Studies on Biologically Active Pteridines. II.[1] Synthesis of Some 2-Pyrimidinyl- and 2-Pteridinyl-amino Acids

Takashi SUGIMOTO,* Keiko SHIBATA, Sadao MATSUURA, and Toshiharu NAGATSU**

Department of Chemistry, College of General Education, Nagoya University, Chikusa-ku, Nagoya 464
***Laboratory of Cell Physiology, Department of Life Chemistry, Graduate School at Nagatsuta, Tokyo Institute of Technology, Midori-ku, Yokohama 227*

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Synthesis of some pyrimidines and pteridines with an amino acid residue at the 2-position was investigated. 4-Amino-6-hydroxy-2-methylthio-5-nitrosopyrimidine, on heating with amino acids in water, underwent aminolysis at the 2-position to give the corresponding *N*-(4-amino-6-hydroxy-5-nitroso-2-pyrimidinyl)amino acids. These (5-nitroso-2-pyrimidinyl)amino acids, after reduction of the nitroso group to an amino group, condensed with biacetyl or a pentose phenylhydrazone to give *N*-(4-hydroxy-6,7-dimethyl-2-pteridinyl)amino acids and *N*-(4-hydroxy-6-polyhydroxypropyl-2-pteridinyl)amino acids, respectively.

Various 2-amino-4-hydroxypteridines and their 5,6,7,8-tetrahydro derivatives having an alkyl or polyhydroxyalkyl substituent at the 6-position possess distinctive biological activities. For example, biopterin [2-amino-4-hydroxy-6-(L-*erythro*-1,2-dihydroxypropyl)-pteridine] is a growth factor for *Crithidia fasciculate*[2,3] and the 5,6,7,8-tetrahydro derivative is a potent coenzyme for phenylalanine,[4] tyrosine,[5,6] and tryptophan hydroxylases.[7]

The relation between the structure of 6-substituted 5,6,7,8-tetrahydro-2-amino-4-hydroxypteridines and the coenzyme activities for tyrosine hydroxylase has been studied extensively by Nagatsu and his coworkers.[6,8,9] However, the pteridines used in their study vary only in the substituents attached to the pyrazine ring of the compounds except a few cases.[8] It is of interest to investigate how the biological activities of such pteridines will be changed by modification in the pyrimidine rather than the pyrazine moiety. This paper describes the synthesis of 6- or 6,7-disubstituted 2-amino-4-hydroxypteridines possessing a carboxyalkyl group attached to the 2-amino group. The choice to introduce a carboxyalkyl group rather than a simple alkyl group stemmed from an anticipation that such polar group would cause a stronger interaction of the pteridine with the biological systems. In addition, it is expected that further modification of the compounds at the carboxyl group, for example the formation of an amido linkage to polyamines or proteins, would widen the utility of these compounds.

The amino acid residue of the pteridines (**3**, **4**, and **5**) was introduced prior to the formation of the pteridine ring by nucleophilic displacement of the methylthio group of 4-amino-6-hydroxy-2-methylthio-5-nitrosopyrimidine (**1**) by an appropriate amino acid in analogy to the aminolysis of **1** by simple aliphatic amines.[10] When the methylthiopyrimidine was heated with an excess amount of 6-aminohexanoic acid in an aqueous solution, the solution changed the color from blue to dark red and evolved methanethiol. The displacement became complete after refluxing for 1 h and the yield of 6-[(4-amino-6-hydroxy-5-nitroso-2-pyrimidinyl)amino]hexanoic acid (**2a**) was about 70%. The reaction of **1** with 4-aminobutanoic acid and β-alanine took place in a similar manner to give the 4-(2-pyrimidinylamino)butanoic and 3-(2-pyrimidinylamino)propanoic acids (**2b** and **2c**), respectively. However, the displacement reaction of **1** with several α-amino acids proceeded much easier when the sodium salts of the acids in place of the free acids were employed. Thus, heating of **1** with L-alanine in the presence of an equivalent amount of sodium hydroxide yielded *N*-(4-amino-6-hydroxy-5-nitroso-2-pyrimidinyl)-L-alanine (**2d**) in 53% yield; in the absence of sodium hydroxide, the yield was 22%. Under similar conditions, **1** reacted with D-alanine and glycine to give the *N*-(2-pyrimidinyl)-D-alanine (**2e**) and the *N*-(2-pyrimidinyl)glycine (**2f**), respectively.

These *N*-(2-pyrimidinyl)amino acids showed two pK_a values at about 2.4 and 7.9, which were attributed to the ionization of the pyrimidine ring to form a cation (pK_a 2.4) and to form an anion (pK_a 7.9); pK_a values due to the ionization of the carboxyl group could not be determined on a spectroscopic measurement. Thus, all the *N*-(2-pyrimidinyl)amino acids, despite of the different carbon chain between the carboxyl and amino groups, exhibited UV spectra (Table 1) very similar to those of known 4-amino-6-hydroxy-2-methylamino-5-nitrosopyrimidine.[11] These physical data, together with elemental analyses, confirmed the structure of the aminolysis products.

The nitrosopyrimidines (**2a**—**f**) were converted to the corresponding 2-amino-4-hydroxy-6,7-dimethylpteridines (**3a**—**f**) having a carboxyalkyl group attached to the 2-amino group in a conventional way: by reduction of the 5-nitroso group to an amino group by catalytic hydrogenation, followed by condensation with biacetyl. By using glyoxal instead of biacetyl in the above course, **2f** gave *N*-(4-hydroxy-2-pteridinyl)glycine (**4**).

Several pteridines analogous to biopterin or neopterin having a di- or trihydroxypropyl substituent at the 6-position were also synthesized starting from the 5-nitrosopyrimidines (**2a** and **2f**). Condensation of the 5-amino analogue of **2a** or **2f** with L-arabinose phenylhydrazone at pH 2—3, followed by air oxidation at pH 7—8 gave 6-[[4-hydroxy-6-(L-*erythro*-1,2,3-trihydroxypropyl)-2-pteridinyl]amino]hexanoic acid (**5a**) and the lower homologue (**5b**), respectively. Condensation of the 5-amino analogue of **2a** with 5-deoxy-L-arabinose phenylhydrazone in a way similar to that for biopterin[1] afforded 6-[[4-hydroxy-6-(L-*erythro*-1,2-dihydroxypropyl)-2-pteridinyl]amino]hexanoic acid (**5c**).

TABLE 1. THE pK_a VALUES AND UV SPECTRA OF *N*-(2-PYRIMIDINYL)AMINO ACIDS AND *N*-(2-PTERIDINYL)AMINO ACIDS

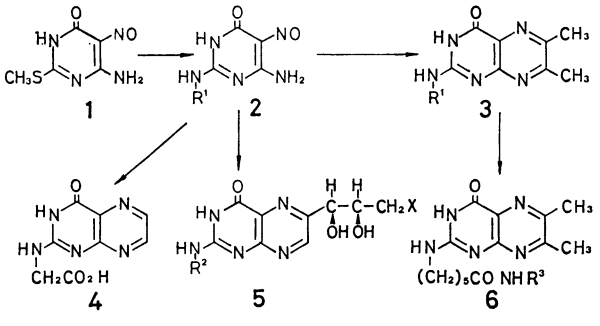
Compound	pK_a	pH of buffer[a]	λ_{max} (log ε)[b]	Ionic species[c]
2a	2.49±0.01	0.25	212(3.90), 268(4.06), *310(3.96)*	+
	8.10±0.01	5.5	216(3.77), 230(3.89), 258(3.66), 326(4.34)	○
		10.5	214(3.82), 328(4.36)	−
2b	2.46±0.01	0.25	212(3.92), 270(4.10), *310(3.81)*	+
	7.87±0.01	5.0	215(3.82), 230(3.93), 257(3.70), 326(4.36)	○
		10.0	213(3.86), 327(4.39)	−
2c	2.41±0.03	0.25	211(3.98), 269(4.16), *310(4.03)*	+
	7.91±0.02	5.0	215(3.85), 230(3.98), 257(3.75), 326(4.40)	○
		10.0	213(3.92), 327(4.44)	−
2d	2.31±0.01	0.25	209(3.96), 271(4.13), *310(3.99)*	+
	7.86±0.01	5.0	214(3.83), 230(3.93), 261(3.68), 326(4.37)	○
		10.0	213(3.89), 328(4.40)	−
2e	2.30±0.01	0.25	209(3.96), 271(4.13), *310(4.00)*	+
	7.86±0.01	5.0	215(3.82), 230(3.93), 261(3.68), 327(4.37)	○
		10.0	214(3.89), 328(4.38)	−
2f	2.31±0.01	0.25	209(3.99), 270(4.17), *310(4.03)*	+
	7.98±0.01	5.0	214(3.85), 230(3.98), 260(3.74), 326(4.41)	○
		10.0	211(3.89), 327(4.43)	−
3a	−2.36±0.03	−4.0	220(4.18), 254(4.11), 340(3.94)	2+
	2.60±0.01	0.25	219(4.29), 253(4.06), 324(3.96), 400(3.10)	+
	8.59±0.02	6.0	213(4.23), 279(4.24), 350(3.85)	○
		10.5	260(4.31), 366(3.91)	−
3b	−2.45±0.02	−4.0	219(4.22), 253(4.14), 339(3.98)	2+
	2.40±0.02	0.25	219(4.26), 253(4.05), 293(3.73), 323(3.93), 397(3.26)	+
	8.92±0.05	6.0	223(4.24), 279(4.25), 350(3.86)	○
		11.0	259(4.34), 365(3.94)	−
3c	−2.55±0.02	−4.0	220(4.18), 253(4.10), 338(3.95), 398(3.76)	2+
	2.22±0.02	−0.25	218(4.26), 253(4.05), 290(3.77), 323(3.91), 395(3.42)	+
	8.89±0.02	5.5	224(4.21), 278(4.24), 349(3.86)	○
		11.0	259(4.34), 364(3.93)	−
3d	−2.95±0.05	−4.0	222(4.13), 253(4.11), 286(3.76), 336(3.88), 393(3.53)	2+
	1.46±0.05	−0.75	218(4.09), 261(4.07), 285(4.04), 324(3.66), 389(3.88)	+
	3.2±0.1	2.5	220(4.21), 276(4.18), 344(3.84)	+[d]
	8.97±0.02	6.0	224(4.20), 279(4.25), 350(3.87)	○
		11.0	259(4.34), 364(3.93)	−
3e	−3.02±0.05	−4.0	221(4.11), 253(4.08), 286(3.72), 336(3.85), 392(3.46)	2+
	1.57±0.05	−0.75	218(4.06), 261(4.03), 285(4.00), 324(3.61), 390(3.84)	+
	3.6±0.1	2.5	221(4.16), 276(4.15), 344(3.80)	+[d]
	9.02±0.05	6.0	224(4.18), 278(4.23), 350(3.84)	○
		11.0	259(4.32), 364(3.91)	−
3f	−2.91±0.03	−4.0	221(4.12), 252(4.08), 287(3.68), 336(3.87), 391(3.41)	2+
	1.30±0.01	−0.75	219(4.11), 255(4.05), 285(3.98), 323(3.72), 387(3.82)	+
	3.36±0.01	2.5	220(4.20), 275(4.15), 343(3.83)	+[d]
	8.45±0.01	6.0	222(4.21), 278(4.23), 350(3.86)	○
		10.5	259(4.25), 360(3.89)	−
4	−3.2±0.1	−4.0	209(4.06), 235(4.02), *280(3.70)*, 318(3.72), 390(3.43)	2+
	0.88±0.02	−1.0	209(4.13), 235(4.08), *280(3.73)*, 315(3.78), 384(3.44)	+
	3.26+0.02	2.0	214(4.09), 234(4.06), 273(4.12), 342(3.76)	+[d]
	7.90±0.01	5.5	221(4.07), 275(4.19), 347(3.78)	○
		10.0	261(4.34), 365(3.84)	−
5a	2.10±0.04	0.25	213(4.22), 242(4.26), 324(3.94)	+
	7.96±0.01	5.0	221(4.15), *241(4.08)*, 281(4.31), 353(3.81)	○
		10.0	216(4.07), 265(4.41), 372(3.90)	−
5b	−1.6±0.1	−4.0	210(4.11), 251(4.09), 285(3.77), 323(3.80), 394(3.46)	2+
	1.01±0.03	−0.5	211(4.14), 239(4.12), 282(4.19), 322(3.74), 389(3.35)	+
	3.10±0.02	2.0	213(4.11), 239(4.08), 277(4.21), 347(3.79)	+[d]
	7.81±0.01	5.0	225(4.05), 240(4.04), 280(4.29), 352(3.79)	○
		10.0	218(4.00), 266(4.37), 369(3.87)	−

Table 1. (Continued)

Compound	pK_a	pH of buffer[a]	λ_{max} (log ε)[b]	Ionic species[c]
5c	2.13±0.01	0.25	212(4.03), 242(4.23), 325(3.90)	+
	7.95±0.02	5.0	221(4.12), *241(4.06)*, 281(4.29), 353(3.80)	○
		10.5	218(4.05), 265(4.41), 372(3.89)	−
6a	−2.45±0.02	−4.0	220(4.21), 254(4.13), 340(3.97)	2+
	2.59±0.02	0.25	219(4.32), 252(4.09), 324(3.98), 400(3.13)	+
	8.46±0.02	6.0	223(4.26), 279(4.26), 350(3.87)	○
		10.5	260(4.34), 365(3.94)	−
6b	−2.42±0.06	−4.0	220(4.20), 253(3.13), 339(3.97)	2+
	2.51±0.02	0.25	219(4.31), 252(4.09), 324(3.98), 396(3.09)	+
	8.50±0.01	5.5	225(4.22), 279(4.25), 350(3.86)	○
		10.5	260(4.33), 366(3.93)	−

a) Negative figures are H_0 values. b) Wavelength in nm measured in aqueous buffer of given pH; shoulders or inflexions in italics. c) Ionic species shown by 2+ (dication), + (monocation), ○ (neutral molecule), and − (monoanion) are regarded only to the pyrimidine or pteridine chromophore unless otherwise indicated. d) Zwitter ions.

The structures of these *N*-(2-pteridinyl)amino acids (**3**, **4**, and **5**) were confirmed by the elemental analyses and the close similarity of their UV spectra (Table 1) to those of 4-hydroxy-6,7-dimethyl-2-methylaminopteridine[12] or biopterin.[13] The pteridines (**3d**—**f**, **4**, and **5b**) derived from α-amino acids exhibited four pK_a values at about 8.8, 3.3, 1.0, and −3.0 which were measured by a spectroscopic method.[14] The values at 8.8, 3.3, and −3.0 are attributed to the ionization of the pteridine ring to form anionic species by losing a proton from the ring (pK_a 8.8) and to form cationic species by single or double protonation (pK_a 3.3 and −3.0, respectively). The pK_a value at 1.0 could be attributed to the ionization of the carboxyl group, since no pK_a was detected by a spectroscopic measurement at this range with **3a**—**c** where the carboxyl group was separated far from the pteridine chromophore by a long carbon chain of two or more carbon atoms. The acidity of the monocations (protonated at the pteridine ring) of **3d**—**f**, **4**, and **5b** (pK_a about 1.0) was fairly strengthened compared to those of glycine (pK_a 2.22[15]) and alanine (pK_a 2.22[15]) owing to the electron withdrawing pteridinyl group attached to the nitrogen atom.



2, 3	R¹
a	-(CH₂)₅CO₂H
b	-(CH₂)₃CO₂H
c	-CH₂CH₂CO₂H
d	-C(CH₃)H-CO₂H (L)
e	-C(H)CH₃-CO₂H (D)
f	-CH₂CO₂H

5	R²	X
a	-(CH₂)₅CO₂H	-OH
b	-CH₂CO₂H	-OH
c	-(CH₂)₅CO₂H	-H

6	R³
a	-H
b	-C₄H₉

As a model for making a peptide linkage between the present synthesized *N*-(2-pteridinyl)amino acids and a protein, conversion of the carboxyl group of **3a** into a carbamoyl group was examined. First, the carboxylic acid (**3a**) was treated with hydrogen chloride in methanol to give the methyl ester which on subsequent treatment with ammonia gave 6-[(4-hydroxy-6,7-dimethyl-2-pteridinyl)amino]hexanamide (**6a**). The conversion, however, was found to undergo much easier by employing a mixed anhydride[16] as an intermediate: **3a** was treated with ethyl chloroformate in *N*,*N*-dimethylformamide in the presence of triethylamine to form an anhydride. Without isolation, the anhydride was treated with butylamine to give the *N*-butyl-6-(2-pteridinylamino)hexanamide (**6b**) in a good yield. The latter method involving an anhydride would be applicable for making a peptide linkage between the present synthesized pteridines and a protein.

Experimental

The elemental analyses were conducted at the Analytical Section, Meijo University, Nagoya. The pK_a values were determined spectroscopically,[14] and the UV spectra in an appropriate buffer solution on a Shimadzu UV-300 spectrophotometer.

Synthesis of N-*(4-Amino-6-hydroxy-5-nitroso-2-pyrimidinyl)amino Acids(2a—f).* *Method A*: A mixture of 4-amino-6-hydroxy-2-methylthio-5-nitrosopyrimidine (**1**)[17] (10 g) and 6-aminohexanoic acid (20 g) in water (400 ml) was heated under reflux for 1 h. The resulting solution was adjusted to pH 2—3 with formic acid and chilled. Filtration and washing with water gave an orange powder (8.5 g) of 6-[(4-amino-6-hydroxy-5-nitroso-2-pyrimidinyl)amino]hexanoic acid (**2a**), which was pure enough for further reactions. The analytical sample (orange needles) was prepared by dissolving in hot dilute ammonia then acidifing with formic acid, mp 232.5—233.5 °C, dec (Found: C, 44.53; H, 5.65; N, 25.69%. Calcd for $C_{10}H_{15}N_5O_4$: C, 44.60; H, 5.62; N, 26.01%). A similar treatment of **1** with 4-aminobutanoic acid or β-alanine gave the 4-(2-pyrimidinylamino)butanoic acid(**2b**)[77% yield, mp 240—241 °C, dec (from water)(Found: C, 39.33; H, 4.92;

N, 29.32%. Calcd for $C_8H_{11}N_5O_4$: C, 39.83; H, 4.60; N, 29.04%)] and the 3-(2-pyrimidinylamino)propanoic acid(**2c**) [68% yield, mp >300 °C (Found: C, 37.58; H, 4.11; N, 30.65%. Calcd for $C_7H_9N_5O_4$: C, 37.01; H, 3.99; N, 30.83 %)], respectively.

Method B: A solution of **1** (9.0 g) and L-alanine (8.9 g) in 0.25 M sodium hydroxide (400 ml) was heated under reflux for 2 h. After concentration *in vacuo* to about 200 ml, the solution was adjusted to pH 2—3 with formic acid and chilled. The orange flakes (5.8 g, 53%) of the *N*-(2-pyrimidinyl)-L-alanine (**2d**), purified in the same way as above, darkened without melting above 245 °C (Found: C, 34.42; H, 4.50; N, 28.46%. Calcd for $C_7H_9N_5O_4\cdot H_2O$: C, 34.29; H, 4.52; N, 28.56%). Similarly, **1** reacted with D-alanine and glycine to give the *N*-(2-pyrimidinyl)-D-alanine(**2e**)[18% yield; darkened above 245 °C without melting (Found: C, 34.38; H, 4.52; N, 28.28%. Calcd for $C_7H_9N_5O_4\cdot H_2O$: C, 34.29; H, 4.52; N, 28.56%)] and the *N*-(2-pyrimidinyl)-glycine(**2f**)[63% yield; darkened above 280 °C without melting (Found: C, 31.42; H, 3.71; N, 30.33%. Calcd for $C_6H_7N_5O_4\cdot H_2O$: C, 31.17; H, 3.92; N, 30.30%)], respectively.

Synthesis of N-*(4-Hydroxy-6,7-dimethyl-2-pteridinyl)amino Acids (**3a**—**f**)*. A solution of **2a** (4.3 g) in 2 M sodium hydroxide (60 ml) was hydrogenated over palladium on carbon (5%, 2 g) at room temperature and atmospheric pressure till the calculated amount of hydrogen was absorbed. After acidifing with concd hydrochloric acid (20 ml), the catalyst was removed by filtration. The filtrate, after being adjusted to pH 2—3 with ammonia, was heated with biacetyl (3.0 g) under reflux for 1 h. Refrigeration gave a yellow solid, which was crystallized from 50% aqueous methanol to give yellow needles (70 % yield) of 6-[(4-hydroxy-6,7-dimethyl-2-pteridinyl)amino]-hexanoic acid (**3a**), mp 215—219 °C, dec (Found: C, 51.94; H, 6.61; N, 21.30%. Calcd for $C_{14}H_{19}N_5O_3\cdot H_2O$: C, 52.00; H, 6.55; N, 21.66%).

In an analogous way, its lower homologues (**3b**—**f**) were synthesized from the corresponding pyrimidines (**2b**—**f**): the 4-(2-pteridinylamino)butanoic acid (**3b**) [80% yield, mp 238—240 °C, dec (from water) (Found: C, 48.72; H, 5.82; N, 23.40%. Calcd for $C_{12}H_{15}N_5O_3\cdot H_2O$: C, 48.80; H, 5.80; N, 23.72%)], 3-(2-pteridinylamino)propanoic acid (**3c**) [70% yield, darkened above 275 °C without melting (Found: C, 49.73; H, 4.98; N, 26.44%. Calcd for $C_{11}H_{13}N_5O_3$: C, 50.18; H, 4.98; N, 26.61%)], *N*-(2-pteridinyl)-L-alanine (**3d**) [50% yield, darkened above 180 °C and did not show sharp mp (from water) (Found: C, 46.95; H, 5.40; N, 24.63%. Calcd for $C_{11}H_{13}N_5O_3\cdot H_2O$: C, 46.97; H, 5.38; N, 24.90%)], *N*-(2-pteridinyl)-D-alanine (**3e**) [45% yield, darkened above 180 °C without showing sharp mp (from water) (Found: C, 46.84; H, 5.14; N, 24.59%. Calcd for $C_{11}H_{13}N_5O_3\cdot H_2O$: C, 46.97; H, 5.38; N, 24.90%)], *N*-(2-pteridinyl)glycine (**3f**) [87 % yield, mp 206—210 °C, dec (from water) (Found: C, 45.13; H, 4.87; N, 25.95%. Calcd for $C_{10}H_{11}N_5O_4\cdot H_2O$: C, 44.94; H, 4.90; N, 26.21%)].

N-*(4-Hydroxy-2-pteridinyl)glycine (**4**)*. A solution of the 5-amino analogue of **2f** (2.0 g) in water (pH 6—7, 80 ml), prepared in the same way as above, was heated with glyoxal sodium hydrogensulfite (3.5 g) under reflux for 1 h. The solution was adjusted to pH 2—3 with hydrochloric acid and chilled to give a solid. Crystallization of the solid from water gave colorless prisms (1.1 g) of **4** which darkened above 250 °C without melting (Found: C, 40.28; H, 3.77; N, 28.94%. Calcd for $C_8H_7N_5O_3\cdot H_2O$: C, 40.17; H, 3.79; N, 29.28%).

*6-[[4-Hydroxy-6-(*L-erythro-*1,2,3-trihydroxypropyl)-2-pteridinyl]amino]hexanoic Acid (**5a**) and the Homologue (**5b**)*. A solution of **2a** (4.0 g) in 2 M sodium hydroxide (100 ml) was hydrogenated and separated from the catalyst as above. The filtrate was adjusted to pH 3—4, diluted with methanol (100 ml), and then heated with L-arabinose phenylhydrazone (3.8 g) under nitrogen and reflux for 2 h. After cooling, the solution was adjusted to pH 8—9 with ammonia and stirred under bubbling air at room temperature for 20 h. The orange solution was evaporated to dryness *in vacuo* and the residue was extracted with 1 M ammonia (200 ml). The extract was adjusted to pH 2—3 with formic acid and chromatographed on a Florisil column[1] (4.5×40 cm) using water as the elution solvent. The eluate was evaporated to dryness and the residue was extracted with hot water (100 ml). The extract, on concentration to about 50 ml and chilling, gave pale yellow prisms (2.0 g) of **5a**, mp 186—190 °C, dec (from water) (Found: C, 46.55; H, 5.84; N, 17.95%. Calcd for $C_{15}H_{21}N_5O_6\cdot H_2O$: C, 46.75; H, 6.02; N, 18.17%).

By using **2f** in place of **2a** in the foregoing condensation, *N*-[4-hydroxy-6-(L-*erythro*-1,2,3-trihydroxypropyl)-2-pteridinyl]glycine (**5b**) was obtained in 53% yield as colorless needles, mp 177—180 °C, dec (from water) (Found: C, 40.15; H, 4.49; N, 20.98%. Calcd for $C_{11}H_{13}N_5O_6\cdot H_2O$: C, 40.12; H, 4.59; N, 21.27%).

*6-[[4-Hydroxy-6-(*L-erythro-*1,2-dihydroxypropyl)-2-pteridinyl]-amino]hexanoic Acid (**5c**)*. The 5-nitrosopyrimidine (**2a**) (4.0 g) was hydrogenated in the same way as above to the 5-amino analogue, which was heated under reflux with 5-deoxy-L-arabinose phenylhydrazone (4.5 g) in 50% aqueous methanol (200 ml) at pH 3—4 and under nitrogen for 20 min. The solution, after cooling in an ice bath, was added to an ice chilled solution of potassium hexacyanoferrate (III) (30 g), potassium iodide (5 g), 35% hydrogen peroxide (10 ml), and formic acid (10 ml) in water (200 ml). The mixture was stirred under bubbling oxygen at 0—5 °C for 2 h and then at 20—25 °C for 10 h. The resulting solution was concentrated *in vacuo* to about 100 ml and chromatographed on a Florisil column as above. The eluate was evaporated to dryness and the residue was extracted with methanol (200 ml). Evaporation of the extract and subsequent crystallization from 90% ethanol gave pale yellow needles (0.47 g) of **5c**, mp 216—218 °C, dec (from 90% ethanol) (Found: C, 48.55; H, 6.25; N, 18.55%. Calcd for $C_{15}H_{21}N_5O_5\cdot H_2O$: C, 48.77; H, 6.28; N, 18.96%).

*Conversion of **3a** into the Carboxamides (**6a** and **6b**)*.

Method A: To a solution of **3a** (300 mg) in anhydrous methanol (30 ml), dry hydrogen chloride was passed till saturation. The solution, after being allowed to stand at 25 °C for 10 h, was evaporated to dryness *in vacuo*. The residue was dissolved in 20% aqueous ammonia (100 ml) and kept at 25 °C for 10 h. Evaporation of the solution gave a solid which was chromatographed on a Florisil column (2× 20 cm), eluted by 0—2% ammonia (500 ml) gradiently. The eluate, on evaporation to dryness and subsequent crystallization from dil formic acid, gave colorless needles (200 mg) of the 6-(2-pteridinylamino)hexanamide (**6a**), mp 212—215 °C dec (from water) (Found: C, 52.07; H, 6.88; N, 26.01%. Calcd for $C_{14}H_{20}N_6O_2\cdot H_2O$: C, 52.16; H, 6.88; N, 26.07%).

Method B: Ethyl chloroformate (50 mg) was added to a solution of **3a** (125 mg) and triethylamine (100 mg) in *N*,*N*-dimethylformamide (1 ml) at −5 °C. After stirring for 15 min, butylamine (75 mg) was added to the solution. The mixture was stirred at −5 °C for 30 min and then at 25 °C for 1 h. Evaporation of the solution *in vacuo* gave an oily residue, which was chromatographed on a Florisil column (1×20 cm) eluted by 20% aqueous acetone containing 2% ammonia. The eluate was evaporated to dryness and the residue was extracted with ethanol (50 ml). Concentration of the extract to about 5 ml and chilling gave pale yellow

needles (47 mg) of the *N*-butyl-6-(2-pteridinylamino)hexanamide (**6b**), which decomposed above 150 °C (from 90% ethanol) (Found: C, 57.21; H, 7.53; N, 22.04%. Calcd for $C_{18}H_{28}N_6O_2 \cdot H_2O$: C, 57.11; H, 7.99; N, 22.21%).

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