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BORON ANALOGUES OF PHOSPHONOACETATES SYNTHESIS, CHARACTERIZATION AND ANTITUMOR PROPERTIES OF SODIUM DIETHYLPHOSPHITE-CARBOXYBORANE AND RELATED COMPOUNDS

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Abstract Several methods have been investigated for the synthesis of functionalized phosphite-borane adducts As part of these investigations, the monosodium salt of diethylphosphite-carboxyborane (a boron analogue of sodium diethylphosphonoacetate) and related precursors and derivatives have been prepared A brief description of their cytotoxic and antitumor properties is also presented

INTRODUCTION

Borane adducts of phosphites[1-5] have received little attention despite the facts that i) they are more stable[1] than corresponding amine-boranes or other base-borane adducts, ii) they can be readily activated[2] for use in hydroboration under mild conditions, iii) they may be considered as analogues of alkylphosphates, $(RO)_3P{=}O$ vs $(RO)_3PBH_3$, where an oxygen atom has been replaced by an isoelectronic BH_3 group and finally, iv) they may be considered as analogues of alkylmethylphosphonates, $(RO)_2P(O)CH_3$ vs $(RO)_2P(O)BH_3^-$, where the methyl group has been replaced by an isostructural and isoelectronic BH_3 group Since phosphate and phosphonate groups are present in a variety of biologically important molecules, e g DNA, RNA, phospholipids, aminophosphonates, etc , a better understanding of the chemistry and stability of modified phosphite-borane adducts as analogues of natural molecules is important

Additionally, several synthetic phosphonates, e g phosphonoacetic acid, phosphonoformic acid, etc , have been found[6-11] to possess significant antiviral activity. This, coupled with the recently established pharmacological activity of amine-borane adducts,[12-16] makes the phosphite boranes even more interesting Finally, substitution of boron for carbon in isoelectronic species has a pronounced effect on charge and for carboxylic acids, on the pKa of the acid [17] Similar substitution in, e g phosphonoacetic acid, should affect pKa's, lipid solubility and biological activity

Thus, in order to investigate various pathways for the synthesis of substituted phosphite-boranes and at the same time obtain chemically and biologically intriguing molecules, the boron analogue of phosphonoacetic acid (phosphorous acid-carboxyborane adduct) was chosen as the target In this paper, we describe the synthesis of derivatives of phosphorous acid-carboxyborane and attempts towards the synthesis of phosphorous acid-carboxyborane itself, a boron analogue of phosphonoacetic acid

RESULTS AND DISCUSSION

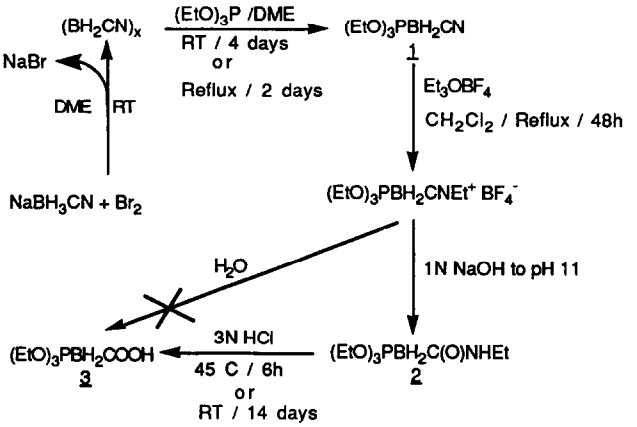
Triethylphosphite-cyanoborane, 1, was prepared in ca 58% yield (Scheme 1) by a method similar to the one described for the synthesis of trimethylphosphite-cyanoborane [4] 1 could also be prepared from aniline-cyanoborane, by the displacement of aniline with $(EtO)_3P$, in refluxing THF The product was obtained in a lower yield (ca 40%) even though the displacement was complete in 3 5 h Similar displacement of Me_3N from Me_3NBH_2CN was slower and only 50-60% reaction was observed (by ^{11}B NMR) after 10 days The slower reaction was expected due to the greater basicity of Me_3N as compared to $PhNH_2$ However, the reaction was performed because the complete displacement of gaseous Me_3N would have simplified the purification

Synthesis of 1, by the intermediate generation of $(BH_2CN)_x$, has also been reported by Das et al [3] However, in this case, I_2 was used as the oxidizing agent In our lab, attempts to synthesize 1 using I_2 completely failed Since both I_2 and Br_2 have been used previously[4] to synthesize $(BH_2CN)_x$, the differences in the final outcome of these reactions must be related to the differences in the conditions used during adduct formation A distinct difference in the conditions used for the adduct formation was the presence or absence of NaX When Br_2 was used as the oxidizing agent, the NaBr formed was insoluble and was filtered prior to the reaction of $(BH_2CN)_x$ with $(EtO)_3P$ In case of I_2, the soluble NaI was left in the reaction mixture during adduct formation The NaI must therefore alter the outcome of this reaction This is indeed the case as was confirmed by the quantitative formation of the sodium salt of diethylphosphite-cyanoborane, 5a, when equimolar amounts of 1 and NaI were heated at reflux in anhydrous DME for 1 h 5a was also observed in the reaction mixture (by ^{31}P NMR) during the attempted synthesis of 1 using I_2 These facts and the ^1H NMR data reported by Das et al [3] clearly suggest that the compound isolated previously was not 1 Since the previously isolated compound was a liquid and was soluble in both benzene and CH_2Cl_2, the possibility of it being 5a can also be ruled out

1 has been used for the synthesis of several new compounds Alkylation with Et_3OBF_4, followed by basic hydrolysis led to the formation of triethylphosphite-N-ethylcarbamoylborane, 2, in ca 73% yield (Scheme 1) During hydrolysis, the pH of the reaction mixture had to be adjusted to ca 11 At lower pH, the reaction was slow, while at higher pH, in addition to the hydrolysis of the nitrilium group, one of the ester groups (P-OR) was also hydrolyzed Even at pH 11, the product was contaminated with small amounts of over hydrolyzed product and the unreacted nitrilium salt However, due to high polarity of these two impurities, purification by chromatography was very simple

Attempts to synthesize 2 by base exchange showed limited success With ammonia-N-ethyl-carbamoylborane as substrate, no exchange was observed Some exchange (ca 25% after 3 days) did occur with trimethylamine-N-ethylcarbamoylborane, but it was accompanied by the formation of some three-coordinate boric acid type species The corresponding aniline derivative, which would have given better results, is an unknown species

Scheme 1

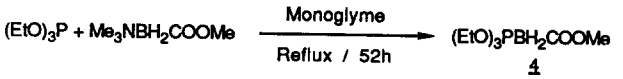


and various attempts at its synthesis have failed [18]

Conversion of 2 to triethylphosphite-carboxyborane, 3, was achieved by hydrolysis with 3N HCl at RT or 45°C At 45°C, the reaction was complete in ca 6 h, while at RT, 2 weeks were required for complete hydrolysis The reaction was also slow at lower concentration of HCl Higher concentrations (6N or 12N) led to hydrolysis of B-H bonds to form increasing amounts of boric acid Boric acid was also the only product, when synthesis of 3 was attempted by an exchange reaction using $(EtO_3)P$ and trimethylamine-carboxyborane or by acid hydrolysis of alkylated cyanoborane Another route, which was successful, involved the hydrolysis of triethylphosphite-carbomethoxyborane, 4 Under similar conditions, the hydrolysis of 4 was much faster than that of 2 The yield of pure product after chromatography, however, was low (ca 46%)

4 itself was prepared in ca 50% yield, by the reaction of $(EtO)_3P$ with trimethylamine-carbomethoxyborane in refluxing DME, Scheme 2 The lower yield was due to loss of product during purification, the exchange itself was > 85% Slower exchange was observed in THF or in large excess of $(EtO)_3P$ as solvent In the case, where excess $(EtO)_3P$ was used as solvent, an additional species with ^{11}B chemical shift similar to 4 was also observed No attempts were made to isolate this species

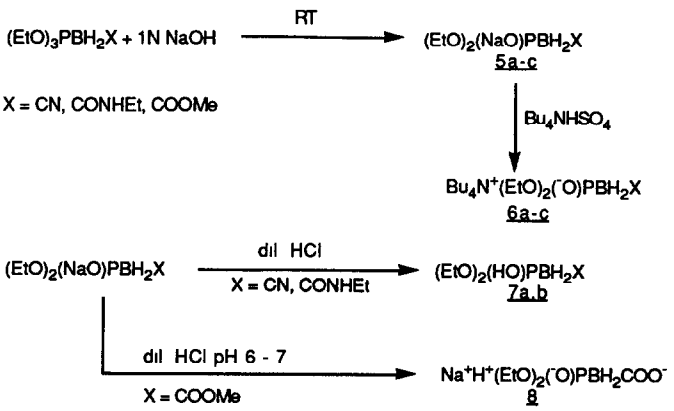
Scheme 2



Reaction of 1, 2 and 4 with 1N NaOH yielded the sodium salts of corresponding diethylphosphite-derivatives, 5a-c, Scheme 3 These were isolated either as such by eva-

poration of water followed by extraction into EtOAc or as tetrabutyl-ammonium salts, 6a-c in > 90% yield Reaction of 3 with 1N NaOH was very slow at room temperature In addition to the formation of a new four coordinate species (based on chemical shift), which could be the desired product, decomposition to sodium borate was observed At higher temperature, ca 45^0C, decomposition was the major reaction No attempts were made to isolate the product

Scheme 3



Acidification of 5a and 5b (Scheme 3) gave the formation of corresponding diethyl-phosphite derivatives 7a-b, while, at a controlled pH (between 6-7), 5c was converted into the monosodium salt of triethylphosphite-carboxyborane, 8 Acidification of 5c to lower pH always led to two major decomposition species in addition to the possible formation of desired product Attempted hydrolysis of 7b to the corresponding carboxyborane with 3N HCl at room temperature or 0 3 N HCl at 42^0C, also resulted in decomposition These results clearly suggest that the diethylphosphite-carboxyborane is unstable under acidic conditions The B-H bonds of 7b itself may also not be very stable under these conditions which may be the reason for its lower yield (ca 32%) from 5b

Several attempts to synthesize and isolate substituted borane adducts of H_3PO_3 have so far been unsuccessful Most of the attempts were directed towards the synthesis of phosphorous acid-cyanoborane, 9, as a model, for two reasons (1) The starting materials could be easily synthesized and thus, were available in sufficient quantities (2) It was expected to be more stable than carboxy- or carboalkoxyborane adducts due to greater electron withdrawing effect of nitrile group strengthening the P-B bond

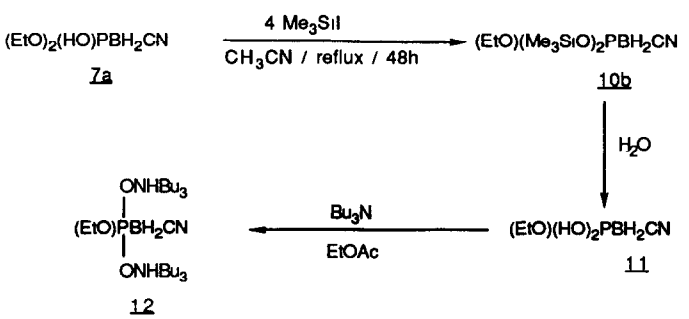
Formation of 9 by direct reaction of H_3PO_3 with $(BH_2CN)_x$ or base BH_2CN is not feasible as H_3PO_3 exists as $(HO)_2P(O)H$ and in addition, it would probably react with $(BH_2CN)_x$ to liberate H_2 Therefore, attempts were directed towards the synthesis of

cyanoborane adducts of highly reactive phosphorous compounds, which could easily be converted into 9 Reaction of PCl_3 with triphenylphosphine-cyanoborane was attempted under various conditions to form Cl_3PBH_2CN Even under very drastic conditions, i e , refluxing in large excess of PCl_3, no exchange was observed

Reaction of excess $(Me_3SiO)_3P$ [ca 90%, remaining $(Me_3SiO)_2P(O)H$] with aniline-cyanoborane did give tris(trimethylsilyl)phosphite-cyanoborane, 10a After removal of solvent, aniline and excess reagent under reduced pressure, ^{31}P NMR showed several impurities in addition to 10a Hydrolysis of 10a to give 9, by addition of water led to decomposition Since the solution became very acidic, acidity may be the cause of decomposition Addition of 1N NaOH to 10a did give a stable sodium salt of 9 (^{11}B nmr$_{(D_2O)}$ δ = -37 2 ppm, $^1J_{B,P}$ = 178 Hz, $^1J_{B,H}$ = 91 ± 2 Hz, ^{31}P nmr$_{(D_2O)}$ δ = 47 8 ppm, $^1J_{B,P}$ = 178 ± 2 Hz), but attempted isolation as the tris(tetra-butylammonium) salt was unsuccessful

Attempted synthesis of 10a by other methods was unsuccessful No reaction was observed when 1 was heated at reflux with excess Me_3SiCl Similarly, with 7a, except for the formation of monosilylated product, no reaction occurred Reaction of 7a with Me_3SiI in acetonitrile at reflux resulted in the formation of ethylbis(trimethylsilyl)phosphite-cyanoborane 10b, which was isolated as ethyl-phosphite-cyanoborane, 11 in ca 50% overall yield (Scheme 4) The product retained some water, which couldn't be removed by keeping it in vacuo for 1 week In addition, the product was usually contaminated with small amounts of phosphorous containing impurities It was reacted with Bu_3N and was isolated as bis(tributylammonium) salt, 12 Although the ^1H, ^{11}B, ^{13}C and ^{31}P NMR corresponded to 12, the elemental analyses of this compound were very unsatisfactory Attempted synthesis of 11 under identical conditions, using 1 as substrate gave a mixture of 11 and 7a Heating 1 in large excess of Me_3SiI at 75-80°C for 48 h also gave the same results

Scheme 4



Though these methods didn't show much success in the formation of 9, these were still tried for the synthesis of corresponding carbomethoxy- or carboxy-borane adducts. When Me_3NBH_2COOMe was heated at reflux in DME with 3 equiv of $(Me_3SiO)_3P$, no reaction was

observed by ^{11}B NMR after 18 h Prolonged heating led to decomposition Since trimethylamine is more difficult to displace than aniline, this result wasn't totally unexpected

Reaction of 4 with Me_3SiI, under the conditions used for the synthesis of 10b, led to complete decomposition within 3 h At RT, using 2 equiv of Me_3SiI, a new product was formed After workup, when a small portion was taken in acetone-d^6, a reaction occured ^{31}P NMR of this sample, showed presence of a quartet at ~75 ppm, in addition to two singlets at 12 3 and -6 11 ppm ^{11}B NMR showed presence of H_3BO_3 and a multiplet at -43 ppm ^1H NMR showed presence of P-OEt but absence of OMe and $SiMe_3$ When taken in D_2O, the new product completely decomposed to give H_3BO_3 These data indicate that the initially formed product was probably $(Me_3SiO)(EtO)_2PBH_2COOSiMe_3$, which, during workup or when taken in acetone, reacted with moisture to give $(HO)(EtO)_2PBH_2COOH$ The latter reacts further with water to give H_3BO_3

In summary, syntheses of several boron analogs of phosphonoacetates are described Various attempts towards the synthesis and isolation of substituted borane adducts of phosphorous acid were unsuccessful These attempts also indicated that if formed, these adducts would probably be unstable in aqueous medium

The newly synthesized compounds have been characterized by a variety of techniques These compounds were tested for cytotoxicity against a variety of murine and human cell lines The results are presented in Table 1 Among murine cell lines, most compounds showed activity against L_{1210} lymphoid leukemia growth but only 5a and 8 were active in the P_{388} lymphocytic leukemia screen In the human tissue culture lines, most of the compounds demonstrated good activity against $Tmolt_3$ leukemia, HeLa-S^3 uterine carcinoma and osteosarcoma In the colon adenocarcinoma screen only 3, 4, 5c and 6c were significantly active, in KB nasopharynx carcinoma screen compounds 1, 5b, 5c and 8 showed good activity Only 6c demonstrated activity against the lung bronchogenic growth while brain glioma growth was retarded by 3, 8 and 12 As demonstrated in Table 1, the activity of the functionalized phosphite-boranes in certain screens, e g , murine L_{1210} and human $Tmolt_3$, is significantly better than that of standards

In addition to tissue culture screens, compounds 2, 4, 7a and 8 were also significantly active against *in vivo* growth of Ehrlich ascites carcinoma

EXPERIMENTAL

^1H NMR spectra were obtained on a JEOL FX90Q, a Brucker NR80 or a Varian XL-300 spectrometer ^{11}B, ^{13}C and ^{31}P NMR spectra were obtained on JEOL FX90Q or Varian XL-300 spectrometer Chemical shifts are presented with respect to Me_4Si for ^1H and ^{13}C NMR spectra, BF_3 Et_2O for ^{11}B NMR spectra and 85% H_3PO_4 for ^{31}P NMR spectra $CDCl_3$ was used as solvent unless stated otherwise In ^1H NMR, all $^3J_{H,H}$ and $^3J_{P,H}$ were close to the 7Hz and BH_2 protons were not observed In ^{13}C NMR, carbon directly attached to boron was not observed due to quadrupole broadening IR spectra were obtained on a Perkin-Elmer 297

Table 1

Cytotoxicity of Boron Analogues of Phosphonoacetates

Compound #	In Vivo % Inhibition Ehrlich Ascites Carcinoma	Murine L_{1210}	Murine P_{388}	ED_{50} (μg/ml) Cytotoxicity Human Tmolt$_3$	Human Colon	Human HeLa^3	Human KB	Human Lung	Human Glioma	Human Osteo Sarcoma
1	66	3 61		2 42	6 56	2 12	2 86	6 88	4 26	2 88
2	80	4 45	6 46							
3	39	3 47	7 26	4 41	3 87	5 29	4 18	4 48	2 65	6 16
4	97	4 15	4 98	3 96	1 75	1 60	7 25	4 80	6 96	7 64
5a	70	3 15	3 62							
5b	60	2 75		2 89	6 40	3 50	3 67	6 80	4 53	1 52
5c	77	2 77	6 25	4 43	3 00	2 37	2 70	4 53	6 34	2 42
6a	78	3 70	7 38							
6b	72	3 31	8 49	2 29	6 10	4 38	7 21	5 48	4 05	4 25
6c	70	2 69	5 96	2 29	3 13	1 69	5 75	3 52	5 49	3 06
7a	94	1 62	4 11	3 36	5 56	5 25	4 01	6 39	4 01	3 91
8	80	3 75	3 76	3 50	5 60	1 75	3 72	5 36	3 91	3 74
12	49	3 31		3 56	4 24	4 83	6 67	4 89	2 87	3 53
13 5FU		1 41	3 72	2 14	3 09	2 47	1 25	5 64	1 28	
14 araC		2 76	4 06	2 67	3 42	2 13	2 54	7 24	1 88	
15 Hydrourea		2 67		3 18	4 74	1 96	5 29	7 33	2 27	7 57
16 Cycloleucine		3 08		2 38	3 81	2 38	5 74	4 36	5 89	6 18
17 6MP	99									

spectrometer Elemental analyses were performed by Galbraith Labs, Tennessee or M-H-W Labs, Arizona Due to the quadrupole moment of B, the peaks in the boron-11 and phosphorous-31 NMR spectra were also broad and the values of $^{1}J_{B,P}$ obtained from a set of spectra (^{31}P and ^{11}B) of the same compound were not always identical

Triethyloxonium tetrafluoroborate,[19] trimethylamine-carboxyborane,[20] trimethyl-amine-carbomethoxyborane,[21] trimethylamine- and ammonia-N-ethylcarbamoylborane,[22] aniline-,[23] trimethylamine-[23] and triphenylphosphine-cyanoborane,[24] and tris(trimethylsilyl)phosphite[25] were prepared by published procedures All other starting materials were obtained commercially Anhydrous 1,2-dimethoxyethane (DME) was obtained commercially, while other solvents were dried by routine methods

Triethylphosphite-cyanoborane, 1

a) From $(BH_2CN)_x$ $NaBH_3CN$ (9 75 g, 155 16 mmol) was dissolved in anhydrous DME (120 ml) under N_2 To this a solution of Br_2 (3 90 ml, 75 70 mmol) in DME (15 ml) was added dropwise with stirring The mixture was stirred at room temperature (RT) overnight and then filtered to remove NaBr The filtrate was mixed with $(EtO)_3P$ (26 10 ml, 152 21 mmol) under N_2 and was either stirred at RT for 4 days or heated at reflux for 2 days It was filtered and the solvent was removed under reduced pressures to give an oil The oil was dissolved in Et_2O (100 ml), washed with water (5 x 75 ml), dried over Na_2SO_4 and the solvent was removed under reduced pressure The residue was kept *in vacuo* for a week to remove traces of triethylphosphate In most batches, the oil became cloudy at this point and was rewashed and dried as described above Attempted distillation under reduced pressure led to decomposition The pure product can be obtained by keeping it *in vacuo* for a long time, *vide supra*, but for the preparation of 2, the crude product was used Yield 17 91 g, 57 70%

b) by exchange Equimolar amounts of $(EtO)_3P$ and an amine-cyanoborane (amine = $PhNH_2$ or Me_3N) were taken in anhydrous THF (25 ml) under N_2 The mixture was heated at reflux and the reaction was followed by ^{11}B NMR For amine = $PhNH_2$, the reaction was complete in 3 5 h After removal of solvent, it was worked up as described in Method a Yield 41 32% ^{1}H NMR, δ(ppm) = 1 39, t, CH_3 and 4 20, m, CH_2 ^{11}B NMR -41 57, $^{1}J_{B,H}$ = 101 ± 1 Hz, $^{1}J_{B,P}$ = 138 ± 1 Hz ^{13}C NMR 15 93, d, $^{3}J_{P,C}$ = 5 5 Hz, CH_3, 63 96, d, $^{2}J_{P,C}$ = 5 5 Hz, CH_2 ^{31}P NMR 91 90, $^{1}J_{B,P}$ = 137 ± 5 Hz IR 2435, 2400 ν(BH), 2205 ν(CN) Analysis calculated for $BC_7H_{17}NO_3P$ C, 41 01, H, 8 36, N, 6 83, P, 15 11 Found C, 40 98, H, 8 36, N, 7 00, P, 15 23

Reaction of 1 with NaI

1 (0 91 g, 4 44 mmol) and NaI (0 665 g, 4 44 mmol) were taken in anhydrous DME under N_2 and heated at reflux for 1 h The solution was cooled, filtered to remove traces of white solid and the solvent was removed under reduced pressure The residue was kept *in vacuo* overnight and analyzed by ^{1}H, ^{11}B and ^{31}P NMR, which were identical to 5a

Triethylphosphite-N-ethylcarbamoylborane, 2

a) via intermediate generation of nitrilium salt: To a stirring solution of 1 (9 83 g, 47.95 mmol) in anhydrous CH_2Cl_2 (48 ml) under N_2, was added a solution of Et_3OBF_4 in CH_2Cl_2 (48 ml of 2 M solution). The mixture was heated at reflux for 48 h, cooled to RT and 1N NaOH was added with stirring until the pH was approximately 11 The layers were separated and the aqueous layer was extracted with CH_2Cl_2 (3 x 75 ml) The combined organic portions were dried and the solvent was removed under reduced pressure to give an oil The product was purified by flash chromatography on silica using ether CH_2Cl_2 (8 2) Yield 8.75 g, 72 68% ^1H NMR, δ(ppm) 1.09, t, CH_3(NEt), 1 34, t, CH_3, 3 26, m, CH_2(NEt), 4.21, m, CH_2, 5 73, br s , NH ^{11}B NMR -31 93, $^1J_{B,H}$ = 95 ± 5 Hz, $^1J_{B,P}$ = 117 Hz. ^{13}C NMR 15 03, s, CH_3(NEt), 15 98, d, $^3J_{P,C}$ = 5 5 Hz, CH_3, 32 63, s, CH_2(NEt), 63 03, d, $^2J_{P,C}$ = 3 3 Hz, CH_2 ^{31}P NMR 97 90, $^1J_{B,P}$ = 118 ± 3 Hz IR 2390 ν(BH), 1600 ν(CO); 3335 ν(NH) Analysis calculated for $BC_9H_{23}NO_4P$ C, 43 06, H, 9 23, N, 5 58, P, 12.34, B, 4 31 Found C, 42 75, H, 9 48, N, 5 17, P, 12 50, B, 4 33

b) by exchange Procedure same as for 1 Reaction was followed by ^{11}B NMR

Triethylphosphite-carboxyborane, 3

a) by acid hydrolysis of 2: 2 (0 50 g, 1 99 mmol) was taken in 3N HCl (30 ml) and was stirred at RT for 2 weeks or at 45-47°C for ca 6 h The solution was extracted with CH_2Cl_2 (3 x 30 ml) The extracts were dried and the solvent was removed to give a clear oil Yield 0 37 g, 82.5% Often, the product was contaminated with a very small amount of 2 It was easily purified by flash chromatography on silica using ether hexane (7 3)

b) by acid hydrolysis of 4 4 (0 92 g, 3 87 mmol) was stirred with 3N HCl (50 ml) at RT for 24 h The mixture was extracted with CH_2Cl_2 (3 x 30 ml) The organic extracts were dried and the solvent was removed *in vacuo* to give an oil (contained ca 10% 4) The product was purified by flash chromatography using hexane EtOAc (1 1) Yield 0 40 g, 46 20% ^1H NMR, δ(ppm) 1 34, t, CH_3, 4 17, m, CH_2, 10 11, br s , OH ^{11}B NMR -33 76, $^1J_{B,H}$ = 96 ± 1 Hz, $^1J_{B,P}$ = 115 Hz ^{13}C NMR 16 01, d, $^3J_{P,C}$ = 6 6 Hz, CH_3, 63 30, d, $^2J_{P,C}$ = 4.4 Hz, CH_2 ^{31}P NMR 96 20, $^1J_{B,P}$ = 119 ± 3 Hz IR 2420 ν(BH), 1655 ν(CO); 3050, ν br ν(OH) Analysis calculated for $BC_7H_{18}O_5P$ C, 37 53, H, 8 10, P, 13 83, B, 4 83 Found C, 37 71, H, 7 88, P, 13 20, B, 4 38.

c) by exchange Procedure same as for 2

d) via intermediate generation of nitrilium salt Procedure same as for 2 except after reaction with Et_3OBF_4, solvent was removed and the residue was hydrolyzed under acidic conditions according to the procedure reported for Ph_3PBH_2COOH [26]

Triethylphosphite-carbomethoxyborane, 4

a) by exchange in a solvent Trimethylamine-carbomethoxyborane (1 00 g, 7 63 mmol) and $(EtO)_3P$ (6 54 ml, 38 14 mmol) were taken in anhydrous DME (50 ml) under N_2 The mix-

ture was heated at reflux and the reaction was followed by ^{11}B NMR After ca 52 h, the solvent was removed under reduced pressure The excess $(EtO)_3P$ was removed in vacuo at RT The residue was taken in ether (50 ml) and washed with water (5 x 30 ml) The ether layer was dried and the solvent was removed under reduced pressure The clear, colorless oil was kept in vacuo for 4-5 days The product is sufficiently pure for further reaction For analysis and biological testing, the product was purified by flash chromatography on silica using ether hexane (1 1) or hexane ethyl acetate (8 2) Yield 0 92 g, 50 63% ^{1}H NMR, δ(ppm) 1 35, t, CH_3, 3 55, S, OCH_3, 4 61, m, CH_2 ^{11}B NMR -33 97, $^{1}J_{B,H}$ = 95 Hz, $^{1}J_{B,P}$ = 122 Hz ^{13}C NMR 16 11, d, $^{3}J_{P,C}$ = 6 6 Hz, CH_3, 48 67, s, OCH_3, 65 35, d, $^{2}J_{P,C}$ = 4 4 Hz, CH_2 ^{31}P NMR 96 59, $^{1}J_{B,P}$ = 122 Hz IR 2415, 2360 ν(BH), 1670 ν(CO) Analysis calculated for $BC_8H_{20}O_5P$ C, 40 33, H, 8 47, P, 13 01, B, 4 54 Found C, 40 47, H, 8 34, P, 13 27, B, 4 35

b) by exchange without a solvent Trimethylamine-carbomethoxyborane (1 50 g, 11 45 mmol) was taken in excess $(EtO)_3P$ (10 ml) under N_2 and was heated at 80°C The reaction was followed by ^{11}B NMR spectroscopy

Diethylphosphite-cyanoborane, sodium salt, 5a

1 (0 56 g, 2 73 mmol) was taken in 1N NaOH (20 ml) and was stirred at RT till all of it dissolved The solution was washed with CH_2Cl_2 (2 x 20 ml) and water was allowed to evaporate at RT The residue was taken in EtOAc (35 ml), filtered and washed with more ethylacetate (35 ml) The filtrate and washings were dried, and the solvent was removed under reduced pressure to give a white hygroscopic solid in quantitative yield ^{1}H NMR (D_2O) δ(ppm) 1 02, t, CH_3, 3 74, m, CH_2 ^{11}B NMR (D_2O) -39 62, $^{1}J_{B,H}$ = 98 ± 2 Hz, $^{1}J_{B,P}$ = 169 Hz ^{13}C NMR (DMSO-d^6) 16 65, d, $^{3}J_{P,C}$ = 4 Hz, CH_3; 57 32, d, $^{2}J_{P,C}$ = 5 4 Hz, CH_2 ^{31}P NMR (DMSO-d^6) 68 28, $^{1}J_{B,P}$ = 176 ± 3 Hz IR 2415, 2370 ν(BH), 2210 ν(CN)

Diethylphosphite-N-ethylcarbamoylborane, sodium salt, 5b

Procedure same as described for 5a Yield 76 45% ^{1}H NMR (acetone-d^6), δ(ppm) 1 11, t, CH_3(NEt), 1 24, t, CH_3, 3 24, q, NCH_2, 3 93, m, OCH_2, NH was not observed ^{11}B NMR (acetone-d^6) -30 49, $^{1}J_{B,H}$ = 88 ± 1 Hz, $^{1}J_{B,P}$ = 149 Hz ^{13}C NMR (acetone-d^6) 15 35, s, CH_3, 16 66, d, $^{3}J_{P,C}$ = 5 4 Hz, CH_3 (OEt), 31 84, s, NCH_2, 57 14, d, $^{2}J_{P,C}$ = 5 1 Hz, OCH_2 ^{31}P NMR (acetone-d^6) 79 97, $^{1}J_{B,P}$ = 144 ± 6 Hz

Diethylphosphite-carbomethoxyborane, sodium salt, 5c

Procedure same as described for 5a Yield 91 58% ^{1}H NMR (D_2O), δ(ppm) 1 13, t, CH_3, 3 40, s, OCH_3, 3 83, m, CH_2 ^{11}B NMR (D_2O) -30 79, $^{1}J_{B,H}$ = 90 ± 3 Hz, $^{1}J_{B,P}$ = 164 Hz ^{13}C NMR (DMSO-d^6) 16 62, d, $^{3}J_{P,C}$ = 5 Hz, CH_3, 47 31, s, OCH_3, 57 02, d, $^{2}J_{P,C}$ = 5 5 Hz, CH_2 ^{31}P NMR (DMSO-d^6) 73 66, $^{1}J_{B,P}$ = 160 ± 6 Hz IR 2430 ν(BH), 1618, 1595 ν(CO)

Diethylphosphite-cyanoborane, tetrabutylammonium salt, 6a.

1 was hydrolyzed to 5a as described, vide supra. After washing with CH_2Cl_2 (2 x 20 ml), the basic solution was stirred with Bu_4NHSO_4 (1 equiv) for 1 h It was extracted with dichloromethane (3 x 35 ml), dried and solvent was removed under reduced pressure to give an oil Yield 99 01% ^1H NMR, δ(ppm) 1 02, t, CH_3, 1 26, t, CH_3 (OEt), 1 46, m, CH_2, 1 67, br m , CH_2, 3 03, distorted t, NCH_2, 3 95, m, OCH_2. ^{11}B NMR -39.66, $^1J_{B,H}$ = 91 ± 1 Hz, $^1J_{B,P}$ = 178 Hz ^{13}C NMR 13 42, s, CH_3, 16 59, d, $^3J_{P,C}$ = 5 5 Hz, CH_3 (OEt), 19 47, s, CH_2; 23 71, s, $CH_2(\underline{C}H_2CH_2CH_3)$, 58 03, s, NCH_2, 58 38, d, $^2J_{P,C}$ = 4 4 Hz, CH_2(OEt) ^{31}P NMR 69.06, $^1J_{B,P}$ = 178 ± 3 Hz IR· 3295 ν(BH), 2195 ν(CN) Analysis calculated for $BC_{21}H_{48}N_2O_3P$ C, 60 28, H, 11 56, N, 6 70, P, 7 40, B, 2 58 Found C, 60 18, H, 11 39, N, 6 87, P, 7 30, B, 2 43

Diethylphosphite-N-ethylcarbamoylborane, tetra-n-butylammonium salt, 6b

Procedure same as described for 6a Yield. 91 42% ^1H NMR, δ(ppm) 0 80, t, CH_3, 0 86, t, CH_3(NEt), 1 02, t, CH_3(OEt), 1 24, m, CH_2, 1 45, br m , CH_2, 2 99, q, CH_2(NEt), 3 10, distorted t, NCH_2, 3 72, m, CH_2(OEt), 6 80, br t , NH ^{11}B NMR -30 49, $^1J_{B,H}$ = 90 Hz, $^1J_{B,P}$ = 146 Hz ^{13}C NMR 13 33, s, CH_3; 14 98, s, CH_3(NEt), 16 57, d, $^3J_{P,C}$ = 5 5 Hz, CH_3(OEt), 19 37, s, CH_2; 23 71, s, $CH_2(\underline{C}H_2CH_2CH_3)$, 32 05, s, CH_2(NEt), 57.60, d, $^2J_{P,C}$ = 4 4 Hz, CH_2(OEt), 58 33, s, NCH_2. ^{31}P NMR 77 85, $^1J_{B,P}$ = 146 ± 3 Hz IR 2415, 2360, 2320 ν(BH), 1598 ν(CO), 3340 ν(NH) Analysis calculated for $BC_{23}H_{54}N_2O_4P$ C, 59 48, H, 11 72, N, 6 03, P, 6 67, B, 2 33 Found C, 59 44, H, 11 61, N, 5 77, P, 6 59, B, 1 70

Diethylphosphite-carbomethoxyborane, tetrabutylammonium salt; 6c

Procedure as described for 6a Yield 97 83% ^1H NMR, δ(ppm) 1 00, t, CH_3, 1 23, t, CH_3(OEt), 1 45, m, CH_2, 1 66, br m , CH_2; 3 30, distorted t, NCH_2, 3 45, s, OCH_3, 3 94, m, CH_2(OEt) ^{11}B NMR -31 08, $^1J_{B,H}$ = 91 ± 2 Hz, $^1J_{B,P}$ = 161 Hz ^{13}C NMR 13 30, s, CH_3, 16 34, d, $^3J_{P,C}$ = 5 4 Hz, CH_3(OEt) 19 32, s, CH_2, 23 61, s, $CH_2(\underline{C}H_2CH_2CH_3)$, 47 50, s, OCH_3, 57 60, d, $^2J_{P,C}$ = 4 0 Hz, CH_2(OEt), 58 29, s, NCH_2 ^{31}P NMR 73 63, $^1J_{B,P}$ = 161 ± 2 Hz. IR 2370 ν(BH), 1665 ν(CO) Analysis calculated for $BC_{22}H_{51}NO_5P$ C, 58 53, H, 11 39, N, 3.10, P, 6 86, B, 2.39. Found C, 58 33, H, 11 18, N, 3 12, P, 6 54, B, 2 22

Diethylphosphite-cyanoborane, 7a

1 (5 75 g, 28 05 mmol) was stirred with 1N NaOH (180 ml) till it went into solution The solution was washed with CH_2Cl_2 (2 x 50 ml), acidified with conc HCl to pH 1-2 and then extracted with CH_2Cl_2 The extracts were dried and the solvent was removed to give a clear, colorless oil Yield 4 65 g, 93 69% ^1H NMR, δ(ppm). 1.35, t, CH_3; 4 11, m, CH_2, 9 65, br s , OH ^{11}B NMR -40.51, $^1J_{B,H}$ = 103 ± 2 Hz, $^1J_{B,P}$ = 141 Hz ^{13}C NMR. 16 08, d, $^3J_{P,C}$ = 5 5 Hz, CH_3, 62.64, d, $^2J_{P,C}$ = 5.5 Hz, CH_2 ^{31}P NMR 86 06,

$^1J_{B,P}$ = 139 ± 13 Hz IR 2425, 2370 ν(BH), 2220 ν(CN) Analysis calculated for $BC_5H_{13}NO_3P$ C, 33 94, H, 7 41, N, 7 92, P, 17 50 Found C, 33 74, H, 7 20, N, 7 65, P, 17 36

Diethylphosphite-N-ethylcarbamoylborane, 7b

Procedure same as described for 7a except the solution was acidified to pH 3-4 Yield 31 86% ^1H NMR, δ(ppm) 1 15, t, CH_3(NEt), 1 21, t, CH_3, 3 33, m, CH_2(NEt), 3 89, m, CH_2, 8 06, br s , NH, 14 50, br s , OH ^{11}B NMR -33 00, $^1J_{B,H}$ = 91 ± 2 Hz, $^1J_{B,P}$ = 163 Hz ^{13}C NMR 13 77, s, CH_3, 16 30, d, $^3J_{P,C}$ = 6 5 Hz, CH_3(OEt), 35 65, s, CH_2, 59 32, d, $^2J_{P,C}$ = 5 5 Hz, CH_2(OEt) ^{31}P NMR 75 19, $^1J_{B,P}$ = 161 Hz IR 2410 ν(BH), 1615 ν(CO), 3280 ν(NH) Analysis calculated for $BC_7H_{19}NO_4P$ C, 37 70, H, 8 59, N, 6 28, P, 13 89, B, 4 85 Found C, 37 71, H, 8 73, N, 6 10, P, 13 71, B, 4 64

Diethylphosphite-carboxyborane, monosodium salt, 8

5c was prepared as described, vide supra and then acidified to pH between 6-7 The water was allowed to evaporate at RT The residue was stirred with EtOAc (3 x 25 ml) and filtered The filtrate was dried and the solvent was removed under reduced pressure to give a white hygroscopic solid Yield 99 59% ^1H NMR (acetone-d^6), δ(ppm) 1 35, t, CH_3, 4 02, m, CH_2, no peak was observed for OH, integration of the spectrum, however, indicated presence of 0 5 H in the region of 10-12 ppm ^{11}B NMR (acetone-d^6) -32 10, $^1J_{B,H}$ = 95 Hz, $^1J_{B,P}$ = 151 Hz ^{13}C NMR (acetone-d^6) 16 84, d, $^3J_{P,C}$ = 6 7 Hz, CH_3, 59 12, d, $^2J_{P,C}$ = 5 4 Hz, CH_2 ^{31}P NMR (acetone-d^6) 78 25, $^1J_{B,P}$ = 151 ± 3 Hz IR 2405 ν(BH), 1645 ν(CO), 3700-2750, ν br band ν(OH)

Attempted preparation of diethylphosphite-carboxyborane

a) from 3 3 (0 15 g, 0 67 mmol) was taken in 1N NaOH (15 ml) and was heated at 45-47°C for 7 h The mixture was left at RT overnight and then washed with CH_2Cl_2 The aqueous layer was acidified, extracted with CH_2Cl_2, dried and the solvent was removed to give an oil. Yield 0 063 g

b) from 7b 7b (0 10 g, 0 49 mmol) was taken in 0 3N HCl (10 ml) and the mixture was heated at 42°C for 6 5 h It was extracted with CH_2Cl_2 (4 x 15 ml) Nothing extracted into CH_2Cl_2 Water was removed from the aqueous layer at RT and the residue was identified by ^{11}B NMR ^{11}B nmr δ = 20 30 ppm, s, H_3BO_3 (major), -31 55 ppm, dt, and 31 57 ppm, br s (minor unknown species)

c) From 4 Procedure same as described for the synthesis of 8 except solution was acidified to lower pH

Attempted preparation of PCl_3 BH_2CN

Ph_3PBH_2CN (1 00 g, 3 32 mmol) and PCl_3 (1 equiv in THF, 2 equiv in $CHCl_3$ or large excess as solvent) were taken in an anhydrous solvent under N_2 The mixture was heated

at reflux and the reaction was followed by ^{11}B NMR

Attempted preparation of phosphorous acid-cyanoborane, 9 via intermediate formation of tris(trimethylsilyl)phosphitecyanoborane, 10a

Aniline-cyanoborane (0 843 g, 6 39 mmol) and $(Me_2SiO)_3P$ (3 82 g, 11 79 mmol of $(Me_3SiO)_3P$ + 1 31 mmol of $(Me_3SiO)_2P(O)H$) were taken in anhydrous THF under N_2 The mixture was heated at reflux for 5 h and then stirred at RT overnight ^{11}B NMR$_{(CDCl_3)}$ δ = -38 17 ppm, dt, $^1J_{B,H}$ = 96 ± 3 Hz, $^1J_{B,P}$ = 146 Hz The solvent was removed under reduced pressure and the residue was kept in vacuo for 5 days It was taken in ether, filtered and the solvent was removed under reduced pressure to give a brown oil, which turned purple upon standing A small portion when taken in D_2O, decomposed into boric acid In 1N NaOH in D_2O, only a small amount of decomposition occurred in the beginning No further decomposition occurred when it was allowed to stand overnight So the purple oil was dissolved in 1N NaOH The solution was washed with CH_2Cl_2 (2 x 50 ml) and EtOAc (2 x 50 ml) The aqueous layer was taken with ca 3 equiv of Bu_4NHSO_4, stirred for 30 min and then extracted with CH_2Cl_2 to give an oil (1 064 g) ^{11}B and ^{31}P NMR spectra showed no boron or phosphorous

Ethylphosphite-cyanoborane, 11

7a (1 15 g) and Me_3SiI (4 equivalent) were taken in anhydrous CH_3CN and were heated at reflux in the dark for 2 days The solvent and excess Me_3SiI were removed under reduced pressure The residue was taken in CH_2Cl_2/water (1 1, v/v, 50 ml) and was stirred at RT for 1 5 h The aqueous layer was separated and washed with CH_2Cl_2 until the organic layer was colorless The water was evaporated at RT by passing a stream of air over the solution The residue was taken in EtOAc and filtered to remove a solid The filtrate was dried and the solvent was removed under reduced pressure to give an oil Often, the EtOAc solution became dark colored, which couldn't be removed by treatment with charcoal To remove this color, the EtOAc was removed and the whole workup was repeated Yield of light yellow oil 0 49 g, 50 72% ^1H NMR (acetone-d^6), δ(ppm) 1 30, t, CH_3, 4 09, m, CH_2, 6 50, s, OH's, integration >> 2 protons indicating presence of H_2O ^{11}B NMR (acetone-d^6) -39 58, $^1J_{B,H}$ = 97 ± 1 Hz, $^1J_{B,P}$ = 155 ± 1 Hz ^{13}C NMR (acetone-d^6) 16 58, d, $^3J_{P,C}$ = 6 6 Hz, CH_3, 61 83, d, $^2J_{P,C}$ = 6 6 Hz, CH_2 ^{31}P NMR (acetone-d^6) 80 56, $^1J_{B,P}$ = 154 ± 5 Hz IR 2450, 2410 ν(BH), 2225 ν(CN)

Ethylphosphite-cyanoborane, bis(tributylammonium) salt 12

11 (0 52 g, 3 49 mmol) was taken in EtOAc (5 ml) and was cooled to 0°C To this Bu_3N (2 60 equivalent, excess) was added and the mixture was stirred at 0°C for 15 min The solution was filtered and the solvent was removed under reduced pressure The residue was kept in vacuo overnight The oily residue was washed with n-pentane (3 x 15 ml), which was removed by decantation The residue was than taken in CH_2Cl_2/H_2O (1 1, v/v, 30

ml), stirred for 5 minutes and the organic layer was separated The aqueous layer was extracted with CH_2Cl_2 (2 x 15 ml) The combined organic layers were dried and the solvent was removed under reduced pressure to give a clear, colorless oil Yield 0 85 g, 46 85% ^1H NMR, δ(ppm) 0 92, br s , CH_3; 1 21, t, CH_3(OEt), 1 35, br s , CH_2, 1 64, br s , CH_2, 2 90, br s , NCH_2, 3 89, m, OCH_2, 11 52, br , NH ^{11}B NMR -39 32, unresolved ^{13}C NMR 13 46, s, CH_3, 16 51, d, $^3J_{P,C}$ = 6 1 Hz, CH_3(OEt), 19 87, s, CH_2, 24 97, s, $CH_2(\underline{C}H_2CH_2CH_3)$, 51 80, s, NCH_2, 59 10, d, $^2J_{P,C}$ = 5 3 Hz, OCH_2. ^{31}P NMR (D_2O + few drops of acetone-d^6) 68 44, $^1J_{B,P}$ = 179 ± 5 IR 2390 ν(BH), 2185 ν(CN) Analysis calculated for $BC_{27}H_{63}N_3O_3P$ C, 62 41, H, 12 22, N, 8 09, P, 5 96, B, 2 08 Found a) by Galbraith Labs C, 55 80, H, 11 50, N, 8 14, P, 6 04, B, 2 81 b) by M H W labs on same sample C, 53 02, H, 10 41, N, 8 23, P and B not analyzed The analyses are inconsistant with the proposed structure

Attempted preparation of tris(trimethylsilyl)phosphite-carbomethoxyborane

a) Me_3NBH_2COOMe (0 80 g, 6 11 mmol) and $(Me_3SiO)_3P$ (6 06 g, 18 72 mmol of $(Me_3SiO)_3P$ and 2 08 mmol of $(Me_3SiO)_2P(O)H$) were taken in anhydrous DME (40 ml) under N_2 The mixture was heated at reflux and the reaction was followed by ^{11}B NMR

b) 4 (1 13 g, 4 75 mmol) and Me_3SiI (2 70 ml, 18 97 mmol) were taken in anhydrous CH_3CN (45 ml) under N_2 Immediately, the mixture became brown It was heated at reflux for 3 h During this time, the brown color completely disappeared The mixture was cooled and the solvent was removed under reduced pressure to give a yellow solid ^{11}B $NMR_{(CDCl_3)}$ δ = 15 ppm, br s , 0 2 ppm, br s, major and a multiplet at -39 ppm, v small amount

Reaction of 4 with iodotrimethylsilane at RT

4 (0 628 g, 2 64 mmol) and Me_3SiI (0 75 ml, 5 27 mmol) were taken in anhydrous CH_3CN (30 ml) under N_2 and stirred at RT for 6 h The mixture was filtered, concentrated, dissolved in CH_2Cl_2, filtered twice to remove small amounts of white solid (H_3BO_3) and the solvent was removed under reduced pressure A small portion was dissolved in acetone-d^6 for NMR

Cytotoxic Activity

All newly synthesized functionalized phosphite-boranes were tested for cytotoxic activity by preparing a 1 mM solution of the drug in 0 05% tween 80/H_2O by homogenization The drug solutions were sterilized by passing them through an Acrodisc 45 µM The following cell lines were maintained by the literature techniques murine L_{1210} lymphoid leukemia,[27] P_{388} lymphocytic leukemia,[27] human $Tmolt_3$ acute lymphoblastic Tcell leukemia,[28] colorectal adenocarcinoma SW480,[29] lung bronchogenic MB-9812,[30] osteosarcoma TE418,[31] KB epidermoid nasopharynx,[27,32] HeLa-S^3 suspended cervical carcinoma,[33] and glioma EH 118 MG [34]

The protocol used to assess cytotoxicity was that of Geran et al [27] Standards were determined in each cell line Values are expressed for the drug's cytotoxicity as ED_{50} in µg/ml, i e the concentration which inhibits 50% of the cell growth determined by the trypan blue exclusion technique Solid tumor cytotoxicity was determined by the method of Huang et al [35] Ehrlich ascites carcinoma in vivo tumor screens were conducted in CF_1 male mice (~28 g) with test drugs at 8 mg/kg/day I P by the method outlined previously [27] 6-Mercaptopurine was used as an internal standard

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REFERENCES

(1) Reetz, T *J Am Chem Soc* **1960**, *82*, 5039-5042

(2) Pelter, A , Rosser, R , Mills, S *J Chem Soc Chem Comm* **1981**, 1014-1015

(3) Das, M K , Roy, S *Synth React Inorg Met -Org Chem* **1986**, *16*, 67-75

(4) Martin, D R , Chiusano, M A , Denniston, M L , Dye, D J , Martin, E D , Pennington, B T *J Inorg Nucl Chem* **1978**, *40*, 9-13

(5) Muetterties, E L 'The Chemistry of Boron and its Compounds', Wiley, New York, **1967**

(6) Mayer, R F , Varnell, E D , Kaufman, M E *Antimicrob Agents Chemother* **1976**, *9*, 308-311

(7) Oberg, B *Pharmac Ther* **1983**, *19*, 387-415

(8) Mao, J C -H , Otis, E R , M von Esch , A , Merrin, T R ; Fairgrieve, J S , Shipkowitz, N L ; Duff, R G *Antimicrob Agents Chemother* **1985**, *27*, 197-202

(9) Clercq, E D *J Med Chem* **1986**, *29*, 1561-1569

(10) Sarin, P S , Taguchi, Y , Sun, D , Thorton, A , Gallo, R C , Oberg, B *Biochem Pharmacol* **1985**, *34*, 4075-4079

(11) Eriksson, B , Oberg, G ; Wahsen, B *Biochim Biophys Acta* **1982**, *696*, 115-123

(12) Hall, I H , Gilbert, C J , McPhail, A T , Morse, K W ; Hassett, K , Spielvogel, B F *J Pharm Sci* **1985**, *74*, 755-758

(13) Hall, I H , Starnes, C O , McPhail, A T , Wisian-Neilson, P , Das, M K , Harchelroad, Jr , F , Spielvogel, B F *J Pharm Sci* **1980**, *69*, 1025-1029

(14) Hall, I H , Das, M K , Harchelroad, F , Jr , Wisian-Neilson, P , McPhail, A T , Spielvogel, B F *J Pharm Sci* **1981**, *70*, 339-341

(15) Hall, I H , Spielvogel, B F , Sood, A , Ahmed, F U , Jafri, S *J Pharm Sci* **1987**, *76*, 359-365.

(16) Hall, I H , Griffin, T S , Docks, E L , Brotherton, R J , Futch, G *J Pharm Sci* **1986**, *75*, 706-710

(17) Scheller, K H , Martin, R B , Spielvogel, B F , McPhail, A T *Inorg. Chim Acta* **1982**, *57*, 227-228

(18) Spielvogel, B F ; Harchelroad, F , Jr , Wisian-Neilson, P *J Inorg Nucl Chem.* **1979**, *41*, 1223-1227

(19) Meerwein, H *Org Synth.* **1966**, *46*, 113-115.

(20) Spielvogel, B F. in "Boron Chemistry-4", IUPAC, Inorganic Chemistry Divison, Parry, R W , Kodama, G , Pergamon Press, New York, NY, 1980, pp 119-129

(21) a) Spielveogel, B F , Ahmed, F U , McPhail, A T *Synthesis* **1986**, 833-835

b) Spielvogel, B F , Ahmed, F U , Silvey, G L , Wisian-Neilson, P , McPhail, A T *Inorg Chem* **1984**, *23*, 4322-4324

(22) Spielvogel, B.F , Morse, K W., Ahmed, F U , McPhail, A.T *Inorg Chem.* **1984**, *23*, 1776-1777

(23) Wisian-Neilson, P , Das, M K , Spielvogel, B F *Inorg Chem* **1978**, *17*, 2327-2329

(24) a) Das, M K ; Roy, S *Synth. React. Inorg Met -Org Chem* **1985**, *15*, 53-59

b) Wisian-Neilson, P , Wilkins, M A , Weigel, F C., Foret, C J , Martin, D R *J Inorg Nucl Chem* **1981**, *43*, 457-458

(25) Thomas, R H., Fairgrieve, J S , Bower, R R , Shipkowitz, N L , Mao, J C.-H. *J Med Chem* **1977**, *20*, 660-663

(26) Wisian-Neilson, P , Wilkins, M A , Weigel, F C ; Foret, C J , Martin, D R *J Inorg Nucl Chem* **1981**, *43*, 457-458

(27) Geran, R I , Greenberg, N H , Macdonald, M M , Schumacher, A M , Abbott, B J *Cancer Chemotherapy Reports* **1972**, *3*, 7-9

(28) Minowada, J , Ohnuma, T , Moore, G.E *J. Nat Cancer Int* **1972**, *49*, 891-895

(29) Leibovitz, A , Stinson, J C , McCombs, W B III, McCoy, C E , Mazur, K C , Mabry, N D *Cancer Res* **1976**, *36*, 4562-4569

(30) Aaronson, S.A , Todaro, G J , Freeman, A E *Expt. Cell Res* **1970**, *61*, 1-5

(31) Smith, H S , Owens, R.B , Hiller, A J , Nelson-Rees, W A , Johnston, J O *Int J Cancer* **1976**, *17*, 219-234

(32) Eagle, H *Proc Soc Expt Biol* **1955**, *89*, 362-364

(33) Puck, T T , Marcus, P I , Cieciura, S J *J Exp Med* **1956**, *103*, 273-283

(34) Nelson-Rees, W A , Flandermeyer, R R , Hawthorne, P K *Int J Cancer* **1975**, *16*, 74-82

(35) Huang, E S , Lee, K H , Piantodosi, C , Geissman, T A , Pagano, J S *J Pharm Sci* **1972**, *61*, 108-110