Mutagenic Heterocyclic Nitrogen Compounds Related to Protein Pyrolysates. 1. Derivatives of Dipyrido[1,2-*a*:3′,2′-*d*]imidazole

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2-Amino-6-methyldipyrido[1,2-*a*:3′-2′-*d*]imidazole is one of the mutagenic principles of L-glutamic acid and casein pyrolysates. We prepared several isomeric amines of this compound, as well as their homologues methylated in different positions on the heterocyclic ring, either by nitration of the ring followed by reduction or from appropriate 4-azidodipyrido[1,2-*a*:3′,2′-*d*]imidazoles. The latter compounds, the heterocyclic ring itself and various functional derivatives (hydroxylated, carboxylated, hydrazinic) were synthesized from 2-aminoimidazo[1,2-*a*]pyridines *via* various reactions which are described herein.

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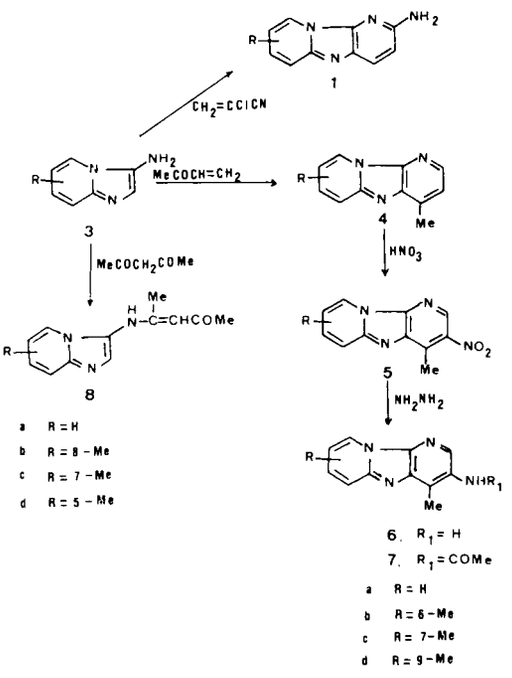
L-Glutamic acid is a constituent of various proteins. Two highly mutagenic compounds were recently identified (1) in pyrolysates of this amino acid: 2-amino-6-methylpyrido[1,2-*a*:3′,2′-*d*]imidazole (**1**) and 2-amino-pyridyl[1,2-*a*:3′,2′-*d*]imidazole (**2**), respectively, designated as Glu-P1 and Glu-P2. These amines may be considered as isosteric nitrogen-containing compounds of 2-amino-fluorene, a well known carcinogen. They also form in the course of combustion of such dietary proteins as casein (2).

These compounds also cause the transformation of embryonic hamster cells *in vitro* (3). It was recently shown that in the presence of microsomal proteins, Glu-P1 modified DNA by covalent binding to C-8 of guanine (4) and also by intercalation between base pairs (5).

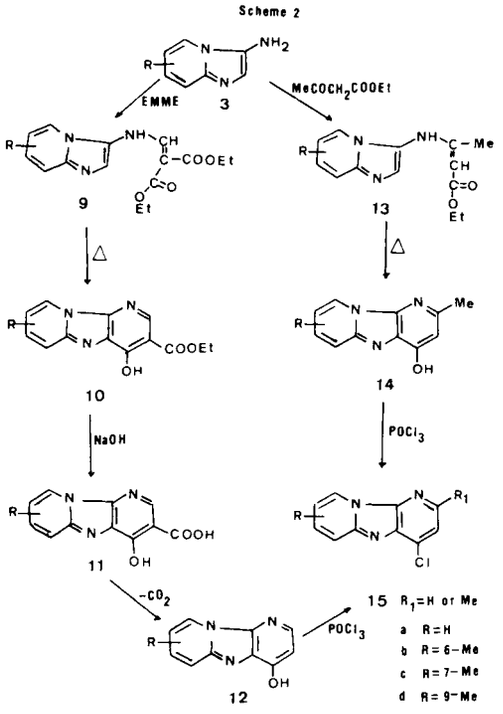
This class of compounds is structurally and biochemically interesting, especially for studies of chemical carcinogenesis. Our interest in the interaction between carcinogenic amines and nucleic acids (19) led us to synthesize various derivatives of dipyrido[1,2-*a*:3′,2′-*d*]imidazole and study their properties. In addition to Glu-P1 and Glu-P2, it is possible that certain of these compounds are formed during the pyrolysis of L-glutamic acid-containing proteins.

Synthetic protocols used are summarized in Schemes 1-3.

Scheme 1



Scheme 2



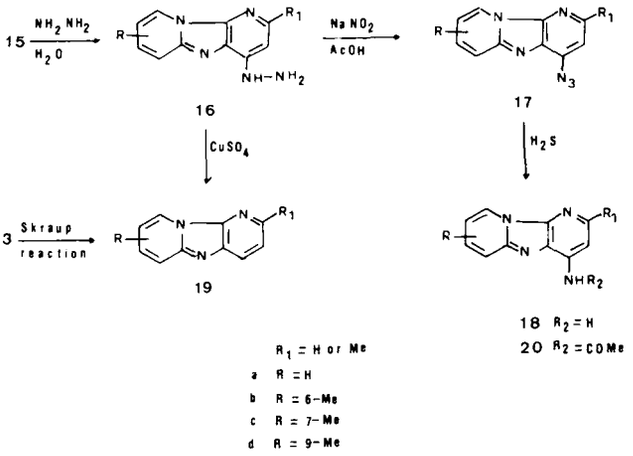
The precursors of these reactions, 3-aminoimidazo-[1,2-*a*]pyridine (**3**) and its methylated derivatives (R = 5-Me, 7-Me or 8-Me) were prepared by reacting the appropriate 2-aminopyridines with formaldehyde sodium bisulfite addition compound in the presence of sodium cyanide. These compounds exist in the form of cyclic B tautomers, rather than in the open A form, as postulated by Bristow, *et.al.*, (6), as shown by the nmr and ir spectrometric data

Table 1

Characteristics of Dipyrido[1,2-*a*:3',2'-*d*]imidazoles Derivatives and Intermediates

Compound	Formula	Mp °C	Yield %	% Calculated C	H	N	% Found C	H	N
1a	$C_{10}H_8N_4$	185	35						
4a	$C_{11}H_9N_3$	150	32	72.1	4.9	22.9	72.1	5.0	22.8
4b	$C_{12}H_{11}N_3$	90	25	72.1	4.9	22.9	72.2	4.8	22.9
4c	$C_{12}H_{11}N_3$	151	25	72.1	4.9	22.9	72.0	4.9	22.7
4d	$C_{12}H_{11}N_3$	149	35	72.1	4.9	22.9	72.3	4.8	22.7
5a	$C_{11}H_8N_4O_2$	250	68	57.9	3.5	24.5	58.0	3.5	24.6
5c	$C_{12}H_{10}N_4O_2$	240	65	59.5	4.2	23.1	59.6	4.1	23.2
6a	$C_{11}H_{10}N_4$	233	54	66.6	5.1	28.3	66.7	5.2	28.1
6c	$C_{12}H_{12}N_4$	199	60	67.9	5.7	26.4	67.9	5.6	26.5
7a	$C_{13}H_{12}N_4O$	245	91	65.0	5.0	23.2	64.9	5.1	23.3
7c	$C_{14}H_{14}N_4O$	241	90	66.1	5.5	22.3	66.3	5.6	22.2
8a	$C_{12}H_{13}N_3O$	100	70	66.9	6.1	19.5	67.1	6.1	19.6
8b	$C_{13}H_{15}N_3O$	96	55	68.1	6.6	18.3	68.2	6.6	18.2
8c	$C_{13}H_{15}NO_3$	116	65	68.1	6.6	18.3	68.0	6.5	18.3
8d	$C_{13}H_{15}NO_3$	112	45	68.1	6.6	18.3	68.2	6.4	18.2
9a	$C_{15}H_{17}N_3O_4$	142	90	59.4	5.6	13.8	59.6	5.7	13.8
9b	$C_{16}H_{19}N_3O_4$	132	70	60.6	6.0	13.2	60.5	6.0	13.5
9c	$C_{16}H_{19}N_3O_4$	148	89	60.6	6.0	13.2	60.6	6.0	13.2
9d	$C_{16}H_{19}N_3O_4$	149	58	60.6	6.0	13.2	60.4	6.1	13.1
10a	$C_{13}H_{11}N_3O_3$	242	59	60.7	4.3	16.3	60.5	4.4	16.5
10b	$C_{14}H_{13}N_3O_3$	172	46	62.0	4.8	15.5	61.9	4.9	15.4
10c	$C_{14}H_{13}N_3O_3$	215	76	62.0	4.8	15.5	61.9	5.0	15.6
11a	$C_{11}H_7N_3O_3$	>350	89	57.6	3.1	18.3	57.7	3.0	18.1
11c	$C_{12}H_9N_3O_3$	>350	90	59.3	3.7	17.3	59.1	3.8	17.1
12a	$C_{12}H_7N_3O$	283	95	64.8	3.8	22.7	64.7	3.9	22.8
12b	$C_{12}H_9N_3O$	285	97	63.3	4.5	21.1	63.1	4.7	21.0
12c	$C_{12}H_9N_3O$	287	93	63.3	4.5	21.1	63.2	4.6	21.0
14a	$C_{11}H_9N_3O$	299	95	63.3	4.5	21.1	63.1	4.7	21.0
14b	$C_{12}H_{11}N_3O$	245	80	67.7	5.2	19.7	67.6	5.1	19.8
14c	$C_{12}H_{11}N_3O$	303	90	67.7	5.2	19.7	67.7	5.2	19.7
14d	$C_{12}H_{11}N_3O$	306	85	67.7	5.2	19.7	67.7	5.3	19.6
15a (R_1 = H)	$C_{10}H_6ClN_3$	177	87	59.0	3.0	20.6	58.8	2.9	20.5
15a (R_1 = Me)	$C_{11}H_8ClN_3$	141	90	60.7	3.7	19.3	60.5	3.8	19.3
15b (R_1 = H)	$C_{11}H_8ClN_3$	169	75	60.7	3.7	19.3	60.6	3.7	19.1
15c (R_1 = H)	$C_{11}H_8ClN_3$	186	88	60.7	3.7	17.3	60.6	3.7	17.4
15b (R_1 = Me)	$C_{12}H_{10}ClN_3$	159	80	62.2	4.3	18.1	62.0	4.5	18.0
15c (R_1 = Me)	$C_{12}H_{10}ClN_3$	207	90	62.2	4.3	18.1	62.1	4.3	18.2
16a (R_1 = H)	$C_{10}H_9N_5$	212	76	60.3	4.5	36.1	60.1	4.7	36.1
16a (R_1 = Me)	$C_{11}H_{11}N_5$	235	82	61.7	5.6	32.7	61.5	5.7	32.6
16c (R_1 = H)	$C_{11}H_{11}N_5$	249	75	61.7	5.6	32.7	61.7	5.6	32.5
16c (R_1 = Me)	$C_{12}H_{13}N_5$	239	80	63.4	5.7	30.8	63.5	5.7	30.7
17a (R_1 = H)	$C_{10}H_6N_6$	156	90	57.7	2.9	40.0	57.5	2.8	40.1
17a (R_1 = Me)	$C_{11}H_8N_6$	161	98	58.9	3.6	37.5	58.8	3.6	37.6
17c (R_1 = H)	$C_{11}H_8N_6$	164	89	58.9	3.6	37.5	58.9	3.6	37.7
17c (R_1 = Me)	$C_{12}H_{10}N_6$	162	90	60.5	4.2	35.3	60.4	4.2	35.3
18a (R_1 = H)	$C_{10}H_8N_4$	206	75	65.2	4.4	30.4	65.2	4.3	30.5
18a (R_1 = Me)	$C_{11}H_{10}N_4$	207	77	66.7	5.1	28.2	66.6	5.0	28.4
18c (R_1 = H)	$C_{11}H_{10}N_4$	203	82	66.7	5.1	28.2	66.7	5.2	28.1
18c (R_1 = Me)	$C_{12}H_{12}N_4$	213	84	67.9	5.7	26.4	67.8	5.5	26.5
19a (R_1 = H)	$C_{10}H_7N_3$	130		71.0	4.2	28.8	71.1	4.3	28.9
19a (R_1 = Me)	$C_{11}H_9N_3$	144	70	72.1	4.9	22.9	72.1	4.8	23.0
19b (R_1 = H)	$C_{11}H_9N_3$	117		72.1	4.9	22.9	72.0	5.0	23.1
19c (R_1 = H)	$C_{11}H_9N_3$	148	84	72.1	4.9	22.9	72.2	4.8	22.9
19c (R_1 = Me)	$C_{12}H_{11}N_3$	143		73.1	5.6	21.3	73.1	5.7	21.2

Scheme 3



(especially by the absence of bands around 2250 cm^{-1} in spectrometry, characteristic of the -CN group).

These amines were condensed with 2-chloroacrylonitrile, as reported by Takeda, *et.al.*, (7), with some modifications, to yield the corresponding 2-aminopyrido[1,2-*a*:3',2'-*d*]imidazoles (**1**).

3A 3B

When the same compounds **3** were condensed with methyl vinyl ketone in the presence of ferric chloride and zinc chloride, according to the method of Campbell and Schaffner (8), we obtained 4-methyldipyrido[1,2-*a*:3',2'-*d*]-imidazoles in moderate yields. Treatment of the latter compounds with hot concentrated (d = 1.49) nitric acid gave a mono nitro compound whose structure was attributed to **5**, based on nmr spectra and in reference to the nitration of pyridine (9) and to that of 3-methyl-3*H*-imidazo[4,5-*b*]pyridine (10). These nitrations occur *meta* to the pyridine nitrogen, the most accessible position for electrophilic substitution reactions. The heterocycle seems to be only slightly reactive, perhaps as a result of the polar effect due to the heteroatom, similar to the case of pyridine derivatives (11). Thus, we were not able to obtain nitration under milder conditions (use of less concentrated nitric acid, even in the presence of sulfuric acid).

The reduction of the nitro compounds by hydrazine hydrate in the presence of Raney nickel enabled us to easily obtain the corresponding amines **6**.

Precursor **3** reacts easily with acetylacetone to yield the compounds **8**, but we were unfortunately unable to perform the Beyer-Combes cyclodehydration (12) of this product. This failure may be attributed to the fragility of this type of molecule under the rather harsh conditions of this reaction. In the presence of concentrated sulfuric acid or of polyphosphoric acid (12c), compounds **8** are hydrolyzed, with regeneration of the amines **3**.

Reaction with ethyl acetoacetate in the presence of Drierite, on the other hand, yielded the crotonate **13**. When heated in Dowtherm A, this compound easily underwent the Conrad-Lipach cyclization reaction (13) to yield the hydroxylated compound **14**.

Similarly, reaction with diethylethoxymethylene malonate (EMME), followed by cyclization of the resulting crotonate **9** in Dowtherm, yielded the hydroxyester **10** in an equivalent yield. After saponification and decarboxylation, the latter yielded the hydroxylated compound **12** *via* the carboxylic acid **11**. The two types of hydroxylated compounds **12** and **14** could be easily transformed to the chlorinated derivatives **15** by the action of phosphorus oxychloride.

When the chlorinated compound **15** was allowed to react with hydrazine hydrate, the hydrazine **16** was obtained. In dilute acetic acid at 0° and in the presence of sodium nitrite, the hydrazine was oxidized to the azide **17**, which was then reduced to the corresponding amine **18** by hydrogen sulfide according to the method of Stanovik, *et.al.*, (14). We unsuccessfully attempted to obtain this amine by aminolysis of the chlorinated derivative **12**.

Amines **6** and **18** were easily transformed to their amides **7** and **20** by acetic anhydride, in order to study their mutagenic and carcinogenic properties.

Finally the heterocycle **19** could be obtained by two different reaction schemes. (a) Action of cupric sulfate pentahydrate on the hydrazine **16** in boiling acetic acid solution. In this case, the 2-methylated compound was obtained. (b) Skraup reaction (15) of glycerol on the amine **3** in the presence of concentrated sulfuric acid and iodine. This reaction yields a compound which does not contain a substituent on position 2 and occurs with a low yield, since sulfuric acid leads to a partial degradation of the amine **3** to 2-aminopyridine.

With the exception of the slightly soluble nitro compounds, the derivatives of dipyrido[1,2-*a*:3',2'-*d*]imidazole synthesized in the present study exhibit a strong steel blue fluorescence in the usual solvents (water, ethanol, ether, benzene, dichloromethane, chloroform, *etc.*). Their precursors **3** do not share this property. Strong absorption between 340 and 380 nm and towards 250-260 nm was observed.

The basic nature of the ring is shown by the well known modification of the uv spectrum in ethanolic hydrogen chloride, as shown by the example of 4-methyldipyrido-[1,2-*a*:3',2'-*d*]imidazole (Figure 1).

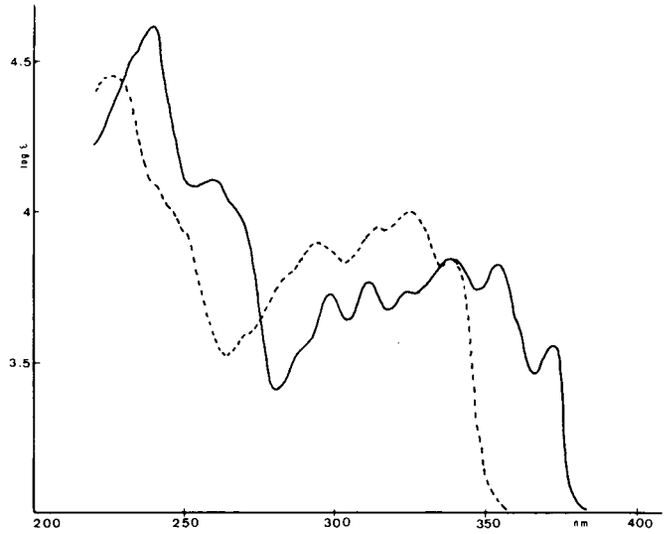


Figure 1. The uv absorption spectra of 4-methyldipyrido-[1,2-*a*:3′,2′-*d*]imidazole in ethanol (——) in ethanol + HCl $10^{-2}M$ (-------).

The attribution of nmr signals of the protons of the compounds studied is based on the comparison of the different spectra, on the examination of coupling constants and on known data on the spectra of imidazo[1,2-*a*]pyridine derivatives (16,17).

Tests for mutagenic properties of the above dipyridoimidazole derivatives were performed and some of them exhibited a strong mutagenic activity towards several strains of *S-typhimurium* in presence and absence of microsomes. The results will be reported in a separate communication.

Experiments to establish the identity of the metabolites responsible for mutagenicity and interactions of these with nucleic acids are currently in progress.

Acknowledgement.

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Table 2

NMR, IR and UV Spectral Data

Compound	δ Values and Proton Assignment	ν NH_2 max/cm^{-1}	λ max (log ε) nm
1a	4.47 (b.s., NH_2) 6.72 (d, H-3 J = 9 cps) 6.75 (m, H-8) 7.2-7.7 (m, H-6, H-7) 7.97 (d, H-4, J = 9 cps) 8.55 (d, H-9, J = 7 cps)	3340	210 (4.17), 255 (4.10), 301 (3.53), 382 (3.74)
4a	2.85 (s, Me) 6.88 (m, H-8) 7.38 (d, H-3, J = 4 cps) 7.55-7.8 (m, H-6, H-7) 8.41 (d, H-2, J = 4 cps) 8.83 (m, H-9)		
4b	2.70 (s, 6-Me) 2.85 (s, 4-Me) 6.70 (m, H-8) 7.2 (m, H-3 and H-7) 8.2 (d, H-2, J = 4 cps) 8.5 (m, H-9)		
4c	2.47 (s, 7-Me) 2.8 (s, 4-Me) 6.8 (d, H-8, J = 7 cps) 7.3 (d, H-3, J = cps) 7.45 (s, H-6) 8.25 (d, H-2, J = 5 cps) 8.65 (d, H-9, J = 7cps)		
5a	3.21 (s, Me) 6.94 (m, H-8) 7.53-7.62 (m, H-6, H-7) 8.21 (s, H-2) 8.76 (d, H-9, J = 8 cps)		
5e	2.5 (s, 7-Me) 2.83 (s, 4-Me) 6.84 (d, H-8) 7.4 (s, H-6) 8.23 (s, H-2) 8.65 (d, H-9)		
6a	2.65 (s, Me) 4.5 (b, s, NH_2) 6.5 (s, H-2) 6.75 (m, H-8) 7.20-7.7 (m, H-6 and H-7) 8.45 (d, H-9)	3400	212 (4.31), 255 (4.22), 302 (3.68), 380 (3.83)
15a (R_1 = H)	6.8 (m, H-8) 7.45 (d, H-3, J = 4 cps) 7.25-7.65 (m, H-6 and H-7) 8.15 (d, H-2, J = 4 cps) 8.55 (d, H-9, J = 7 cps)		
15a (R_1 = Me)	2.7 (s, Me) 6.83 (m, H-8) 7.31 (s, H-3) 7.2-7.7 (m, H-6, H-7) 8.61 (d, H-9)		
15c (R_1 = H)	2.45 (s, Me) 6.72 (d, H-8, J = 7 cps) 7.35 (d, H-3, J = 4 cps) 7.4 (s, H-6) 8.23 (d, H-2, J = 4 cps) 8.52 (d, H-9, J = 7 cps)		
15c (R_1 = Me)	2.41 (s, 7-Me) 2.65 (s, 2-Me) 6.65 (d, H-8, J = 7 cps) 7.26 (s, H-3) 7.35 (s, H-6) 8.47 (d, H-9), J = 7 cps)		
18a (R_1 = H)	5.18 (b, s, NH_2) 6.63 (d, H-3, J = 5 cps) 6.82 (m, H-8) 7.25-7.7 (m, H-6, H-7) 8.15 (d, H-2, J = 5 cps) 8.72 (d, H-9, J = 7 cps)	3400-3300	254 (4.5), 303 (3.64), 344 (3.88), 360 (3.84)
18a (R_1 = Me)	2.55 (s, 2-Me) 5.1 (b.s., NH_2) 6.5 (s, H-3) 6.75 (m, H-8) 7.2-7.65 (m, H-6, H-7) 8.7 (d, H-9)	3400-3280	257 (4.46), 304 (3.62), 346 (3.77), 360 (3.73)
18c (R_1 = H)	2.45 (s, 7-Me) 5.03 (b.s., NH_2) 6.6 (d, H-3, J = 5 cps) 6.7 (d, H-8, J = 7 cps) 7.32 (s, H-6) 8.07 (d, H-2, J = 5 cps) 8.52 (d, H-9, J = 7 cps)	3330-3200	255 (4.33), 302 (3.57), 340 (3.77), 355 (3.64)
18c (R_1 = Me)	2.4 (s, 7-Me) 2.6 (s, 2-Me) 5.03 (b.s., NH_2) 6.49 (s, H-3) 6.62 (d, H-8, J = 7 cps) 7.35 (s, H-6) 8.57 (d, H-9, J = 7 cps)	3320-3200	258 (4.35), 276 (3.74), 303 (3.60), 340 (3.61), 355 (3.55)
19a (R_1 = H)	6.93 (m, H-8) 7.3-7.7 (m, H-3, H-6, H-7) 8.23 (1, H-4) 8.5 (q, H-2) 8.82 (q, H-9)		
19a (R_1 = Me)	2.78 (s, 2-Me) 6.82 (m, H-8) 7.33 (d, H-3, J = 10 cps) 7.36 (m, H-7) 7.56 (m, H-6) 8.1 (d = H-4, J = 10 cps) 8.78 (q, H-9)		
19b (R_1 = H)	2.68 (s, 6-Me) 6.8 (m, H-4) 7.25-7.45 (m, H-3,H-7) 8.22 (q, H-4) 8.45 (q, H-2) 8.65 (q, H-9)		
19c (R_1 = H)	2.41 (s, 7-Me) 6.7 (d, H-8, J = 7 cps) 7.31 (s, H-6) 7.41 (m, H-3) 8.21 (q, H-4) 8.42 (q, H-2) 8.68 (d, H-9, J = 7 cps)		
19c (R_1 = Me)	2.48 (s, 7-Me) 2.69 (s, 2-Me) 6.61 (d, H-8, J = 7 cps) 7.29 (d, H-3, J = 5 cps) 7.29 (s, H-6) 7.98 (d, H-4, J = 5 cps) 8.55 (d, H-9, J = 7 cps)		

EXPERIMENTAL

The nmr spectra were recorded with a varian A 60 spectrometer in deuteriochloroform solutions with TMS as the internal standard: ir absorption spectra (potassium bromide) were recorded on a Perkin-Elmer 457 apparatus and uv absorption spectra was measured with a DB Beckman spectrophotometer.

Characteristics of the compounds synthetized are reported in Tables 1 and 2.

Amines **3** were previously reported (6,7,18). Glu-P1 and Glu-P2 **1a** and **1b** were previously described (7).

4-Methyldipyrido[1,2-*a*:3',2'-*d*]imidazole (**4a**).

A mixture of 8.3 g of 3-aminoimidazo[1,2-*a*]pyridine (0.0625 mole), 27 g of ferric chloride hexahydrate (0.1 mole) and 1 g of anhydrous zinc chloride in 50 ml of ethanol was heated to an inside temperature 65-70°; 3.5 g of methyl vinyl ketone (0.05 mole) was added dropwise during about 10 minutes. The mixture was then refluxed for 12 hours. After evaporation of the solvent, the residue was treated with 25% sodium hydroxide. The dark solid obtained upon evaporation *in vacuo* was extracted with hexane in a Soxhlet extractor.

The compound **4a** thus obtained was recrystallized in hexane giving colorless prisms.

The methyl homologues **4b**, **4e** and **4d** were prepared in the same way from the appropriate amines **3**.

4-Methyl-3-nitrodipyrido[1,2-*a*:3',2'-*d*]imidazole (**5a**).

Five g of **4a** (0.027 mole) was added in small portions at 20 ml of nitric acid (d, 1.49) under magnetic stirring and the mixture was then heated for 2 hours. After cooling the reaction mixture was poured into ice and the precipitate was filtered, washed with cold water and the nitro compound recrystallized from ethanol to give yellow needles. The methyl homologue **5e** was obtained in the same way.

Amines **6a** and **6e**.

To a solution of 0.013 mole of the nitro compounds **5** and 98% hydrazine hydrate (3 ml) in ethanol (120 ml), Raney nickel (0.75 g) was added in small portions, and the mixture was refluxed for 1.5 hours. The nickel was then filtered off, the solvent evaporated and the residue recrystallized from benzene or ethanol-benzene.

3- And 4-Acetamidodipyrido[1,2-*a*:3',2'-*d*]imidazoles (**7**) and (**20**).

These compounds were obtained by refluxing for 50 minutes, a solution of 0.5 g of amines **6** and **18** in 5 ml of acetic anhydride, respectively. After cooling the mixture was treated with sodium hydroxide to *p*H 7 and extracted with methylene chloride. Upon removing of the solvent, the residue was recrystallized from benzene as colorless needles.

Preparation of the Compounds **8**.

An equimolecular mixture of the amine **3** and acetylacetone was heated gently *ca.* 110° for 3 hours. The cooled product was then recrystallized from cyclohexane.

Crotonates **9**.

A mixture of 15 g of the amine **3** (0.11 mole) and 30.4 g of ethoxymethylene malonic ester (0.14 mole) was refluxed for 30 minutes. Recrystallization of the solid obtained from ethanol following cooling yielded colorless prisms.

3-Carbethoxy-4-hydroxydipyrido[1,2-*a*:3',2'-*d*]imidazoles (**10**).

Ten g of the crotonate **9** in 100 ml of Dowthem A was heated under reflux with magnetic stirring for 40 minutes and allowed to cool, followed by dilution with ethyl ether. The insoluble material was removed by filtration, washed with ethyl ether and recrystallized from ethanol-benzene, yielding the hydroxy-ester as colorless prisms.

4-Hydroxydipyrido[1,2-*a*:3',2'-*d*]imidazoles (**12**).

The ester **10** (15 g) was hydrolysed by boiling with 30% sodium hydroxide (100 ml) for 12 hours. After cooling, the reaction product was filtered, washed with water and recrystallized from acetic acid. The acid thus obtained was decarboxylated by sublimation *in vacuo* at *ca.* 250°. The sublimate was recrystallized from ethanol-benzene as yellow needles.

4-Hydroxy-2-methyldipyrido[1,2-*a*:3',2'-*d*]imidazoles (**14**).

These compounds were prepared by reflux for 24 hours of a mixture of the amines **3** (0.04 mole), of ethyl acetoacetate (0.04 mole) and 35 g of Drierite in 50 ml of absolute ethanol. After filtering and removing the ethanol *in vacuo*, the residue was recrystallised from hexane as colorless needles. Cyclisation of 100 g of the crotonate, thus obtained, of refluxing Dowtherm A for 30 minutes gave, after washing with ethyl ether and recrystallization from ethanol-benzene 80 g (89%) of the cyclic compound.

4-Chlorodipyrido[1,2-*a*:3',2'-*d*]imidazoles (**15**).

A mixture of 50 g of the hydroxylated compound **12** and **14** and 100 ml of phosphorus oxychloride was refluxed for 6 hours and excess phosphorus oxychloride removed by vacuum distillation. After cooling the crude product was treated with ammonia to *p*H 7. The solid obtained was filtered, washed thoroughly with water, and recrystallized in colorless needles from benzene.

4-Hydrazinodipyrido[1,2-*a*:3',2'-*d*]imidazole (**16**).

A mixture of 5 g of the chloro derivative **15** and 25 ml of 98% hydrazine hydrate was refluxed for 40 minutes. After cooling, water (90 ml) was added and the precipitate obtained was washed with hot benzene, dried and recrystallized from water as colorless microprisms.

4-Azidodipyrido[1,2-*a*:3',2'-*d*]imidazoles (**17**).

To a cold solution at 0° containing 4 g of hydrazine **16** dissolved in 5 ml of acetic acid and 50 ml of water was added dropwise a solution of 2.3 g of sodium nitrite in 5 ml of water. The mixture was stirred at 0° for 5 minutes, filtered and the residue recrystallized from ethanol; ir: strong band at 2150-2155 cm^{-1} (N^3 group).

4-Aminodipyrido[1,2-*a*:3',2'-*d*]imidazoles (**18**).

Into a hot solution of 3.7 g of the azide **18** in 50 ml of methanol, hydrogen sulfide was bubbled for 40 minutes. After cooling, the precipitate was filtered, the filtrate evaporated *in vacuo* and the residue recrystallized from benzene as pale yellow microprisms.

Dipyrido[1,2-*a*:3',2'-*d*]imidazoles (**19**).

These heterocycles were obtained by the two procedures which are described below.

Method a.

To a solution of 15 g of hydrazino compound **16** in 37 ml of acetic acid and 154 ml of water was added a solution of 37 g of cupric sulfate pentahydrate in 370 ml of water. The mixture was refluxed for 1 hour, then treated with sodium hydroxide and the precipitate was filtered. Extraction of the latter with chloroform in a Soxhlet extractor gave upon evaporation *in vacuo* **19**, which was recrystallized from hexane as colorless microprisms.

Method b.

A mixture of 13.3 g of the amine **3**, 4.9 g of glycerol, 27 g of concentrated sulfuric acid and 0.49 g of iodine was heated at 170° for 3 hours. After cooling, the reaction mixture was poured on ice and made basic with ammonia and extracted into dichloromethane. The solid obtained after removal of the solvent was purified by distillation *in vacuo* and recrystallization from hexane.

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