

A Convenient Synthesis of Aminopterin and Homologs *via* 6-(Bromomethyl)-2,4-diaminopteridine Hydrobromide (1)

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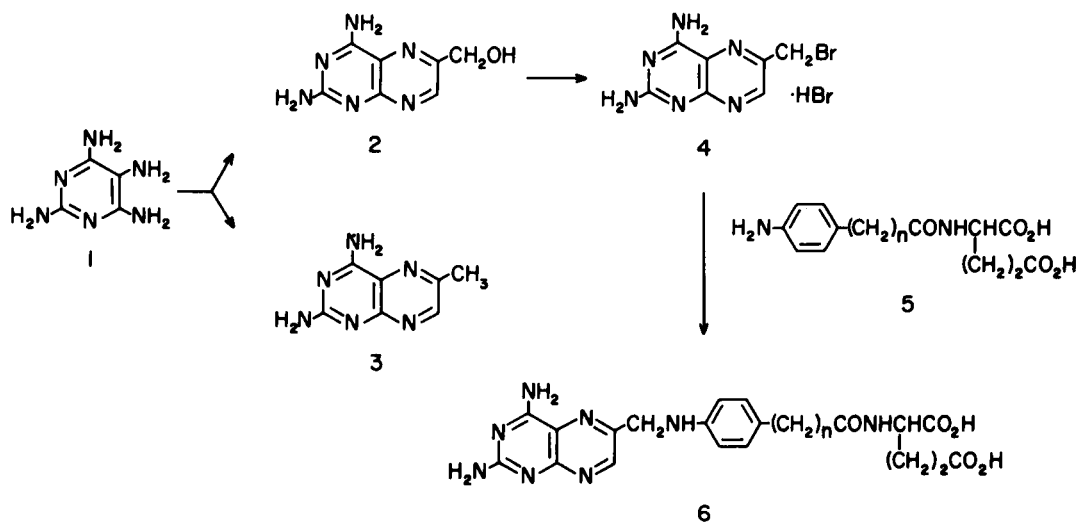
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Sir:

We wish to describe a facile preparation of 6-(bromomethyl)-2,4-diaminopteridine hydrobromide (**4**) and its conversion to the anticancer agent aminopterin (**6**, $n = 0$) and homologs (**6**, $n = 1, 2$) in good yields and high purity. This simple approach to **6**-types represents a marked improvement over methods used previously that give low yields of products requiring purification by laborious and tedious techniques (**2**).

The preparation of **4** has not heretofore been reported. There have been several reports on the preparation of folic acid from 2-amino-6-(halogenomethyl)-4-pteridinol (**3**), but the approach apparently preferred in more recent syntheses of folic acid and its analogs is that *via* 2-acetamido-4-hydroxy-6-pteridinecarboxaldehyde (**4**). 7-Methylaminopterin and 7-methylmethopterin were recently prepared (**5**) from the corresponding 6-(bromomethyl)-substituted pteridine by methods similar to those reported earlier for the preparation of 7-methylfolic acid and 7,10-dimethylfolic acid (**6**). In those reports the 6-(bromomethyl)-7-methyl-substituted pteridines were derived from 6,7-bis(bromomethyl)-precursors, which were prepared by condensation of the appropriate pyrimidines and dibromodiacetyl.

Essential features of the preparation of **4** are as follows. The material obtained directly from the condensation of 2,4,5,6-tetraaminopyrimidine (**1**) and 1,3-dihydroxyacetone according to a reported procedure (**7**) consisted mainly of 2,4-diamino-6-pteridinemethanol (**2**) and 2,4-diamino-6-methylpteridine (**3**) with **2** in dominance over **3** by a ratio of approximately 5:1. The relative amounts were estimated from pmr spectral data in deuterio-trifluoroacetic acid; compound **2** gave signals at δ 5.3 (6-CH₂O-) and δ 9.1 (7-H), and **3** at δ 2.8 (6-CH₃) and δ 8.8 (7-H). Treatment of a suspension of the crude mixture of **2** and **3** in boiling ethanol with an equimolar amount of 48% hydrobromic acid gave their hydrobromides, and the greater solubility of **3**·HBr in ethanol allowed its nearly complete removal from the desired **2**·HBr. The pmr spectrum of the product (typically obtained in 39% yield) showed only **2**·HBr and **3**·HBr with **2**·HBr in dominance by 16-20:1, depending on the extent of extraction with boiling ethanol. Treatment of the **2**·HBr thus prepared with triphenylphosphine dibromide (**8**) (four molar equivalents, preformed *in situ* from triphenylphosphine and bromine) in DMAC at 20-25° for 1.5-2 hours led to **4**, but pmr spectral data showed the relative proportion of **4** (δ 4.7, 6-CH₂Br, in deuterio-trifluoroacetic acid) to **3**·HBr in each of three runs to be



only slightly improved over that of 2·HBr to 3·HBr in the starting material. The work-up procedure was as follows. The reaction mixture was treated with ethanol, left in a refrigerator overnight, and evaporated *in vacuo* (bath up to 45°). The residue, a dark oil, gave a solid when stirred with warm benzene. The liquid phase was then removed by decantation, and the benzene-insoluble solid was dissolved in glacial acetic acid at 80°. The cooled solution deposited an off-white crystalline solid, which was ultimately freed of acetic acid by drying *in vacuo* (phosphorus pentoxide) at 110° to give yellow, crystalline product in 60-65% yield (three runs). A sample of 4 (C₇H₇BrN₆·HBr) that gave a satisfactory elemental analysis (C, H, Br, and N) (9), although it still contained detectable 3·HBr and was estimated to be of 95% purity, gave the following uv spectral data: λ max, nm (ε × 10⁻³), 0.1 N hydrochloric acid, 249 (17.1), 339 (10.6), 353 (sh) (9.4); 0.1 N sodium hydroxide, 258 (22.1), 372 (7.2). The 4 obtained in this manner proved to be suitable for the preparation of 6-types.

Treatment of *N*-(*p*-aminobenzoyl)glutamic acid (5, n = 0) and its homologs (5, n = 1, 2) (three molar equivalents) with 4 in DMAC (20 hours at 20-25°) gave 6 (n = 0, 1, 2) in respective yields of 68, 73, and 39%. Addition of water to the reaction mixtures caused precipitation of the products. Two of the 6-types (n = 0, 1) required no purification other than thorough washing with water, and 6 (n = 2) was obtained pure following reprecipitation from Norit-treated 0.1 N sodium hydroxide solution by addition of an equivalent amount of hydrochloric acid. Each of these products gave satisfactory elemental analyses (C, H, N) (9) and migrated as single uv-absorbing spots on thin-layer chromatograms. The spots from 6 (n = 1, 2) had a thin, faintly fluorescent cap, and that from 6 (n = 0) showed no fluorescence (10). Their pmr spectra were as expected with no indication of the continued presence of 3. The uv spectrum of 6 (n = 0) agrees with that previously reported (2a, e).

Acknowledgment.

N-(*p*-Aminophenylacetyl)glutamic acid (5, n = 1) was prepared by Mr. Jerry L. Frye by reduction of *N*-(*p*-nitrophenylacetyl)glutamic acid. *N*-(*p*-Aminohydrocinnamoyl)-

glutamic acid (5, n = 2) was prepared by Mr. Jerry D. Rose by the following sequence: *p*-nitrocinnamic acid → acyl chloride → *N*-acylated diethyl glutamate → *N*-acylated glutamic acid → 5 (n = 2). Reduction steps in both sequences were by catalytic hydrogenation (5% Pd on charcoal) in water.

REFERENCES

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- (7) C. M. Baugh and E. Shaw, *J. Org. Chem.*, **29**, 3610 (1964).
- (8) Cf. G. A. Wiley, R. L. Hershkowitz, B. M. Rein, and B. C. Chung, *J. Am. Chem. Soc.*, **86**, 964 (1964).
- (9) Satisfactory elemental analyses (± 0.4%) were obtained on designated compounds for the elements given in parentheses. Results for 6 (n = 0) correspond to C₁₉H₂₀N₈O₅·1.75H₂O, those for 6 (n = 1) to C₂₀H₂₂N₈O₅·H₂O, and for 6 (n = 2) to C₂₁H₂₄N₈O₅·2H₂O.
- (10) Thin-layer chromatograms were run on Bakerflex DEAE-cellulose sheets using 0.5 M sodium chloride, 0.2 M in mercaptoethanol, in 0.005 M potassium phosphate buffer at pH 7.0. The chromatograms exhibited characteristics like those of related compounds as described by R. B. Angier and W. V. Curran [*J. Am. Chem. Soc.*, **81**, 2814 (1959)].