)–P(1)–O(3)	114.34(16)
P(1)–O(4)	1.553(3)	O(3)–P(1)–C(13)	105.22(19)
P(1)–C(13)	1.814(4)	O(2)–P(1)–O(4)	117.11(17)
N(1)–C(8)	1.318(6)	O(4)–P(1)–C(13)	100.57(17)
N(1)–C(9)	1.312(6)	O(2)–P(1)–C(13)	114.78(18)
N(2)–C(12)	1.334(5)	C(8)–N(1)–C(9)	115.2(4)
N(2)–C(13)	1.458(5)	C(12)–N(2)–C(13)	120.1(3)
O(1)–C(12)	1.215(5)	C(14)–O(3)–P(1)	122.9(2)
O(3)–C(14)	1.464(5)	C(17)–O(4)–P(1)	123.7(3)
O(4)-C(17)	1.477(6)		

X-Ray Crystallography

Data were collected at 298 K on a Bruker SMART CCD X-ray diffractometer fitted with Mo Kα radiation ($\lambda = 0.7013$ Å). The structures were solved by direct methods yielding the positions of all non-hydrogen atoms, and refined with a full-matrix least squares procedure based on F^2 using the SHELXL-97 program system.

The crystal data and the final refinement details of **2a, 2b**, and **2c** are given in Table 2, selected bond distances and angles are shown in Tables 3–5, respectively [19,20]. The crystal structures and

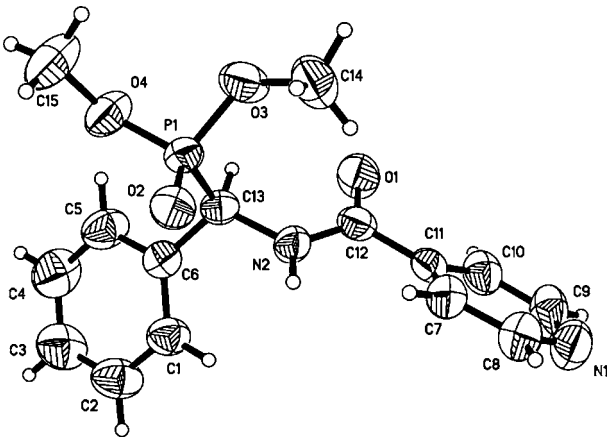


FIGURE 1 ORTEP drawing of the molecular structure **2a.** Displacement ellipsoids are at 50% probability level.

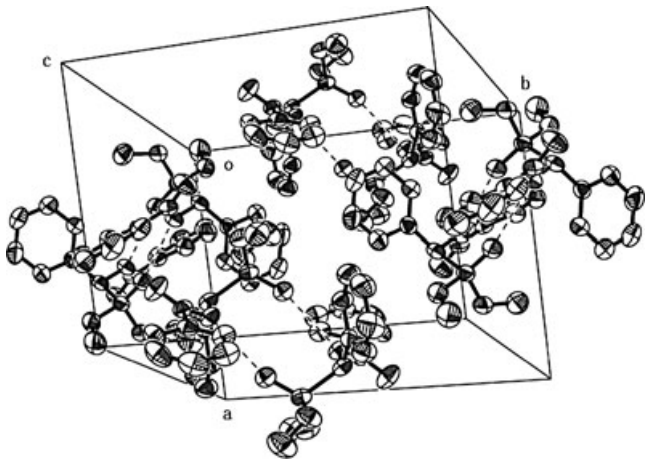


FIGURE 2 ORTEP drawing of the unit cell packing **2a.** Displacement ellipsoids are at 50% probability level. Hydrogen atoms are omitted for clarity.

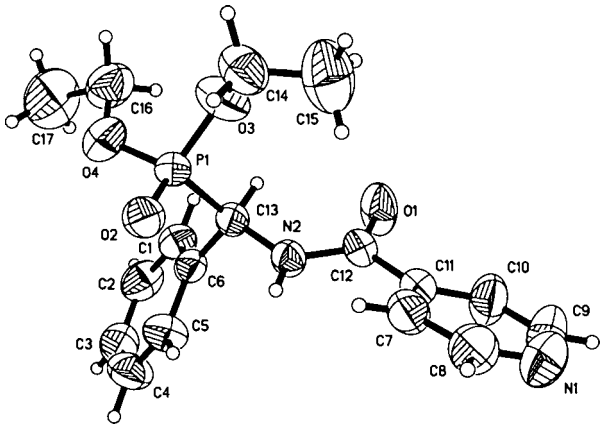


FIGURE 3 ORTEP drawing of the molecular structure **2b.** Displacement ellipsoids are at 50% probability level.

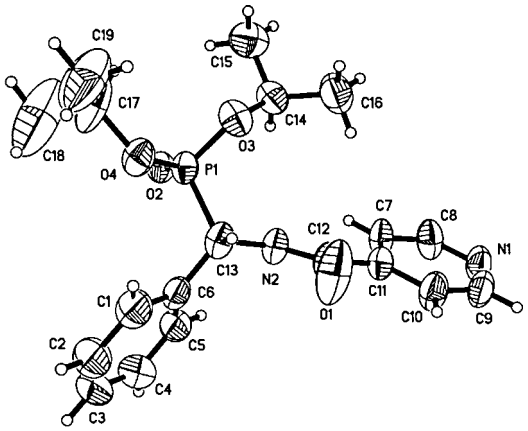


FIGURE 5 ORTEP drawing of the molecular structure **2c.** Displacement ellipsoids are at 50% probability level.

unit cells packing figures are shown in Figs. 1–6 [21,22].

The adjacent bonds O(4)—P(1)—O(3) (mean, 1.561(2) Å for **2a**; 1.564(3) Å for **2b**; 1.562(4) Å for **2c**) are similar to ammonium dimethylphosphate (1.559(7) Å) and methylethylenephosphate (1.57(1) Å) [23,26]. The phenyl and pyridine groups are planar well within the experimental error. The packing of the molecules assumed to be dictated by van der Waal interactions and by intermolecular hydrogen bonds. Two hydrogen bond donors formed hydrogen bonds in the crystal, as depicted in Figs. 2, 4, and 6 (unit-cell packing diagram viewed). In **2a, 2b**, and **2c**, the length of the intermolecular hydrogen bonds (N-2···O-2) are 2.949(3) Å, 2.891(3) Å, and 2.919(5) Å, respectively; and N-2—H···O-2 angles are 166.9(3)°, 169.1(3)°, and 171.2(5)°, respectively. C(12), O(1), and N(2) are coplanar with the pyridine.

Biological Activities

Antibacterial and antitumor activities were also determined [27]. Five microbes (*Aspergillus sp., Cadida albicans, Escherchia coli, Staphylococcus allreus, and Bacillus stubtilis*) were used as indicator organisms to determine the antimicrobial activity of **1a, 1b, 1c, 2a**, and **2b**. These compounds displayed weak antibacterial activity against *Staphylococcus allreus* (inhibition zone = 7 mm); however, **1c** displayed high antibacterial activity against *Bzcillus subtilis* (inhibition zone = 12 mm). The cytotoxicty

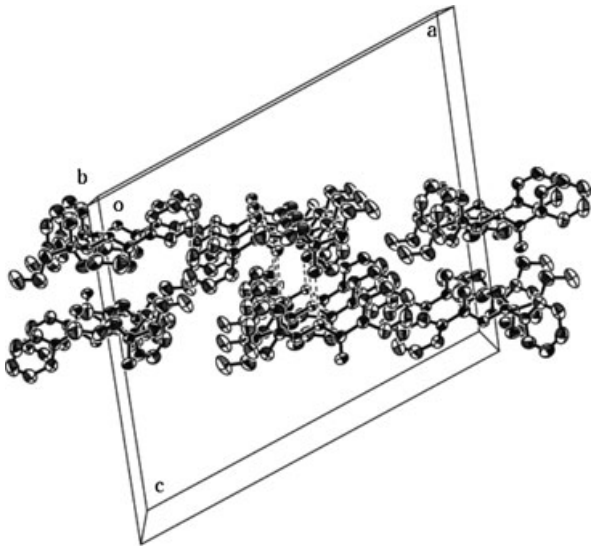


FIGURE 4 OR TEP drawing of the unit cell packing **2b.** Displacement ellipsoids are at 50% probability level. Hydrogen atoms are omitted for clarity.

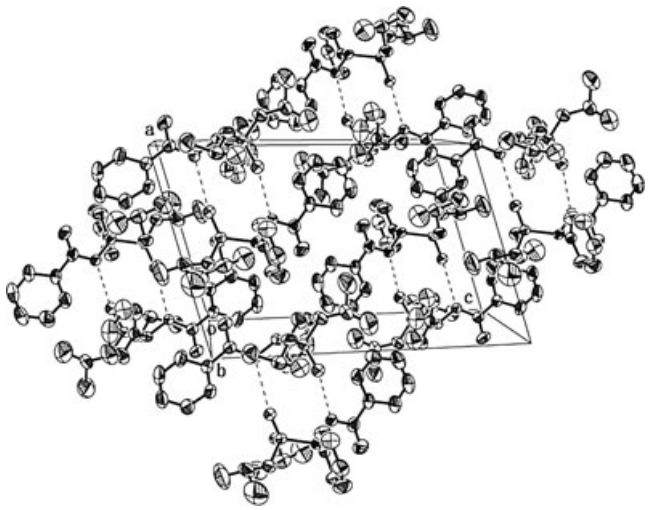


FIGURE 6 ORTEP drawing of the unit cell packing **2c.** Displacement ellipsoids are at 50% probability level. Hydrogen atoms are omitted for clarity.

of the compounds was tested in a concentration of 10 μg/mL by MTT assay according to the procedure described in the literature [28]. The cell line used was a human cancer cell line, KB cells. **2a, 2b**, and **2c** all displayed antimicrobial activity against KB cells. The IC_{50} values for **2a, 2b**, and **2c** were 114.1μg/mL, 68.5 μg/mL, and 51.8 μg/mL respectively. Bioactivities increased with the bulk of the alkyl group, Me > Et > ^{i}Pr. Further study on the antibacterial and antitumor activities of these compounds is underway.

Supplementary Data

Crystallographic data of the structural analyses (excluding structure factors) have been deposited with the Cambridge Crystallographic Data Center, CCDC nos. 248275, 248276, 248277. Copies of this information may be obtained free of charge from the Director, CCDC, 12 Union Road, Cambridge CB2 1EZ UK on request (fax: +44 1223-336-033; email: deposit@ccdc.cam.ac.uk or http://www.ccdc.cam.ac.uk) quoting the deposition numbers for **2a, 2b**, and **2c**, respectively.

REFERENCES

[1] Baylis, E. K.; Campbell, C. D.; Dingwall, J. G. J Chem Soc, Perkin Trans I 1984, 12, 2845.
[2] Maier, L. Phosphorus, Sulfur Silicon 1991, 61, 65.
[3] Kowalik, J.; Sawka, D.; Gowiak, T. J Chem Soc, Chem Commun 1984, 7, 446.
[4] Chen, R.-Y.; Liu, L. Z.; Zhang, Zh. -B. Heteroatom Chem 1995, 6, 503.
[5] Shimonishi, Y.; Sakakibara, S. Bull Chem Soc Jpn 1962, 35, 1966.
[6] Veber, D. F.; Hirschmann, R.; Denkewalter, R. G. J Org Chem 1969, 34, 753.
[7] DeTar, D. F.; Silverstein, R. J Am Chem Soc 1966, 88(5), 1013.
[8] Wuensch, E.; Drees, F. Chem Ber 1966, 99, 110.
[9] Ogura, H.; Nagai, S.; Takeda, K. Tetrahedron Lett 1980, 21, 1467.
[10] Kiso, Y.; Miyazaki, T.; Satomi, M.; Hiraiwa, H.; Akita, T. J Chem Soc, Chem Commun 1980, 22, 1029.
[11] Ueda, M.; Oikawa, H. J Org Chem 1985, 50, 760.
[12] Dong, S.-Z.; Fu, H.; Zhao, Y.-F. Synth Commun 2001, 31, 2067.
[13] Miao, Zh.-W.; Fu, H.; Tu, G.-Zh.; Zhu, J.-G. Ai, H. W.; Zhao, Y.-F. Heteroatom Chem 2003, 14, 62.
[14] Takahashi, H.; Yoshioka, M.; Imai, N.; Onimura, K. Synthesis 1994, 9, 763.
[15] Yuan, C.-Y.; Wang, G.-H.; Chen, Sh.-J. Synthesis 1990, 6, 522.
[16] Yuan, C.-Y.; Qi, Y.-M. Acta Chim Sinica 1986, 44(3), 280.
[17] Appel, R.; Baeumer, G.; Struever, W. Chem Ber 1975, 108, 2680.
[18] Appel, R.; Halstenbery, J. Chem Ber 1977, 110, 2374.
[19] Sheldrick, G. M. (1997). SHELXS-97 and SHELXL97. University of Gottingen, Germany.
[20] Accelrys. ViewerPro (V4.2). Accelrys Inc., Burlington, MA, USA, 2001.
[21] Bruker. SAINT (V6.22), SMART (V5.625), and SADABS (V2.03). Bruker AXS Inc., Madison, WI, USA; 2001.
[22] Farrugia, L. J. J Appl Cryst 1997, 30, 565.
[23] Giarda, L.; Garbassi, F.; Calcaterra, M. Acta Cryst Sect B 1973, 29, 1826.
[24] Bryan, A.; Ronald, M. M.; John, M. H.; Glen, B. R.; Alan, M. S. J Am Chem Soc 1977, 99(8); 2652.
[25] Steitz, T. A.; Lipscomb, W. N. J Am Chem Soc 1965, 87, 2488.
[26] Furberg, S.; Solbakk, J. Acta Chem Scand 1973, 27, 1226.
[27] Scudiero, D. A.; Shoemaker, R. H.; Paull, K. D.; Monks, A.; Tiermey, S.; Nofziger, T. H. J Cancer Res 1998, 48(4), 827.
[28] Mosmann, T. J Immunol Methods 1983, 65, 55.