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Herbicidal Derivatives of Aminomethylenebisphosphonic Acid. Part III. Structure—Activity Relationship

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Abstract. Derivatives of aminomethylenebisphosphonic acids constitute a class of promising herbicides. More than 40 *N*-substituted aminomethylenephosphonic acids were synthesized and evaluated for their herbicidal activity on common cress (*Lepidium sativum* L.) and cucumber (*Cucumis sativus* L.). Some of the tested compounds were found to exhibit strong herbicidal properties being equal in activity with the popular herbicide glyphosate as well as parent *N*-pyridylaminomethylenephosphonic acids. *N*-Substituted iminodi(methylenephosphonic) acids, which may be considered as close analog of glyphosate, were inactive toward test plants.

Key Words. Shikimate pathway—Glyphosate—Bisphosphonates

Derivatives of aminomethylenebisphosphonic acid, developed in Japan (e.g., compounds **1, 2,** and **3;** Suzuki et al. 1979) and the United States (Cromartie and Fisher 1995) constitute a new class of promising herbicides (Fig. 1). Although some attempts to define their mechanism of action have been undertaken (Cromartie and Fisher 1995, Forlani et al. 1996, Lejczak et al. 1996) it still remains unclear. Physiologic activity of the structurally related series of compounds is routinely examined to define the structural requirements of target receptors. Thus, in trying to understand better the mode of action of this class of compounds we have synthesized more than 40 *N*-substituted derivatives of aminomethylenebisphosphonic acids (compounds **4**) and determined the influence of the structural variation of their *N*-substitutent on their herbicidal activities. *N*-Substituted iminodi(methylenephosphonic) acids (compounds **5**) may be considered as close structural analogs of compounds **4,** and thus we also have determined the influence of chosen representatives of this class on the growth of test plants.

Materials and Methods

Chemical Syntheses

N-Substituted aminomethylenebisphosphonic acids were obtained by reacting the corresponding amine with equimolar quantities of ethyl orthoformate and triethyl phosphite (Maier 1981, Suzuki et al. 1979). In the case of substrates bearing carboxylate moieties two equivalents of triethyl phosphite had to be used. Compounds **4dc** and **4dd** were obtained by catalytic (Pd/C) reduction of control compounds **1** and **2** with hydrogen. Melting points and yields of these compounds are given in Tables 1 and 2. Substituted iminodi(methylenephosphonic) acids were prepared according to the described procedure (Moedritzer and Irani 1966).

Evaluation of Herbicidal Activity of Aminomethylenebisphosphonic Acids

The physiologic activity of the studied compounds was tested using common cress (*Lepidium sativum* L.) and cucumber (*Cucumis sativus* L. cv. Wisconsin) seedlings. Each experiment was replicated four times.

The effect of some of these compounds on the growth of suspension cultured cells of maize (*Zea mays* L. cv. Black Mexican Sweet) was also evaluated.

Effects of Studied Compounds on the Growth of Test Plants. Seeds were germinated at 33°C for 1.5–4 days in darkness. Groups of 10–40 uniform seedlings (depending on the species used) were transferred to Petri dishes (9 cm) lined with two discs of Whatman No. 2 filter paper wetted with 10 mL of distilled water (control) or solutions of the test compounds to give final concentrations of 0.05, 0.15, or 1.5 mM. Plants

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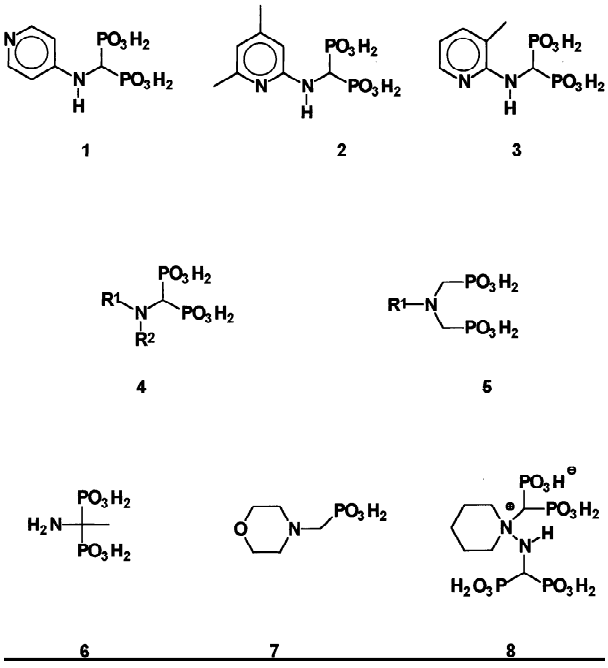


Fig. 1. *N*-(Pyridylamino)aminomethylenebisphosphonic acids (**1, 2,** and **3**) discovered by Suzuki et al. (1979); their analogs: derivatives of aminomethylenebisphosphonic acids (**4**); *N*-substituted iminodi(methylenephosphonic) acids (**5**); 1-amino-1,1-bisphosphonoethylbisphosphonic acid (**6**); and compounds **7** and **8.**

were grown at 25°C with a 12-h day length for 5–9 days (depending on species) under fluorescent tubes (about 300 μE m^{-2} s^{-1} at plant level). Separated roots and shoots were then weighed on a torsion balance. The herbicidal effect was expressed as percentage change in plant root and shoot fresh weight in relation to untreated control.

Inhibition of Plant Cell Culture Growth. Plant cell cultures were grown in Erlenmeyer flasks in MS medium (Murashige and Skoog 1962) containing 1 mg $liter^{-1}$ 2,4-dichlorophenoxyacetic acid. Incubation was in the dark at 26 ± 1°C on a rotary shaker (120 rpm). Subcultures were made every 2 weeks by transferring 25-mL aliquots to 100 mL of fresh medium.

The effect of increasing concentrations of aminomethylenebisphosphonic acid derivatives on exponentially growing cultures was measured as described previously (Forlani et al. 1994). Cell samples withdrawn from stock cultures in the early stationary growth phase were used to inoculate 100-mL culture flasks containing 20 mL of fresh medium to a density of about 1.0 mg mL^{-1} (dry weight). Cultures were incubated as above, and filter-sterilized bisphosphonates were added just after the density of cells reached 1.7 mg mL^{-1} (dry weight). After a further 8 days of incubation, when the untreated controls reached the late exponential phase of growth, cells were harvested by vacuum filtration, and the dry weight change was determined on each sample following drying in an oven at 90°C for 48 h. The dry weight for untreated controls was 4.6 ± 0.2 mg mL^{-1}. For each compound at least seven concentrations (ranging from 0.01 to 1 mM) with three replicates were tested. Two independent experiments were performed for cell culture; since error variances were found to be homogenous, means over the two replicates were considered.

Table 1. *N*-Substituted aminomethylenebisphosphonic acids **4** and compounds **6** and **8.**

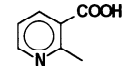
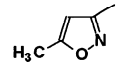
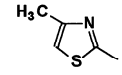
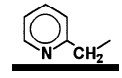
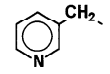
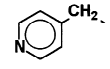
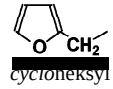
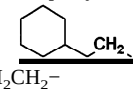
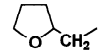
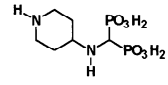
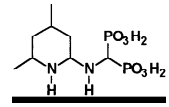
Compound	Structure R^1	Structure R^2	Yield (%)	m.p. decomp. (°C)
4aa	H	*4*-$CH_3C_6H_4$	81	242–246
4ab	H	*3*-$O_2NC_6H_4$	90	207–209
4ac	H	*3*-$HOOCC_6H_4$	14	238–239
4ad	H	*3,5*-$Cl_2C_6H_3$	87	239–243
4ae	H	*2*-$CH_3C_6H_5CH_2$	32	172–173
4af	H	*2*-$CH_3OC_6H_4CH_2$	24	231–235
4ag	H	*4*-$CH_3OC_6H_4CH_2$	50	150–151
4ah	H	*4*-$HOC_6H_4CH_2CH_2$	20	226–227
4ai	H	*3*-$O_2NC_6H_4CH_2$	31	217–220
4ba	H		48	250–252
4bb	H		14	238–239
4bc	H		47	259–260
4bd	H		29	100–102
4be	H		36	128–130
4bf	H		26	259–262
4bg	H		23	274–276
4bh	H		52	191–193
4bi	H		11	149–150
4bj	H		3	223–225
4bk	H		3.5	239–242
4ca	H	*cyclo*heksyl	67	245–247
4cb	*cyclo*heksyl	*cyclo*heksyl	22	234–236
4cc	H	*cyclo*propyl	67	272–273
4cd	H	*sec*-butyl	85	232–235
4ce	H	*n*-pentyl	77	212–214
4cf	H	*iso*-pentyl	78	228–230
4cg	H		81	225–226
4ch	$-CH_2CH_2CH_2CH_2-$		57	205–207
4da	$-CH_2CH_2OCH_2CH_2-$		70	205–210
4db	H		31	232–233
4dc	H		49	256–260
4dd	H		60	235–240
6			78	204–206
8			38	208–211

Table 2. *N*-Substituted aminobis(methylphosphonic) acids **5** and compound **7.**

Compound	Structure R^1	Yield (%)	m.p. decomp. (°C)
5a	*cyclo*hexyl	48	225–228
5b	$-CH_2C_6H_5$	51	229–232
5c	*sec*-butyl	62	256–259
5d	*iso*-pentyl	43	
5e		59	172–174
5f		87	257–259
7		78	260–266

Statistical Treatment

Dixon's *Q*-test was used to reject unreasonable results. The means for samples and controls were compared by testing the null hypothesis at the 5% significance level (Miller and Miller 1984). Concentrations causing 50% inhibition of cell growth rate (LD_{50}) were estimated using linear regression equations of the values of growth yield, expressed as a percentage of untreated controls, plotted against the logarithm of bisphosphonate concentration.

Results statistically not significant are marked in tables as N.

Results and Discussion

Our first attempt was to find out how the replacement of pyridyl moieties in compounds **1, 2,** and **3** by other substituents influenced herbicidal properties. The data collected in Table 3 show that the influence of the tested compounds on plant root development was usually more pronounced than their action on shoots. With the exception of compounds **4af, 4ai, 4be, 4bk, 4cg,** and **4da,** which were inactive, and compound **4ad,** which was stimulatory, most of the studied compounds exhibited quite significant herbicidal activity toward cress. Many of them exhibited exceptionally strong herbicidal activity. Thus, compounds **4bd, 4bh, 4ce, 4dc,** and **8** were comparably active to the most herbicidally active control compound **3,** whereas ten (compounds **4ab, 4ac, 4ae, 4ah, 4ba, 4bb, 4bc, 4bf, 4cc,** and **4db**) were comparable in activity or greater than the second control compound **1.**

The synthesized compounds represent at least four structurally diverse groups of analogs. The first one (compounds **4a**) was obtained by replacement of pyridyl moieties of model compounds **1–3** by substituted phenyl or benzyl groups of varying hydrophobicity. Among nine representatives of this class, two did not exhibit notable activity (and are not included in Table 3), and four exhibited activity similar to the control compound **1** and thus fall into the category of the strongest herbicides. We

Table 3. Effect of derivatives of aminomethylenebisphosphonic acid on the growth of cress, measured as percentage change in root and shoot weight compared with that of control.

Compound	Root or shoot	Concentration (mM) 0.05	0.15	0.5	1.5
Glyphosate	R	−(86 ± 8)	−(88 ± 4)	−(90 ± 7)	−(94 ± 9)
	S	−(13 ± 1)	−(15 ± 2)	−(20 ± 2)	−(44 ± 2)
1	R	N	−(42 ± 7)	−(50 ± 3)	−(84 ± 8)
	S	N	−(15 ± 2)	−(36 ± 7)	−(49 ± 4)
2	R	N	−(32 ± 9)	−(35 ± 7)	−(32 ± 2)
	S	N	N	N	N
3	R	−(76 ± 7)	−(82 ± 13)	−(89 ± 9)	−(95 ± 15)
	S	−(27 ± 6)	−(42 ± 9)	−(51 ± 7)	−(58 ± 9)
4aa	R	N	−(31 ± 9)	−(37 ± 8)	−(34 ± 11)
	S	N	N	−(8 ± 2)	−(16 ± 4)
4ab	R	−(16 ± 4)	−(48 ± 3)	−(82 ± 3)	−(90 ± 8)
	S	N	N	−(27 ± 3)	−(55 ± 2)
4ac	R	N	N	−(69 ± 8)	−(93 ± 11)
	S	N	N	N	−(42 ± 9)
4ad	R	+(27 ± 4)	+(40 ± 6)	+(70 ± 1)	+(25 ± 6)
	S	N	N	N	−(18 ± 6)
4ae	R	N	−(55 ± 7)	−(69 ± 9)	−(76 ± 3)
	S	N	N	N	−(25 ± 4)
4ag	R	N	N	−(24 ± 5)	−(40 ± 13)
	S	N	N	N	N
4ah	R	−(29 ± 5)	−(38 ± 8)	−(69 ± 5)	−(82 ± 4)
	S	N	N	−(32 ± 4)	−(62 ± 8)
4ba	R	−(22 ± 4)	−(50 ± 8)	−(55 ± 7)	−(88 ± 1)
	S	N	N	N	−(48 ± 3)
4bb	R	−(32 ± 5)	−(64 ± 7)	−(80 ± 11)	−(91 ± 4)
	S	N	N	N	N
4bc	R	−31	−(48 ± 13)	−(50 ± 12)	−(75 ± 10)
	S	N	N	N	−(30 ± 9)
4bd	R	−(76 ± 4)	−(81 ± 2)	−(87 ± 5)	−(92 ± 3)
	S	−(16 ± 2)	−(26 ± 5)	−(32 ± 1)	−(47 ± 4)
4bf	R	−(46 ± 7)	−(43 ± 5)	−(56 ± 11)	−(58 ± 2)
	S	−(17 ± 5)	−(22 ± 6)	−(32 ± 9)	−(30 ± 4)
4bg	R	N	N	−(17 ± 5)	−(57 ± 9)
	S	N	N	N	N
4bh	R	−(80 ± 4)	−(88 ± 3)	−(98 ± 1)	−(95 ± 2)
	S	−(51 ± 4)	−(50 ± 1)	−(64 ± 3)	−(68 ± 4)
4bi	R	−(28 ± 7)	−(33 ± 8)	−(49 ± 4)	−(69 ± 8)
	S	N	N	N	N
4bj	R	N	N	−(41 ± 4)	−(63 ± 3)
	S	N	−(15 ± 11)	−(18 ± 3)	−(21 ± 3)
4ca	R	−(35 ± 4)	−(46 ± 5)	−(52 ± 4)	−(56 ± 9)
	S	−(12 ± 4)	−(17 ± 4)	−(26 ± 4)	−(34 ± 5)
4cb	R	+(41 ± 4)	+(40 ± 7)	N	−(88 ± 2)
	S	N	N	−(32 ± 2)	−(77 ± 3)
4cc	R	−(17 ± 2)	−(43 ± 8)	−(76 ± 7)	−(91 ± 10)
	S	N	N	N	N
4cd	R	N	N	−(35 ± 13)	−(45 ± 14)
	S	N	N	N	−(16 ± 10)
4ce	R	−(64 ± 5)	−(79 ± 4)	−(82 ± 7)	−(87 ± 3)
	S	N	N	N	N
4cf	R	N	−(20 ± 2)	−(38 ± 6)	−(44 ± 12)
	S	N	N	N	N
4ch	R	−(24 ± 6)	−(30 ± 2)	−(54 ± 5)	−(65 ± 8)
	S	N	N	N	N
4db	R	−(36 ± 2)	−(59 ± 6)	−(71 ± 1)	−(74 ± 5)
	S	N	N	N	N

Table 3. Continued.

Compound	Root or shoot	Concentration (mM) 0.05	0.15	0.5	1.5
4dc	R	−(43 ± 3)	−(81 ± 6)	−(87 ± 3)	−(92 ± 7)
	S	N	−(17 ± 9)	−(41 ± 1)	−(59 ± 3)
4dd	R	N	N	N	−(79 ± 14)
	S	N	N	N	−(52 ± 11)
5e	R	N	N	N	−(45 ± 13)
	S	N	N	N	N
5f	R	N	N	−(16 ± 6)	−(77 ± 8)
	S	N	N	N	−(22 ± 11)
6	R	+33	N	−(25 ± 5)	−(54 ± 7)
	S	N	N	N	N
7	R	N	N	N	−(47 ± 12)
	S	N	N	N	−(22 ± 10)
8	R	−(58 ± 5)	−(70 ± 3)	−(86 ± 2)	−(83 ± 11)
	S	N	N	N	−(75 ± 10)

were unable to find any relationship between their herbicidal potency and hydrophobicity of the substituent.

Replacement of the aminomethylenebisphosphonic acid with heteroaromatic moieties resulted in 11 compounds **4b,** which are the closest structural analogs of *N*-pyridyl derivatives **1–3.** As might be expected most of them showed potent herbicidal activity, with compounds **4bd** and **4bh** being equally potent with the most active control compound **3** and thus being comparable in activity to glyphosate. Also, in this case, it is difficult to draw any meaningful structure-activity relationship. It is perhaps most clearly seen for compounds **4bf** and **4bg** where the minor modification, namely insertion of the methyl group into the thiazole ring of **4bf,** caused a dramatic decrease of herbicidal activity of the resulting **4bg.**

A third group of analogs (compounds **4c**) was obtained by replacing aminomethylenebisphosphonic acid with aliphatic or alicyclic substituents. Among eight compounds synthesized only *N*-(*n*-pentylamino)methylenebisphosphonic acid (compound **4ce**) was of activity equal to glyphosate. Also, in this case, no relation between activity and structure of the *N*-substituent was found. Moreover, compound **4cg,** which is the saturated analog of moderately herbicidal *N*-(benzylamino)methylenebisphosphonic acid (Lejczak et al. 1996), appeared to be completely inactive and thus is not included in Table 3.

Finally, a group of derivatives substituted with aliphatic heterocycles was obtained. Three out of four compounds **4d,** as well as compound **8,** exhibited quite strong herbicidal activity. Compounds **4dc** and **4dd** are saturated, nonaromatic analogs of compounds **1** and **2,** respectively. Whereas compound **4dc** exhibited slightly stronger herbicidal activity than the starting compound **1,** the activity of compound **4dd** was significantly different

Table 4. Effect of derivatives of aminomethylenebisphosphonic acid on the growth of cucumber, measured as percentage change in root and shoot weight compared with that of control.

Compound	Root or shoot	Concentration (mM) 0.05	0.15	0.5	1.5
Glyphosate	R	−(60 ± 4)	−(77 ± 4)	−(78 ± 7)	−(84 ± 2)
	S	N	N	N	−(22 ± 2)
1	R	−(62 ± 5)	−(74 ± 5)	−(81 ± 9)	−(81 ± 3)
	S	−(34 ± 2)	−(34 ± 6)	−(47 ± 3)	−(49 ± 5)
3	R	−(76 ± 11)	−(88 ± 12)	−100	−100
	S	−(48 ± 10)	−(66 ± 9)	−(68 ± 7)	−100
4ab	R	N	−(45 ± 3)	−(49 ± 6)	−(58 ± 5)
	S	N	N	N	N
4ac	R	−(42 ± 10)	−(46 ± 11)	−(44 ± 15)	−(48 ± 13)
	S	N	N	N	−(49 ± 10)
4ae	R	−(31 ± 5)	−(32 ± 6)	−63	−70
	S	N	N	N	−15
4ah	R	−(23 ± 6)	−(27 ± 6)	−(52 ± 4)	−(84 ± 7)
	S	−(15 ± 7)	−(20 ± 5)	−(28 ± 3)	−(48 ± 5)
4ba	R	N	N	−18	−34
	S	+(36 ± 13)	N	N	N
4bb	R	−(34 ± 3)	−(48 ± 4)	−(51 ± 3)	−(69 ± 1)
	S	N	−(31 ± 5)	−(25 ± 3)	−(67 ± 4)
4bc	R	N	−(20 ± 5)	−(51 ± 4)	−(72 ± 9)
	S	N	N	N	N
4bd	R	−(18 ± 4)	−(42 ± 4)	−(65 ± 7)	−(79 ± 7)
	S	N	N	N	−(27 ± 5)
4bh	R	−(68 ± 15)	−(72 ± 13)	−(66 ± 16)	−(77 ± 11)
	S	N	−(29 ± 11)	−(53 ± 9)	−(61 ± 10)
4bi	R	−(15 ± 7)	−(40 ± 8)	−(41 ± 9)	−(52 ± 2)
	S	N	N	N	−(27 ± 5)
4bj	R	−(35 ± 11)	−(35 ± 13)	−(41 ± 10)	−(42 ± 15)
	S	N	N	N	N
4ca	R	N	−(21 ± 6)	−(72 ± 3)	−(71 ± 11)
	S	N	N	N	N
4cb	R	N	−(30 ± 5)	−(55 ± 3)	−(70 ± 4)
	S	N	N	N	−(30 ± 5)
4cc	R	N	N	N	N
	S	N	N	N	N
4cd	R	N	N	N	−41
	S	N	N	N	N
4ce	R	−(58 ± 3)	−(66 ± 2)	−(80 ± 3)	−(86 ± 9)
	S	N	N	N	N
4ch	R	N	−(24 ± 4)	−(31 ± 6)	−(35 ± 5)
	S	N	N	N	N
4db	R	−(38 ± 4)	−(51 ± 6)	−(65 ± 4)	−(72 ± 2)
	S	+(22 ± 7)	+(18 ± 3)	+(10 ± 2)	N
4dc	R	−(47 ± 4)	−(61 ± 3)	−(69 ± 7)	−(68 ± 4)
	S	−(21 ± 3)	−(24 ± 5)	−(28 ± 6)	−(47 ± 1)
4dd	R	N	N	−(48 ± 11)	−(61 ± 10)
	S	N	N	−(35 ± 9)	−(50 ± 7)
6	R	+(33 ± 11)	+(55 ± 13)	+(38 ± 11)	N
	S	+(51 ± 5)	+(49 ± 7)	+(34 ± 9)	+(33 ± 11)
8	R	N	−(22 ± 3)	−(58 ± 6)	−(75 ± 2)
	S	N	+(20 ± 1)	−(24 ± 2)	−(28 ± 3)

from that observed for compound **2.** It was less active at low concentrations than the parent compound **2** and strongly active at high concentrations. The fact that re-

Table 5. Effect of the most active derivatives of aminomethylenebisphosphonates on the growth of *Zea mays* cultured cells.

Compound	LD_{50} (mM)	Concentration (mM) 0.01	0.02	0.05	0.1	0.2	0.5
4ah	0.14	N	−(16 ± 5)	−(36 ± 2)	−(43 ± 0.5)	−(47 ± 0.5)	−(50 ± 1)
4ai	u[a]	N	N	N	N	N	N
4ba	1.15	N	+(8 ± 5)	+(9 ± 1)	+(12 ± 5)	+(12 ± 8)	−(18 ± 5)
4bb	u[a]	N	N	N	N	N	N
4bd	0.29	+(11 ± 3)	+(14 ± 2)	−(10 ± 1.5)	−(29 ± 0.5)	−(38 ± 1)	−(42 ± 4)
4bh	0.64	N	N	−(9 ± 2)	−(22 ± 8)	−(37 ± 1)	−(44 ± 4)
4ca	0.73	−(7 ± 1)	−(21 ± 9)	−(27 ± 2)	−(31 ± 9)	−(35 ± 5)	−(47 ± 3)
4ch	3.70	N	−(7 ± 1)	−(14 ± 4)	−(20 ± 5)	−(25 ± 4)	−(34 ± 4)
4ce	1.64	N	−(16 ± 2)	−(20 ± 5)	−(23 ± 5)	−(35 ± 0.5)	−(41 ± 4)
4db	0.70	−(5 ± 1)	N	−(15 ± 1)	−(25 ± 7)	−(30 ± 5)	−(31 ± 3)

[a] u, ineffective.

duction of the pyridyl ring of compounds **1–3** resulted in compounds of equal herbicidal activity, as well as the fact that substitution of these pyridyl moieties by many other groups resulted in compounds of significant herbicidal activity, shows a wide range of activity of the bisphosphonate herbicides for modifications of this part of the molecule.

It is also worth noting that the unsubstituted aminomethylenebisphosphonic acid (Lejczak et al. 1996), as well as its homolog (compound **6**) were significantly less active than compounds **4,** indicating that substitution of the amino group of this acid is indispensable for herbicidal activity.

In summation, we were unable to find any reasonable structure-activity relationship for the synthesized compounds **4.** The lack of such a relationship suggests that multiple sites of action of the studied herbicides might exist and that there might be more than one target enzyme for their action.

N-Substituted iminodi(methylenephosphonic) acids (compounds **5**) may be considered as close structural analogs of compounds **4.** We also have determined the influence of some representatives of these compounds on the growth of cress. Only compounds **5e** and **5f** exhibited weak herbicidal activity. This finding is interesting in that there is a small steric structural difference between these two classes of compounds, and that would indicate that there is indeed tight dependence of herbicidal activity of the studied compounds on a spatial arrangement of both phosphonic groups.

The compounds most active against cress were also tested on cucumber. As in our previous studies cucumber was less sensitive than cress (Kafarski et al. 1995, Lejczak et al. 1996), and again, the influence on the growth of plant roots was usually more pronounced than the action on hypocotyls. Results presented in Table 4 show that there is nearly the same pattern of activity of the studied compounds toward both plants. The only exception is a tolerance of cucumber to the action of compound **4cc,** which exhibited exceptionally strong herbicidal activity on cress. The herbicidal activity observed for many representatives of *N*-substituted aminomethylenebisphosphonic acids indicates their potential usefulness as a new class of herbicides and justifies further search for new compounds of this type.

We have also checked the herbicidal activity of chosen representatives of our compounds using cultured cells of maize. Results presented in Table 5 show that the compounds most active on intact plants are also active in cell suspension, but they were significantly less active. Moreover, the structure-activity relationship was quite different from those found for cress and cucumber. At least two target sites for the action of the derivatives of aminomethylenebisphosphonic acid were proposed: 3-deoxy-D-arabino-heptulosonate-7-phosphate (Forlani et al. 1996) and farnesyl pyrophosphate synthase (Cromartie and Fisher 1995). The observed differences in the action of our compounds on whole plants and cell suspension system may derive from the fact that the growth inhibition resulting from inhibition of farnesyl pyrophosphate synthase, an intermediate in the biosynthesis of carotenoids and chlorophylls, which are not required for growth by heterotrophically grown cultured cells, is not expressed in the latter system. This finding, once more, supports the possibility of multiple action sites of the derivatives of aminomethylenebisphosphonic acid.

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