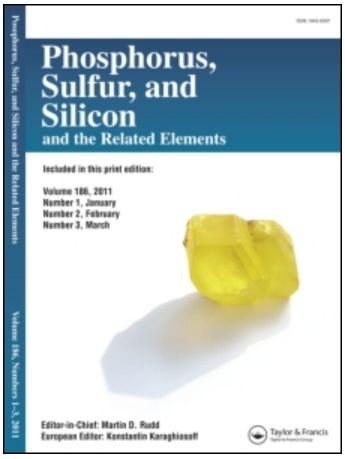
This article was downloaded by:
On: *29 January 2011*
Access details: *Access Details: Free Access*
Publisher *Taylor & Francis*
Informa Ltd Registered in England and Wales Registered Number: 1072954 Registered office: Mortimer House, 37-41 Mortimer Street, London W1T 3JH, UK



Phosphorus, Sulfur, and Silicon and the Related Elements
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713618290

Synthesis and Properties of Novel Nitrogen Mustard Linked Phosphoryl Diamide Derivatives

Ru-Yu Chen[a]; Li-Juan Mao[a]; Hui-Lin Wang[a]; Jia Zhou[a]
[a] Institute of Elemento-Organic Chemistry, Nankai University, Tianjin, People's Republic of China

To cite this Article Chen, Ru-Yu , Mao, Li-Juan , Wang, Hui-Lin and Zhou, Jia(1994) 'Synthesis and Properties of Novel Nitrogen Mustard Linked Phosphoryl Diamide Derivatives', Phosphorus, Sulfur, and Silicon and the Related Elements, 89: 1, 89 — 95
To link to this Article: DOI: 10.1080/10426509408020437
URL: http://dx.doi.org/10.1080/10426509408020437

PLEASE SCROLL DOWN FOR ARTICLE

Full terms and conditions of use: http://www.informaworld.com/terms-and-conditions-of-access.pdf

This article may be used for research, teaching and private study purposes. Any substantial or systematic reproduction, re-distribution, re-selling, loan or sub-licensing, systematic supply or distribution in any form to anyone is expressly forbidden.

The publisher does not give any warranty express or implied or make any representation that the contents will be complete or accurate or up to date. The accuracy of any instructions, formulae and drug doses should be independently verified with primary sources. The publisher shall not be liable for any loss, actions, claims, proceedings, demand or costs or damages whatsoever or howsoever caused arising directly or indirectly in connection with or arising out of the use of this material.

Phosphorus, Sulfur, and Silicon, 1994, Vol. 89, pp. 89–95
Reprints available directly from the publisher
Photocopying permitted by license only

© 1994 Gordon and Breach Science Publishers SA
Printed in the United States of America

SYNTHESIS AND PROPERTIES OF NOVEL NITROGEN MUSTARD LINKED PHOSPHORYL DIAMIDE DERIVATIVES

RU-YU CHEN,* LI-JUAN MAO, HUI-LIN WANG and JIA ZHOU
Institute of Elemento-Organic Chemistry, Nankai University, Tianjin, 300071, People's Republic of China

(Received December 27, 1993; in final form March 28, 1994)

Two series of novel phosphoryl diamide derivatives (**3**, **4**) containing the nitrogen mustard group were synthesized, and the effects of substituents on the reactivity and the spectral properties are discussed. All the compounds prepared were confirmed by ^{1}H NMR, IR, MS, ^{31}P NMR and elemental analysis. Preliminary bioassays indicate that compounds **3** have good antitumor activities, and some of compounds **4** display high inhibitory activities against the tobacco mosaic virus (TMV).

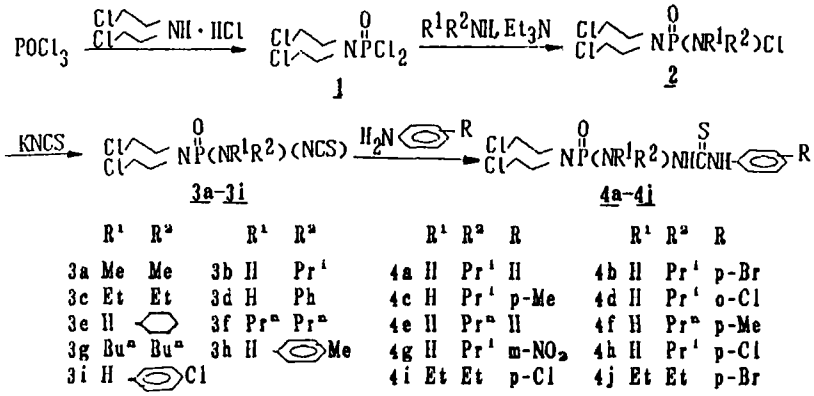
Key words: Antitumor; antivirus; phosphoryl diamide; nitrogen mustard.

Downloaded At: 12:07 29 January 2011

INTRODUCTION

The synthesis and the antitumor mechanism of the phosphoryl mustard derivatives have been reported in the literature.[1–3] Some are widely used in cancer chemotherapy, such as Endoxan, Holoxan etc., but their by-effects on marrow cells and urinary system[4] somewhat limit their clinical use. Therefore, attempts to modify the structures of the mustard derivatives of cyclophosphamide for an enhancement of their chemotherapeutic properties were made.

For the straight chain phosphoryl mustard derivatives, it has been reported that if the substituents attaching to the phosphorus atom are electron-attracting groups, they tend to have good antitumor activity, but instead, if they are electron-donating groups, there is no activity.[5] To look for antitumor drugs with high activity and low toxicity, we designed the structures of compounds **3** and **4**, which were synthesized by the following route (Scheme I):



SCHEME I

*Author to whom correspondence should be addressed.

All of the compounds **3** and **4** were confirmed by microanalyses and spectroscopic methods, and their biological activities were also tested.

RESULTS AND DISCUSSION

A. Spectral Properties and Structures of **3** and **4**

In the ^{1}H NMR spectra of compounds **3** and **4**, the methylene protons of the nitrogen mustard appear as a group of characteristic complex absorption peaks in the range of δ 3.2–3.9 ppm. Since the phosphorus atom is chiral, the two alkyls in the dialkyl amino group are magnetically non-equivalent. Taking **3a** as an example, the protons of one methyl in the dimethyl amino are coupled to be a doublet by phosphorus ($^3J_{NCH}$ = ~1.4 Hz), and appear at the lower field, but with regard to the protons of the other one the absorption signal is just a singlet. Similarly, the two methyls in the compounds containing —NHPr^i or —NEt_2 group are split into multiplets. In the ^{1}H NMR of **4**, the two N—H protons of the thioureido group appear at the lower field. The one, N—H_a, near the P atom displays phosphorus coupling, and appears as a doublet ($^2J_{PNH}$ = ~9 Hz) in the range of δ 7–9 ppm, while the other one, N—H_b, appears as a singlet at the chemical shift of about 11.0 ppm. This abnormal phenomenon might be explained by intermolecular and intramolecular hydrogen-bonding. Taking **4a** as an illustration, its molecular structure is shown in Figure 1. By X-ray diffraction analysis, it was found that the atoms P, O(1), N(3), C(8), N(4) and H(3) bonded to N(4) are coplanar. The bond distances of H(3) ··· O(1) and H(2) ··· O(1)^1 are 1.80 Å and 1.90 Å respectively, and the occurrence of intramolecular hydrogen-bonding (represented by the broken line in Figure 1 and H(3) is omitted) results in the formation of the six-membered ring. It must be pointed out that, when R is a strong electron-attracting group such as m-NO_2 or a bulky group such as p-Me, there might be less tendency to form the intramolecular hydrogen-bonding, therefore, no absorption peak was observed at about 11.0 ppm in the ^{1}H NMR spectra of **4c** and **4g**.

For the IR spectra of compounds **3**, the normal stretching absorption bands indicate the existence of P=O (~1200 cm^{-1}) and —N=C=S (~2000 cm^{-1}). When

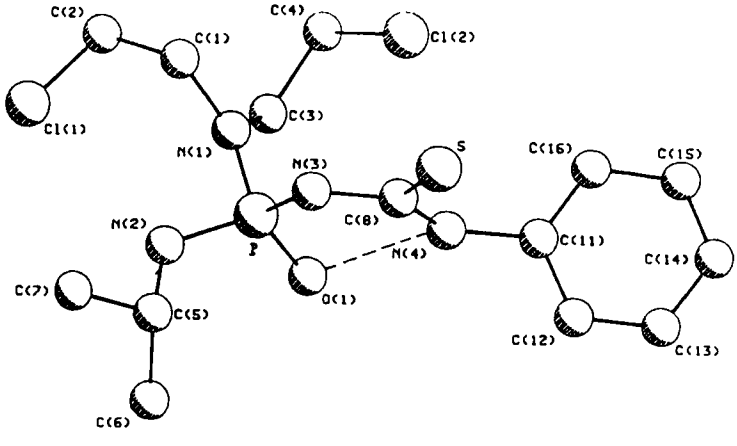


FIGURE 1 The structure of compound **4a**.

Downloaded At: 12:07 29 January 2011

compounds 3 are converted to 4, the signal at ~2000 cm^{-1} disappears and compounds 4 display the absorption peak of $\diagdown\!\!\diagup C{=}S$ (1305–1338 cm^{-1}) in the $\diagdown N{-}C(S){-}N\diagup$ group and the stretching absorption peak of the N—H (3100–3406 cm^{-1}).

TABLE I

The physical and chemical data of compounds 3a–3i

No.	^1H NMR ($CDCl_3$, δ, ppm)	IR (cm^{-1})	Elemental Analysis/Found(Cacld.) or MS Data(m/e)*		
			C(%)	H(%)	N(%)
3a	2.8(s,3H,NCH_3),2.7-2.8 (d,3H,NCH_3),3.2-3.7(m, 8H,2ClCH$_2$CH$_2$N)	2950,2038(s,C=N=S) 1425,1338,1230(m, P=O),1140,972	28.81 (28.97)	4.88 (4.86)	14.58 (14.47)
3b	1.15(s,3H,CH_3),1.25(s, 3H,CH_3),2.90-3.10(w, 1H,NH),3.2-3.7(m,8H, 2ClCH$_2$CH$_2$)	3181(m,NH),2961, 2040(s,C=N=S),1428 1222(P=O),1123, 902	305(M^++2),303(M^+),261, 245,(M^+-NCS),213,140, 58,57		
3c	1.0-1.3(t,6H,2CH_3, $^3J_{HCCH}$=3.6Hz),2.9-3.3 (m,4H,2NCH_2,$^3J_{HCCH}$=3.6 Hz),3.3-3.8(m,8H, 2ClCH$_2$CH$_2$N)	2958,1980(s,C=N=S) 1589,1434,1340, 1237(s,P=O), 1157,1084,972	33.90 (33.96)	5.53 (5.70)	12.97 (13.20)
3d	3.2-3.8(m,8H,2ClCH$_2$CH$_2$ N),4.0-4.2(w,1H,NH), 7.2-7.5(m,5H,Ph)	3274(m,NH),3080, 2971,2038(s,C=N=S) 1572,1480,1254(s, P=O),1145,963	38.71 (39.06)	3.65 (4.17)	11.97 (12.42)
3e	0.9-2.1(m,11H,C_6H_{11}), 2.8-3.2(w,1H,NH), 3.2-3.7(m,8H,2ClCH$_2$CH$_2$N)	2917,2014(s,C=N=S) 1428,1221(s,P=O), 1083,1002,908	38.64 (38.37)	5.88 (5.86)	11.99 (12.49)
3f	0.7-1.0(t,6H,2CH_3, $^3J_{HCCH}$=3.2Hz),1.38-1.8 (m,4H,2CH_2),2.70-3.1(m,4H,2NCH_2)	2949,1994(s,C=N=S) 1235(s,P=O),1102, 1003,971	347(M^++2),345(M^+),318, 245,205,117,140, 100,58		
3g	0.7-1.0(t,6H,2CH_3),1.1 -1.7(m,8H,2CH$_2$CH$_2$), 2.8-3.2(t,4H,2NCH_2)	2943,1986(s,C=N=S) 1440,1338,1227(s, P=O),1148,1032, 973	376(M^++2),373(M^+),332, 330,288,245,191, 149,140,90,58		
3h	2.2-2.3(d,3H,CH_3),3.2-3.8(m,8H,2ClCH$_2$CH$_2$), 6.9-7.4(q,4H),3.8-4.0 (w,1H,NH)	3254,3026,2987, 2020(s,C=N=S),1563 1480,1293(s,P=O), 1166,982	41.24 (40.91)	4.09 (4.58)	11.58 (11.92)
3i	5.8-6.0(w,1H,NH),3.0-3.8(m,8H,2ClCH$_2$CH$_2$N), 6.8-7.4(m,4H,Ph-Cl)	3427(m,NH),3080, 2968,2016(s,C=N=S) 1572,1481,1286(m, P=O),1172,1058,963	343(M^++2),341(M^+),306, 313,292,230,140, 111,58		

* The compounds with MS data are viscous,and their elemental analysis data exceed ±0.5% of the theoretical values.

Downloaded At: 12:07 29 January 2011

TABLE II

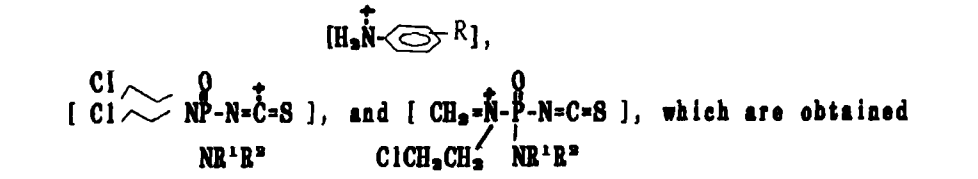
The physical and chemical data of compounds 4a–4j

No.	^{1}H NMR (TMS, $CDCl_3^{**}$, δ, ppm)	m.p. (℃)	Yield (%)	Ele. Anal. /Found (Calcd.)		
				C (%)	H (%)	N (%)
4a	1.23(m,6H,$2CH_3$),3.1(w,1H,NH), 3.38-3.70(m,8H,$2ClCH_2CH_2$,CH), 7.20-7.38(m,5H,5Ph-H),11.0(s, 1H,NH)	133 - 134	82.1	43.03 (43.32)	5.90 (5.79)	14.22 (14.11)
4b	1.23(m,6H,$2CH_3$),3.07(w,1H,NH), 3.39-3.69(m,8H,$2ClCH_2CH_2$,CH), 7.42-7.52(m,5H,4Ph-H,NH),11.0 (s,1H,NH)	135 - 135.5	76.8	35.40 (35.29)	4.45 (4.62)	11.66 (11.76)
4c	1.20(dd,6H,$2CH_3$),3.02(m,1H,NH) 3.32-3.88(m,8H,$2ClCH_2CH_2$,CH), 7.12-7.56(m,6H,4Ph-H,2NH)	137	85.5	43.45 (43.80)	5.97 (6.08)	13.42 (13.63)
4d*	1.16(d,6H,$2CH_3$),3.20-3.96(m, 8H,$2ClCH_2CH_2$,CH),5.0(m,1H,NH), 7.20-7.96(m,4H,4Ph-H),9.52(d, 1H,NH),11.30(s,1H,NH)	142 - 142.5	73.2	39.06 (38.93)	5.25 (5.10)	12.76 (12.98)
4e	0.86(t,3H,CH_3),1.40(m,2H,CH_2), 3.0(m,2H,CH_2),3.22-3.69(m,8H, $2ClCH_2CH_2$,NH),7.12-7.80(m,5H, 5Ph-H),7.76(d,1H,NH),10.82(s, 1H,NH)	102 - 103	83.2	43.18 (43.32)	5.74 (5.79)	14.17 (14.11)
4f	0.93(t,3H,CH_3),1.54(m,2H,CH_2), 2.32(s,3H,CH_3),3.02(m,2H,CH_2), 3.30(m,1H,NH),3.40-3.76(m,8H, $2ClCH_2CH_2$),7.1-7.5(m,5H,4Ph-H, NH),10.82(s,1H,NH)	86 - 97	84.0	43.46 (43.80)	5.85 (6.08)	13.60 (13.63)
4g*	1.24(dd,6H,$2CH_3$),3.0(w,1H,NH), 3.26-3.78(m,8H,$2ClCH_2CH_2$,CH), 7.28-8.12(m,5H,4Ph-H,NH),8.68 (m,1H,NH)	99 - 100	46.1	37.73 (38.01)	4.86 (4.98)	15.64 (15.84)
4h	1.24(dd,6H,$2CH_3$),3.2(w,1H,NH), 3.26-3.72(m,8H,$2ClCH_2CH_2$,CH), 7.24-7.57(m,5H,4Ph-H,NH),11.08 1H,NH)	133	77.8	38.66 (38.93)	4.96 (5.10)	12.77 (12.98)
4i	1.10(t,6H,$2CH_3$),2.96-3.72(m, 12H,$2ClCH_2CH_2$,$2CH_2$),7.20-7.60 (m,5H,4Ph-H,NH),11.18(s,1H,NH)	116 - 117	62.2	40.80 (40.40)	5.33 (5.39)	12.68 (12.57)
4j	1.06(t,6H,$2CH_3$),2.84-3.69(m, 12H,$2ClCH_2CH_2$,$2CH_2$),7.10-7.60 (m,5H,4Ph-H,NH),11.18(s,1H,NH)	115 - 116	61.7	38.87 (38.73)	4.66 (4.80)	11.31 (11.43)

** The solvent of compounds marked by asterisk is $CD_3C(O)CD_3$

Downloaded At: 12:07 29 January 2011

The EI mass spectra of 3 demonstrate the existence of the molecular ion peaks and ($M^+ + 2$) peaks (see Table I). However, those of compounds 4 indicate that the fragmentations are very complicated, and no molecular ion peak can be found. Their mass spectra indicate that the main fragmentation products of compounds 4 are



via four-membered H-rearrangement. The fragmentation products of both 3 and 4 are consistent with their structures.

B. Effects of Substituents on Reactivity

In the synthesis of 3, when the group R^1 and R^2 in HNR^1R^2 are H or alkyl, the reaction can be carried out in CH_2Cl_2 under a mild condition. But when R^1 and R^2 are bulky, the steric factor should be considered, and the solvents should be used according to their polarity in the order of $HCCl_3 <$ THF $<$ MeCN. Moreover, elevated temperature and longer time are needed. For instance, compound **3h** was obtained in MeCN by heating the reaction solution at 80°C for 10 h.

The conversion of compounds 3 to 4 is a nucleophilic addition to the unsaturated carbon. The substituent R in the phenyl group of the amine affects the reaction intensively. When R is an electron-donating group, the reaction goes on easily, but in the case of an electron-attracting one, the addition is extremely slow and the yield is lower. When R = p—NO_2, no product was obtained.

C. Biological Activities

The preliminary anticancer tests in vitro were carried out by the MTT method. The results which are given in Table III indicate that some of compounds 3 have a high inhibitory effect to the growth of leukaemia L_{1210} cells in vitro, but 4 are inactive. Their anticancer activities in vivo are being tested. From the bioassays,

TABLE III

The antitumor activity of compounds 3

No.	Cancer cell	Drug amount (μg/mL)	Time (h)	Inhibition rate (IC_{50})
3a	L_{1210}	0.933	72	50
3b	L_{1210}	0.619	72	50
3c	L_{1210}	1.260	72	50
3d	L_{1210}	0.312	72	50
3f	L_{1210}	0.697	72	50

Downloaded At: 12:07 29 January 2011

TABLE IV
The antivirus activity of compounds 4 against TMV

No.	Concentr. (ppm)	Inhibition rate (%)	No.	Concentr. (ppm)	Inhibition rate (%)
4a	100	46	4c	100	72
4b	100 500	52 70	4d	100 500	60 84
4g	100	66	4h	100	36
4i	100	68	4j	100	16

however, compounds 4 display good antivirus activity against tobacco mosaic virus (TMV). The results are listed in Table IV.

EXPERIMENTAL

Elemental analysis was performed with a CHNCORDERD MT-3 elementary analyzer. Mass spectra were recorded with a VG-7070E spectrometer using the GAB method. ^{1}H NMR spectra were recorded with a JEOL-FX-90Q spectrometer and BRUKER ACP200. TMS was used as an internal standard for ^{1}H NMR, and 85% H_3PO_4 was used as an external standard for ^{31}P NMR. The IR spectra were measured by using a SHIMADZU-435 instrument. The X-ray diffraction analysis was performed on a PDP 11/44 computer with SDP-PLUS programme package. Melting points were determined with a model YANACO MP-500 apparatus and were uncorrected. Column chromatography was performed on silica gel H(10–40μ, Hai Yang Chemical Factory of Qingdao).

The reagents and solvents were available commercially and purified according to conventional methods.

Bis(2-chloroethyl) amino phosphoryl dichloride (1). A mixture of dry bis(2-chloroethyl)amine hydrochloride (30.0 g, 168.9 mmol) and phosphoryl chloride (78 mL, 0.81 mol) was heated under reflux at 120–140°C for 14 h. Excess phosphoryl chloride was evaporated under reduced pressure to yield the crude product and recrystallized from acetone-petroleum ether as colorless crystals (34.0 g, 78.2%), m.p. 53–54°C (Lit.[6]: yield 80%, m.p. 53–56°C).

N,N-bis(2-chloroethyl)-N'-alkyl (or aralkyl) isothiocyanate phosphoryl diamide (3a–3i) *(General procedure).* To a solution of 2.0 g (7.7 mmol) of bis (2-chloroethyl) amino phosphoryl dichloride (1) in 20 mL of CH_2Cl_2 (or anhydrous $HCCl_3$, THF, MeCN) was added dropwise a mixture of equimolecular amounts of amine and NEt_3 in the corresponding solvent at −12–−20°C. The reaction was kept at this temperature for 1.5 h, then at 25–80°C for 3–8 h until the phosphoryl dichloride (1) disappeared, being traced by TLC. The mixture was filtered, and the solvent removed under reduced pressure. To the residue in 30 mL of anhydrous MeCN was added 1.5 g (15.4 mmol) of KNCS at ambient temperature and stirred for 2–8 h until the phosphoryl chloride (2) disappeared, being traced by TLC, then the mixture was filtered and the solvent was removed on a rotary evaporator by heating on a hot water bath (<60°C) under reduced pressure. The residue was purified by flash column chromatography on silica gel with a mixture of chloroform, ether and petroleum ether (60–90°C) (1:1:3) as the eluent, to give the product (3) in yield of 20–78%. The physical and chemical data are given in Table I.

N,N-bis(2-chloroethyl)-N'-alkyl-N''-aryl thioureido phosphoryl diamide (4a–4j) *(General procedure).* To a solution of 4.0 mmol of N,N-bis (2-chloroethyl)-N'-alkyl isothiocyanato phosphoryl diamide (3) in 15 mL of anhydrous MeCN was added dropwise the mixture of 4.8 mmol aryl amine and 5 mL of MeCN on ice bath. The mixture was allowed to warm to ambient temperature, stirred for 5–6 h, and the product (4) appeared as a solid from the solution. The mixture was filtered, and the solvent removed under reduced pressure. The residue was crystallized from acetone-ether in the refrigerator for 24 h, to give another part of product 4. All the solid products were combined and recrystallized (46–86%). The physical and chemical data are listed in Table II.

Downloaded At: 12:07 29 January 2011

ACKNOWLEDGEMENT

This project was supported by the National Natural Science Foundation of China.

REFERENCES

1. O. M. Friedman and S. K. Carter, *Semin. Oncol.*, **5,** 193 (1987).
2. M. Colvin, C. A. Padgett and C. Fenselan, *Cancer Res.*, **33,** 915 (1973).
3. W. Scheef, H. O. Klein, *et al.*, *Cancer Treat. Rep.*, **63,** 501 (1979).
4. H. Arnold and F. Bourseaux, *Angew. Chem.*, **70,** 539 (1958).
5. F. R. Borch, *et al.*, *PCT Int. Appl.*, *WO*, **8911,** 484 (1989).
6. Kyle Ward, Jr., *J. Am. Chem. Soc.*, **57,** 914 (1935).

Downloaded At: 12:07 29 January 2011