TABLE I

N-(2-CHLOROETHYL)-DL-β-AMINOBUTYRIC ACID HYDROCHLORIDE

$$ROOCCH_2CHCH_3$$
$$|$$
$$NHCH_2CH_2X$$

Compd no.	R	X	Yield, %	Mp, °C	Recrystn solvent[a]	Formula	Calcd, % C	H	N	Cl	Found, % C	H	N	Cl
V	H	OH	82	181[b]	A	$C_6H_{13}NO_3$								
VI	CH_3	OH	95	Oil	..	$C_7H_{16}NO_3Cl$			7.09	17.90			6.80	16.70
VII	CH_3	Cl	90	94	B	$C_7H_{15}NO_2Cl_2$	38.80	6.94	6.48	32.87	38.17	7.55	6.56	32.56
VIII	H	Cl	96	Syrup	..	$C_6H_{13}NO_2Cl_2$			6.98	35.10			6.89	34.92

[a] A = dimethylformamide, B = ethyl acetate–acetone (4:1) or acetonitrile. [b] Lit.[3a] mp 181°.

TABLE II

N-(2-CHLOROETHYL)-DL-β-AMINOISOBUTYRIC ACID HYDROCHLORIDE

$$ROOCCHCH_2NHCH_2CH_2X$$
$$|$$
$$CH_3$$

Compd no.	R	X	Yield, %	Mp, °C	Recrystn solvent[a]	Formula	Calcd, % C	H	N	Cl	Found, % C	H	N	Cl
IX[b]	H	OH	46	158	A	$C_6H_{13}NO_3$	48.97	8.84	9.53		48.72	9.03	9.75	
X	CH_3	OH	90	62–64	B	$C_7H_{16}NO_3Cl$	42.53	8.10	7.09	17.90	42.56	8.43	6.79	17.10
XI	CH_3	Cl	92	131–132	C	$C_7H_{15}NO_2Cl_2$	38.80	6.94	6.48	32.87	38.57	7.37	6.44	32.81
XII	H	Cl	96	Syrup	..	$C_6H_{13}NO_2Cl_2$			6.98	35.10			6.92	34.95

[a] A = dimethylformamide, B = acetone, C = ethyl acetate. [b] The starting material was methacrylic acid and a reflux period of 90 min was necessary.

the reactivity of the alkylating groups but also by the molecular configuration of the nonalkylating or "prosthetic" moiety. Three amino acid nitrogen mustards were synthesized for use in the study of the relation of structure to mutagenic activity. None of the compounds tested showed significant activity.

Experimental Section

N-(2-Hydroxyethyl)amino acids were prepared using a slight modification of the literature procedure.[3] Since the N-(2-chloroethyl)amino acids were synthesized by essentially identical experimental procedures, specific data will be given for only one compound; data on the other analogs will be presented in Tables I and II. All melting points (not corrected) were determined in capillaries.

β-Methyl N-(2-Hydroxyethyl)-DL-aspartate (I).—A solution of 9.8 g (0.1 mole) of maleic anhydride in 25 ml of absolute methanol was heated at reflux for 30 min, and the excess methanol was distilled *in vacuo*. The light yellow reaction mixture, after cooling in ice, was treated dropwise (stirring) with 30 ml of ice-cooled pyridine. Then, 6.1 g (0.1 mole) of ethanolamine was added, and the solution was refluxed for 1 min. The solution was left to cool, and the crystalline product was filtered, triturated once in hot acetone and once in methanol, and recrystallized from dimethylformamide giving I (8.75 g, 46%, mp 184°).

Anal. Calcd for $C_7H_{13}NO_5$: C, 43.97; H, 6.80; N, 7.32. Found: C, 44.29; H, 6.87; N, 7.25.

Dimethyl N-(2-Hydroxyethyl)-DL-aspartate Hydrochloride (II).—To 16 ml of cooled (−10°) methanol was added slowly with stirring, 4.76 g (0.02 mole) of purified thionyl chloride, then 3.8 g (0.02 mole) of I. The solution was left at room temperature for 30 min, and the methanol was eliminated under reduced pressure. The evaporation was repeated each time after the addition of three 5-ml portions of methanol and two 8-ml portions of methanol–carbon tetrachloride to afford 5 g of a hygroscopic product, which was dissolved in 10 ml of methanol (Norit), filtered, and precipitated with 30 ml of dry ether. The white product was filtered, washed with 10 ml of ether, and dried (high vacuum, P_2O_5) to yield II (4.15 g, 86%, mp 122°).

Anal. Calcd for $C_8H_{16}NO_5Cl$: C, 39.75; H, 6.62; N, 5.79; Cl, 14.82. Found: C, 39.60; H, 6.87; N, 5.65; Cl, 14.76.

Dimethyl N-(2-Chloroethyl)-DL-aspartate Hydrochloride (III).—To a stirred suspension of 8.45 g (0.035 mole) of II in 30 ml of $CHCl_3$ was added a solution of 8.3 g (0.07 mole) of thionyl chloride in 20 ml of $CHCl_3$. The mixture was stirred at room temperature for 10 min, then at reflux temperature for 40 min, and evaporated *in vacuo* to an oil. The evaporation was repeated after each of three additions of 15-ml portions of $CHCl_3$ and two 10-ml portions of methanol. The crystals were collected, washed with ethyl acetate, and dried. Recrystallization from ethyl acetate–acetonitrile afforded III (8.20 g, 90%, mp 150°).

Anal. Calcd for $C_8H_{15}NO_4Cl_2$: C, 36.92; H, 5.76; N, 5.38; Cl, 27.33. Found: C, 36.48; H, 6.02; N, 5.29; Cl, 27.16.

N-(2-Chloroethyl)-DL-aspartic Acid Hydrochloride (IV).—A solution of 1 g (0.004 mole) of III in 10 ml of concentrated HCl was refluxed for 20 hr. At the end, it was evaporated to dryness under reduced pressure, three times with water and two with benzene, to afford IV, a very hygroscopic syrup which could not be characterized. The yield was essentially quantitative. No suitable solvent for crystallization was found, and no crystalline derivative was obtained, using picric and picrolonic acids and ammonium reineckate.

Anal. Calcd for $C_6H_{11}NO_4Cl_2$: N, 6.08; Cl, 30.87. Found: N, 5.95; Cl, 30.52.

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Some Derivatives of Natural Isoflavones

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Isoflavones and their glycosides are widely distributed in plants. Some of them have been found responsible for disorders of the female reproductive system in cattle[1] and in experimental studies have shown estrogenic activity.[2] These findings and the structural relationships which may be envisaged between

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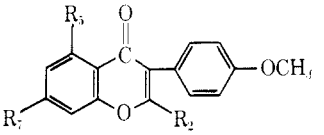
TABLE I

FORMONONETIN AND BIOCHANIN A DERIVATIVES



Compd	R_2	R_5	R_7	Method	Mp, °C	Recrystn solvent[a]	Formula	%C Calcd	%C Found	%H Calcd	%H Found	%N Calcd	%N Found	%other Calcd	%other Found
I	H	H	$OCH_2CH_2N(CH_3)_2$	A	139–141	M	$C_{20}H_{21}NO_4$	70.78	70.26	6.24	6.51	4.13	4.12		
II	H	H	$OCH_2CH_2N(C_2H_5)_2$	A, B	102.5–104	PE	$C_{22}H_{25}NO_4$	71.91	71.61	6.86	6.82	3.81	3.63		
	Hydrochloride				188–190	A								Cl: 8.59	8.58
III	CH_3	H	$OCH_2CH_2N(C_2H_5)_2$	A		PE	$C_{23}H_{27}NO_4$	72.41	72.85	7.13	7.05	3.67	3.73		
	Hydrochloride				219–221	A–E								Cl: 8.48	8.51
IV	H	OH	$OCH_2CH_2N(CH_3)_2$	A	119–120	PE	$C_{20}H_{21}NO_5$	67.59	68.04	5.96	6.14	3.92	3.91		
V	H	OH	$OCH_2CH_2N(C_2H_5)_2$	A, B	78–80	PE	$C_{22}H_{25}NO_5$	68.91	69.13	6.57	6.43	3.65	3.65		
	Hydrogen sulfate				180–182	M						2.91	2.95	S: 6.65	6.75
VI	H	OH	OCH_2CH_2N 	A	80–81	PE	$C_{22}H_{23}NO_5$					3.51	3.14		
VII	H	OH	$OCH_2CH_2CH_2N(CH_3)_2$	A	90–91	PE	$C_{21}H_{23}NO_5$					3.79	3.71		
VIII	CH_3	OH	$OCH_2CH_2N(CH_3)_2$	A	115–117	PE	$C_{22}H_{25}NO_5$	68.28	68.18	6.28	6.42	3.79	3.69		
IX	CH_3	OH	$OCH_2CH_2N(C_2H_5)_2$	A	71.5–72.5	PE	$C_{24}H_{27}NO_5$	69.50	69.47	6.85	6.90	3.52	3.56		
	Hydrochloride				239–242	M–E								Cl: 8.17	8.31
X	H	H	OCH_2CH_2Br	B	174–175	A	$C_{18}H_{16}BrO_4$							Br: 21.35	20.80
XI	H	OH	OCH_2CH_2Br	B	163–165	B	$C_{18}H_{15}BrO_5$	55.26	56.02	3.86	4.06			Br: 20.43	20.70
XII	H	H	$OCH_2COOC_2H_5$	C	142–144	EA	$C_{20}H_{18}O_6$	67.79	67.45	5.12	5.12				
XIII	H	H	OCH_2COOH		223–225	DMF–M	$C_{18}H_{14}O_6$	66.26	66.59	4.32	4.51				
XIV	CH_3	H	$OCH_2COOC_2H_5$	C	111–113	B–PE	$C_{21}H_{20}O_6$	68.47	68.43	5.47	5.31				
XV	H	OH	$OCH_2COOC_2H_5$	C	160–163	EA	$C_{20}H_{18}O_7$	64.86	64.52	4.90	4.97				

[a] A = ethanol, B = benzene, E = ether, EA = ethyl acetate, PE = petroleum ether (bp 80–120°), M = methanol.

isoflavones and estrogens (such as estradiol and diethylstilbestrol)[3] as well as between isoflavones and bioflavonoids have prompted us to synthesize a series of derivatives of two natural isoflavones, biochanin A (5,7-dihydroxy-4-methoxyisoflavone) and formononetin (7-hydroxy-4′-methoxyisoflavone), different in estrogenic potency.[2] The new compounds are shown in Table I.

Experimental Section[4]

Biochanin A and formononetin were prepared by the Baker method.[5] The final step (decarboxylation of the corresponding 2-carboxylic acids) was accomplished, for both compounds, by heating under nitrogen at 300° for 15 min and subliming the crude products at 10^{-3} mm; yields, 88–90%. 2-Methylbiochanin A and 2-methylformononetin were synthesized by known methods.[6]

7-Diethylaminoethoxy-5-hydroxy-4′-methoxyisoflavone (V). Method A.—To a stirred suspension of 5.68 g (0.02 mole) of biochanin A in 40 ml of anhydrous methanol was added 0.02 mole of $NaOCH_3$ (9.2 ml of 11.75% solution in methanol). After a few minutes, 60 ml of xylene was added and methanol was distilled completely under reduced pressure. Diethylaminoethyl chloride (4 g, 0.02 mole) was added, and the mixture was heated in an oil bath at 110° for 2 hr, then filtered and extracted with 10% acetic acid. The acid extract was filtered and rendered slightly alkaline with NH_4OH. The precipitate was collected, dried, and recrystallized from petroleum ether (bp 80–120°) giving 6.7 g (88%) of V, mp 78–80°. The ultraviolet spectrum (λ_{max} 260 mμ in ethanol and in 4% ethanolic sodium acetate, 272 mμ in 4% ethanolic $AlCl_3 \cdot 6H_2O$) was consistent with a free 5-hydroxyl group.[7] The infrared spectrum, similarly, did not show absorption in 3500–3300-cm^{-1} region.[8] The hydrogen sulfate, precipitated by adding concentrated H_2SO_4 to an acetone solution of V and recrystallized from methanol had mp 180–182°.

Method B.—To a solution of 5.68 g (0.02 mole) of biochanin A in 130 ml of anhydrous Cellosolve was added 0.02 mole of $NaOCH_3$ (9.2 ml of 11.75% solution in methanol). The mixture was distilled until the boiling point of Cellosolve was reached, then 30 ml of dibromoethane was added, and the mixture was refluxed for 2 hr and evaporated to dryness under reduced pressure. The residue was triturated with 5% NaOH, washed thoroughly with water, dried, and recrystallized from benzene giving 5.5 g (70%) of 7-bromoethoxy-5-hydroxy-4′-methoxyisoflavone (XI), mp 163–165°. A solution of 3.91 g of XI in 35 ml of diethylamine and 35 ml of dimethylformamide was kept for 4 days at room temperature, then evaporated to dryness under reduced pressure. The residue, triturated with water, dried, and recrystallized from petroleum ether gave V, mp 78–80°, identical with the product obtained by method A.

Ethyl 4-Methoxy-7-isoflavonoxyacetate (XII). Method C.—To a stirred suspension of 0.02 mole of formononetin sodium salt in xylene, prepared as in method A, was added 10.6 g (0.06 mole) of ethyl bromoacetate. The mixture was refluxed for 5 hr, filtered, and evaporated under reduced pressure. The residue was triturated with petroleum ether. Recrystallization from ethyl acetate or benzene yielded 5.9 g (83%) of XII, mp 142–144°.

4′-Methoxy-7-isoflavonoxyacetic Acid (XIII).—A mixture of 2 g of XII, 30 ml of acetone, 4 ml of water, and 3.25 ml of 2 *N* NaOH was stirred for 2 hr. Water was added and acetone was evaporated under reduced pressure. On acidification of the clear solution with HCl, 1.45 g (79%) of XIII, mp 223–225°, was precipitated.

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Octamethylbiguanide Perchlorate

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Although large numbers of biguanides have been synthesized as potential antimalarial drugs[1] and as hypoglycemic agents,[2] the number of polysubstituted examples is limited. In this paper, we describe the synthesis of octamethylbiguanide perchlorate (1), the most highly substituted biguanide yet reported.

The reaction of tetramethylchloroformamidine chloride (2) and 1,1,3,3-tetramethylguanidine provided a hygroscopic chloride

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