Acknowledgment. The authors wish to thank Dr. L. D. Freedman for suggesting the dephosphonylation reactions, Dr. R. E. Lutz for samples of dibenzoylethylene and 2,5-diphenylfuran, and the Monsanto Chemical Company, Victor Chemical Works, and the National Aniline Division, Allied Chemical Corporation, for generous sample of starting materials.

PITTSBURGH 13, PA.

[CONTRIBUTION NO. 30 FROM THE L. G. RYAN RESEARCH LABORATORIES OF MONSANTO CANADA LIMITED AND THE RESEARCH LABORATORIES OF AYERST, MCKENNA AND HARRISON LIMITED]

Bacteriostats. IV.[1] ω-Amino Acid Amide Derivatives

D. L. GARMAISE,[2] R. W. KAY,[2] G. Y. PARIS,[2] R. J. RANZ,[2] R. GAUDRY,[3] H. A. BAKER,[3] AND A. F. MCKAY[3]

Received June 15, 1960

A number of *N,N'*-disubstituted ω-amino acid amides have been prepared for evaluation as bacteriostats. The preparations and bacteriostatic properties of the compounds are described.

N,N'-Disubstituted glycineamides have been prepared for evaluation as tuberculostats[4] and tri- and tetrasubstituted glycineamides have been patented as pharmacological agents.[5] Some of the products described by Bersch and Döpp,[4] especially *N,N'*-di(4-ethoxyphenyl)- and *N,N'*-di(4-butoxyphenyl)glycineamides showed high activity *in vitro* against *M. tuberculosis*. Thus it was considered to be of interest to determine the general bacteriostatic effectiveness of *N,N'*-disubstituted ω-amino acid amides, $RNH(CH_2)_nCONHR'$. The substituents were varied to include both aryl and aralkyl groups which are known to increase bacteriostatic activity in other structures.[6]

A variety of methods are available for the preparation of *N,N'*-disubstituted amino acid amides. The most general procedure consists of treating an ω-haloacyl chloride with an amine to yield the *N*-substituted ω-haloacid amide, which is then heated with an amine to yield the desired product.[5] Symmetrically substituted products may be prepared in a single step by refluxing the amine and the haloacyl chloride in toluene in the presence of sodium carbonate. The symmetrically substituted glycineamides also may be obtained by refluxing an amine with glyoxal sodium bisulfite.[7,8] However, the most convenient process for a large scale laboratory preparation of symmetrically substituted glycineamides is the condensation of an amine with ethyl chloroacetate at 125–140°.[9] Symmetrically substituted 3-aminopropionamides are readily prepared in one step by heating an amine with acrylic acid.[10]

Bacteriostatic activities. It may be seen from the results in Table I that chain length is of secondary importance in determining bacteriostatic effectiveness. For values of *n* of 1, 2, and 5 the most effective substituent on either nitrogen atom was the 3,4-dichlorophenyl group. The most active single compound was *N,N'*-di(3,4-dichlorophenyl)-3-aminopropionamide and the homologous glycineamide and 6-aminocaproamide were slightly less effective. When the functional groups were separated by a longer polymethylene chain, as in the undecanoamide derivatives ($n = 10$), the 3,4-dichlorobenzyl group was considerably more effective than the 3,4-dichlorophenyl group when substituted on the amino nitrogen. This result is in accord with other observations made on compounds in which the bacteriostatic activity is largely due to an isolated basic functional group.

N,N'-Di(3,4-dichlorophenyl)glycineamide hydrochloride has a low acute toxicity for mice (>1.2 g./kg.). However, its bacteriostatic potency was considerably reduced in the presence of serum. The minimal inhibitory concentration for *Strept. faecalis*, for example, fell from 1:1,280,000 to 1:160,000 in the presence of serum. *N,N'*-Di(3,4-dichlorophenyl)-3-aminopropionamide showed a similar deactivation by serum.

EXPERIMENTAL[11]

Amines. 3-Phenylpropylamine was prepared by the modified phthalimide synthesis[12] from 3-phenylpropyl bromide.

(1) *Bacteriostats.* III. A. F. McKay, D. L. Garmaise, G. Y. Paris and S. Gelblum, *Can. J. Chem.*, **38**, 343 (1960).

(2) Monsanto Canada Limited, LaSalle, Quebec.

(3) Ayerst, McKenna and Harrison Limited, Montreal, Quebec.

(4) H. W. Bersch and W. Döpp, *Arzneimittel-Forsch.*, **5**, 183, 335 (1955).

(5) W. F. Bruce and J. Seifter, Canadian Pat. 563,659; 563,660; 563,661 (1958), and 579,667 (1959).

(6) A. F. McKay, D. L. Garmaise, R. Gaudry, H. A. Baker, G. Y. Paris, R. W. Kay, G. E. Just, and R. Schwartz, *J. Am. Chem. Soc.*, **81**, 4328 (1959).

(7) O. Hinsberg, *Ber.*, **21**, 110 (1888).

(8) K. Maurer and E. H. Woltersdorf, *Z. Physiol. Chem.*, **254**, 18 (1938).

(9) P. J. Meyer, *Ber.*, **8**, 1152 (1875).

(10) W. Autenrieth and C. Pretzell, *Ber.*, **36**, 1262 (1903).

(11) All melting points are uncorrected. Microanalyses were performed by Micro-Tech Laboratories, Skokie, Ill.

(12) J. C. Sheehan and W. A. Bolhofer, *J. Am. Chem. Soc.*, **72**, 2786 (1950).

TABLE I

BACTERIOSTATIC ACTIVITIES (M.I.C.,[a] 1 X 10^{-3}) OF ω-AMINO ACID AMIDES, $RNH(CH_2)_nC(O)NHR'$

R	R′	n	*M. pyogenes*[b] *var. aureus* (S)	*M. pyogenes*[c] *var. aureus* (R)	*Sarcina lutea*	*Strept. faecalis*	*E. coli* #198	*A. aerogenes*	*S. pullorum*	*Ps. aeruginosa*	*Pr. mirabilis*	*Pr. vulgaris*
3,4-Dichlorophenyl	3,4-Dichlorophenyl	1[d]	1280	1280	1280	1280	20	20	10	10	10	10
4-Chlorobenzyl	4-Chlorobenzyl	1[d]	10	10	20	20	20	10	10	<10	10	10
2,4-Dichlorobenzyl	2,4-Dichlorobenzyl	1[d]	80	40	80	80	40	10	<10	20	40	40
3,4-Dichlorobenzyl	3,4-Dichlorobenzyl	1[d]	80	80	160	80	80	10	20	<10	40	40
3,4-Dichlorobenzyl	3,4-Dichlorophenyl	1[d]	160	160	<320	160	10	10	10	10	10	10
3,4-Dichlorophenyl	3,4-Dichlorophenyl	2	2560	1280	2560	1280	160	20	20	10	40	40
3,4-Dichlorobenzyl	3,4-Dichlorobenzyl	2[d]	320	160	320	160	80	40	20	20	160	80
4-Ethylphenyl	4-Ethylphenyl	2	10	10	<10	<10	<10	10	<10	10	20	<10
4-Ethoxyphenyl	4-Ethoxyphenyl	2	<10	<10	<10	<10	10	<10	<10	<10	<10	<10
3,4-Dichlorophenyl	3,4-Dichlorophenyl	5	1280	640	1280	160	40	20	20	10	40	20
3,4-Dichlorobenzyl	3,4-Dichlorobenzyl	5[d]	80	160	80	80	80	40	40	10	20	10
3,4-Dichlorobenzyl	3,4-Dichlorophenyl	5[d]	640	320	640	160	320	40	40	20	20	20
3,4-Dichlorophenyl	3,4-Dichlorophenyl	10	10	10	40	40	10	10	10	<10	10	10
3,4-Dichlorobenzyl	3,4-Dichlorophenyl	10[d]	160	160	640	1280	320	20	20	10	80	80

[a] Minimal inhibitory concentration determined by serial dilution technique, *e.g.*, the value 1280 is equivalent to a dilution of 1 part in 1,280,000. [b] (S) indicates Penicillin sensitive. [c] (R) indicates Penicillin resistant. [d] Evaluated as the hydrochloride.

TABLE II

ω-HALOACID AMIDES, $X(CH_2)_nCONHR$

Compound	Yield, %	M.P.	Formula	Carbon, % Calcd.	Carbon, % Found	Hydrogen, % Calcd.	Hydrogen, % Found	Halogen,[a] % Calcd.	Halogen,[a] % Found	Nitrogen,[a] % Calcd.	Nitrogen,[a] % Found
N-(3,4-Dichlorophenyl)-3-chloropropionamide	87	110–111[b]	$C_9H_8Cl_3NO$	42.79	42.81	3.27	3.19	42.21	42.12	5.50	5.55
N-(3,4-Dichlorobenzyl)-3-chloropropionamide	54	108–110[b]	$C_{10}H_{10}Cl_3NO$	45.05	45.05	3.67	3.78	40.10	39.90	5.33	5.26
N-(3,4-Dichlorophenyl)-6-bromocaproamide	98	65–66[c]	$C_{12}H_{14}BrCl_2NO$	42.50	43.02	4.16	4.21	44.48	44.05	4.13	3.91
N-(3,4-Dichlorobenzyl)-6-bromocaproamide	44	75–76[c]	$C_{13}H_{16}BrCl_2NO$	44.22	44.54	4.57	4.57	42.72	42.73	3.97	4.16
N-(3,4-Dichlorophenyl)-11-bromoundecanoamide	78	83–84[c]	$C_{17}H_{24}BrCl_2NO$	49.90	49.92	5.91	5.90	36.86	36.51	3.42	3.32
N-(3,4-Dichlorophenyl)-2-chloroacetamide	84	106.5–107[b]	$C_8H_6Cl_3NO$	40.28	40.39	2.54	2.56	44.60	44.43	5.87	5.83
N-(3,4-Dichlorobenzyl)-11-bromoundecanoamide	49	67–68[c]	$C_{18}H_{26}BrCl_2NO$	51.08	50.88	6.19	6.14	35.63	35.88	3.31	3.33

[a] Refers to total halogen. [b] Crystallized from aqueous ethanol [c] Ether–petroleum ether.

TABLE III

ω-AMINO ACID AMIDES, $RNH(CH_2)_nCONHR'$

R	R'	n	Yield, %	M.P.	Formula	Carbon, % Calcd.	Carbon, % Found	Hydrogen, % Calcd.	Hydrogen, % Found	Chlorine, % Calcd.	Chlorine, % Found	Nitrogen, % Calcd.	Nitrogen, % Found
4-Chlorobenzyl	4-Chlorobenzyl	1[a]	29[f]	266 (dec.)[h]	$C_{16}H_{17}Cl_3N_2O$	53.43	53.29	4.76	4.84	29.57	29.58	7.79	7.87
			71[g]	179–180[i]	$C_{22}H_{19}Cl_2N_5O_8$	47.84	48.25	3.46	3.65	12.84	12.97	12.68	13.01
2,4-Dichlorobenzyl	2,4-Dichlorobenzyl	1[a]	50[f]	224–225[h]	$C_{16}H_{15}Cl_5N_2O$	44.85	44.76	3.53	3.65	41.37	41.70	6.59	6.79
3,4-Dichlorobenzyl	3,4-Dichlorobenzyl	1[a]	49[f]	245[j]	$C_{16}H_{15}Cl_5N_2O$	44.85	44.77	3.53	3.61	41.37	41.61	6.54	6.93
3,4-Dichlorophenyl	3,4-Dichlorophenyl	1[b]		147–148[k,l]	$C_{14}H_{10}Cl_4N_2O$	46.18	46.32	2.77	2.94	38.96	38.90	7.70	8.08
			29[f]	201.5–202.5[m]	$C_{14}H_{11}Cl_5N_2O$	41.99	42.26	2.77	2.89	44.26	44.61	6.99	6.98
3,4-Dichlorobenzyl	3,4-Dichlorophenyl	1[c]	41[f]	245–245.5[h]	$C_{15}H_{13}Cl_5N_2O$	43.45	43.59	3.16	3.25	42.77	42.53	6.76	6.98
			94[g]	195.5–196.5[k]	$C_{21}H_{17}Cl_4N_5O_8$	41.54	41.54	2.49	2.76	23.35	23.28	11.54	11.73
2-Phenylethyl	2-Phenylethyl	1[d]	76[f]	233–234[h,n]									
			85[g]	124–126[k]	$C_{24}H_{25}N_5O_8$	56.36	55.95	4.93	4.88			13.70	14.15
3-Phenylpropyl	3-Phenylpropyl	1[b]	54[f]	146.5–148[m]	$C_{20}H_{27}ClN_2O$	69.24	69.56	7.85	7.82	10.22	9.99	8.08	8.08
			99[g]	139–140[h]	$C_{26}H_{29}N_5O_8$	57.88	58.14	5.42	5.58			12.98	13.27
4-Phenylbutyl	4-Phenylbutyl	1[d]	61[f]	130.5–132.5[m]	$C_{22}H_{31}ClN_2O$	70.47	70.45	8.33	8.32	9.46	9.65	7.47	7.68
			98[g]	122–123[h]	$C_{28}H_{33}N_5O_8$	59.25	59.41	5.86	6.09			12.34	12.06
2-(4-Hydroxyphenyl)-ethyl	2-(4-Hydroxyphenyl)-ethyl	1[d]	33[f]	233–234[h]	$C_{18}H_{23}ClN_2O_3$	61.61	61.38	6.61	6.81	10.11	10.31	7.99	7.72
			69[g]	149–151[k]	$C_{24}H_{25}N_5O_{10}$	53.03	52.97	4.64	4.84			12.89	12.46
2-(3,4-Dimethoxy-phenyl)ethyl	2-(3,4-Dimethoxy-phenyl)ethyl	1[b]	56[f]	182–183[h]	$C_{22}H_{31}ClN_2O_5$	60.20	60.48	7.12	7.06	8.08	7.76	6.38	6.12
3,4-Dichlorobenzyl	3,4-Dichlorobenzyl	2[c]	78[f]	252–253[h]	$C_{17}H_{17}Cl_5N_2O$	46.31	46.14	3.90	3.87	40.04	40.05	6.41	6.33
3,4-Dichlorophenyl	3,4-Dichlorophenyl	2[c]	71	114–115[k]	$C_{15}H_{12}Cl_4N_2O$	47.76	47.65	3.18	3.20	37.36	37.51	7.25	7.41
4-Ethoxyphenyl	4-Ethoxyphenyl	2[e]	48	141–142[h]	$C_{19}H_{24}N_2O_3$	69.49	69.67	7.39	7.49			8.53	8.77
4-Ethylphenyl	4-Ethylphenyl	2[e]	43	129–130[g]	$C_{19}H_{24}N_2O$	76.98	76.85	8.16	8.08			9.41	9.77
2-Phenylethyl	2-Phenylethyl	2[c]	73[f]	235.5–236[h]	$C_{19}H_{25}ClN_2O$	68.55	68.65	7.57	7.69	10.65	10.31	8.42	8.54
			95[g]	100–101	$C_{25}H_{27}N_5O_8$	57.14	57.23	5.18	5.13			13.33	13.54
3,4-Dichlorobenzyl	3,4-Dichlorobenzyl	5[c]	52[f]	214–215[h]	$C_{20}H_{23}Cl_5N_2O$	49.56	49.55	4.78	4.64	36.58	36.59	5.78	5.49
3,4-Dichlorobenzyl	3,4-Dichlorophenyl	5[c]	43[f]	216.5–218[h]	$C_{19}H_{21}Cl_5N_2O$	48.48	48.31	4.50	4.44	37.67	37.54	5.95	5.92
3,4-Dichlorophenyl	3,4-Dichlorophenyl	5[c]	58[f]	169–179[h]	$C_{18}H_{19}Cl_5N_2O$	47.34	47.62	4.19	4.24	38.82	38.55	6.13	6.36
3,4-Dichlorophenyl	3,4-Dichlorophenyl	10[e]	47[f]	129–130[h]	$C_{23}H_{29}Cl_5N_2O$	52.44	52.20	5.55	5.40	33.65	33.88	5.32	5.23
3,4-Dichlorobenzyl	3,4-Dichlorophenyl	10[e]	55[f]	184–185[k]	$C_{24}H_{31}Cl_5N_2O$	53.31	53.43	5.78	5.86	32.79	32.65	5.18	5.21

[a] Prepared by method C. [b] Method B. [c] Method A. [d] Method D. [e] Method E. [f] Hydrochloride. [g] Picrate. [h] Crystallized from ethanol. [i] Water. [j] Methanol. [k] Aqueous ethanol. [l] Prepared from the hydrochloride in 94% yield. [m] Crystallized from ethanol-ether. [n] Literature reported m.p. 231° (J. von Braun and W. Munch, *Ber.* 60, 345 (1927)). [o] Crystallized from chloroform–petroleum ether.

The product was obtained in 87% yield, b.p. 97.5–98.5°/10 mm. (lit.,[13] b.p. 112–114°/18 mm.).

4-Phenylbutylamine was prepared by the reduction of 4-phenylbutyronitrile with lithium aluminum hydride-aluminum chloride according to the general procedure of Nystrom.[14] The product was obtained in 89% yield boiling at 114° at 12 mm. (lit.,[15] b.p. 123–124° at 17 mm.).

ω-Haloacid amides. N-(3,4-Dichlorophenyl)-3-chloropropionamide. β-Chloropropionyl chloride (13 g., 0.10 mole) in benzene (100 ml.) was added dropwise to a stirred benzene (150 ml.) solution of 3,4-dichloroaniline (32.5 g., 0.20 mole) at 20°. The precipitate of 3,4-dichloroaniline hydrochloride (19 g., 95% recovery) was filtered, and the filtrate was evaporated to dryness. Crystallization from ether-petroleum ether (b.p. 60–90°) gave the product melting at 108–110°, yield 22 g. (87%). Recrystallization from dilute ethanol solution raised the melting point to 110–111°.

The other compounds listed in Table II were prepared in the same manner.

Preparation of ω-aminoalkylcarboxamides. Method A. N,N'-Di(3,4-dichlorophenyl)-3-aminopropionamide. A mixture of *N*-(3,4-dichlorophenyl)-3-chloropropionamide (12.7 g., 0.05 mole) and 3,4-dichloroaniline (16.2 g., 0.10 mole) was stirred at 180° for 30 min. The reaction mixture was partitioned between 5% sodium carbonate solution (500 ml.) and ether (500 ml.). The ether solution was dried and evaporated, and the residue was steam-distilled until 3,4-dichloroaniline no longer appeared in the distillate. The nonvolatile residue was extracted with ether (300 ml.) and the ether extract was concentrated to a small volume and diluted with petroleum ether to give the product melting at 108–112°, yield 13.5 g. (71%). Crystallization from dilute ethanol raised the melting point to 114.5–115.5°. The compounds prepared by Methods A, B, C, D, and E are described in Table III.

Method B. N,N'-Di(3-phenylpropyl)aminoacetamide hydrochloride. Chloracetyl chloride (8.48 g., 0.075 mole) was added dropwise to a stirred mixture of 3-phenylpropylamine (20.3 g., 0.15 mole) and sodium carbonate (15.9 g., 0.15 mole) in toluene (75 ml.) at 10°. The mixture was then refluxed for 2 hr., while the water liberated in the reaction (1.4 ml.; theory, 1.4 ml.) was removed azeotropically in a Barrett trap. The suspension was filtered and the filtrate was shaken with 3*N* hydrochloric acid (200 ml.). The crude product (m.p. 128–145°) separated at the interface. Crystallization from ethanol-ether raised the melting point to 146.5–148°, yield 14.0 g. (53.8%).

Method C. N,N'-Di(2,4-dichlorobenzyl)aminoacetamide hydrochloride. A mixture of 2,4-dichlorobenzylamine (19 g., 0.108 mole) and glyoxal sodium bisulfite (14.4 g., 0.0504 mole) in 50% aqueous ethanol (160 ml.) was refluxed for 22 hr. The mixture was evaporated to dryness *in vacuo* and the residue was extracted with boiling ethanol (150 ml.). The ethanol extract was concentrated to about 50 ml., and ether (300 ml.) was added. On passing dry hydrogen chloride through the solution the crude product (m.p. 221–223°) precipitated, yield 19.5 g. (50%). Crystallization from ethanol raised the melting point to 224–225°.

Method D. N,N'-Di(2-phenylethyl)aminoacetamide hydrochloride. Ethyl chloroacetate (163 g., 1.33 mole) was added dropwise with stirring to 2-phenylethylamine (485 g., 4.0 moles) without external cooling. During the addition period of 45 min. the temperature rose to 125°. The reaction mixture was heated at 135–140° for 1 hr. and the ethanol from the reaction was removed by distillation. Water (2 l.) and 2*N* hydrochloric acid (712 ml., 1.42 moles) were added at 75°, and the solution was allowed to cool overnight. The crude product (m.p. 220–222°) was recovered by filtration, yield 357 g. (84%). Extraction with acetone (1 l.) removed the colored impurities, 325 g. (76.2%). Crystallization from 1-propanol (5 ml./g.) raised the melting point from 230–232° to 233–234°. (lit.,[16] m.p. 231°).

Method E. N,N'-Di(4-ethoxyphenyl)-3-aminopropionamide. *p*-Phenetidine (137 g., 1.0 mole) was heated with acrylic acid (36.0 g., 0.5 mole) at 180–190° for 5 hr. Crystallization of the dark-brown reaction mixture from dilute ethanol gave the product melting at 140–141°, yield 82 g. (47.8%). Recrystallization from dilute ethanol raised the melting point to 141–142°.

LaSalle, Quebec

(13) J. von Braun, G. Blessing, and F. Zobel, *Ber.*, 56, 1988 (1923).

(14) R. F. Nystrom, *J. Am. Chem. Soc.*, 77, 2544 (1955).

(15) J. von Braun, *Ber.*, 43, 2837 (1910).

(16) J. von Braun and W. Munch, *Ber.*, 60, 345 (1927).

[Contribution from the Department of Biological Sciences, Stanford Research Institute]

Potential Anticancer Agents.[1] XLVII. Alkylating Agents Related to Phenylalanine Mustard. IV.[2] Transformation Products of Ethyl *o*-Amino-α-benzamidocinnamate

ABELARDO P. MARTINEZ, W. A. SKINNER, WILLIAM W. LEE, LEON GOODMAN, AND B. R. BAKER

Received June 23, 1960

Ethyl *o*-amino-α-benzamidocinnamate (VI) readily rearranged to ethyl *o*-benzamidopyruvate (XIII) in aqueous acetic acid at room temperature, indicating that VI had a *cis*-relationship of the *o*-aminophenyl and α-benzamido groups. When VI was treated with hydrazine at room temperature, a hydrazide (VIII) was obtained with the reverse conformation, that is, the *o*-aminophenyl and benzamido groups were *trans*, as VIII was readily cyclized to 3-benzamidocarbostyril (XI).

The *p*-isomer of phenylalanine mustard has excellent anticancer properties in transplanted experimental tumors[2] and some utility in man. As the *m*-isomer of phenylalanine mustard appears to have a better chemotherapeutic index than the *p*-isomer against some tumors such as Sarcoma 180

(1) This work was carried out under the auspices of the Cancer Chemotherapy National Service Center, National Cancer Institute, National Institutes of Health, Public Health Service, Contract No. SA-43-ph-1892. The opinions expressed in this paper are those of the authors and are not necessarily those of the Cancer Chemotherapy National Service Center. For the preceding paper in this series, *cf.* W. A. Skinner, A. P. Martinez, and B. R. Baker, *J. Org. Chem.*, 26, 152 (1961).