A New Synthesis of *as*-Triazines and Pyrimido[4,5-*e*]-*as*-triazines (6-Azapteridines)

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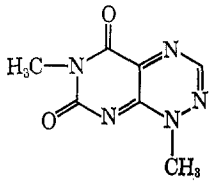
A new synthesis of *as*-triazines has been developed which involves Michael addition of diethyl azodicarboxylate to acyclic enamines, followed by base-catalyzed ring closure. By an appropriate choice of the enamine, *as*-triazines suitably substituted for subsequent annelation of a fused pyrimidine ring may be prepared. We describe in this paper the synthesis of a number of pyrimido[4,5-*e*]-*as*-triazines (6-azapteridines), including 2-methylisofervenulone (**20**) and 3,6,8-triaminopyrimido[4,5-*e*]-*as*-triazine (**23**), by this new route.

The discovery of the triad of naturally occurring antibiotics toxoflavin (**1**),[2,3] fervenulin (**2**),[4] and 2-methylfervenulone (MSD-92) (**3**),[5] and their identification by degradation and total synthesis as derivatives of the pyrimido[5,4-e]-*as*-triazine (7-azapteridine) ring system (**4**), has stimulated considerable recent interest in the synthesis and chemistry of further derivatives.



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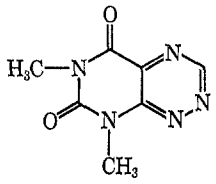

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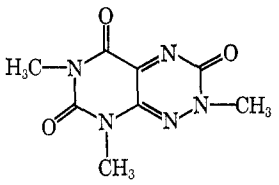

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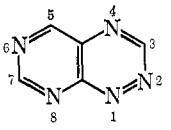

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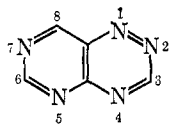

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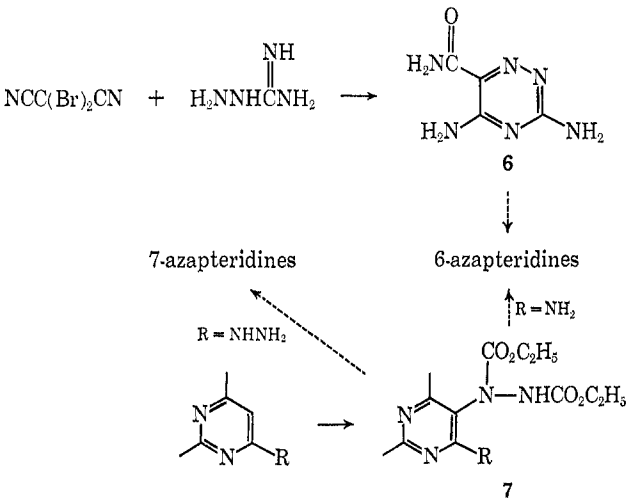
The isomeric pyrimido[4,5-e]-*as*-triazine (6-azapteridine) system (**5**) is also attracting recent attention because of the discovery that some derivatives exhibit antiviral activity.[6]

Almost all previous synthetic routes to pyrimidotriazines have involved the initial construction of a suitably substituted pyrimidine precursor and the eventual annelation of the condensed triazine ring in a terminal reaction step.[7-9] One exception to this order of construction of the bicyclic system was described by Taylor and Morrison,[10] who prepared 3,5-diamino-*as*-triazine-6-carboxamide (**6**) by condensation of dibromomalononitrile with aminoguanidine, and subsequently converted this intermediate to 6-azapteridines by cyclization with appropriate one-carbon reagents. We describe in this paper a further entry into the pyrimido[4,5-*e*]-*as*-triazine system *via* triazine intermediates.



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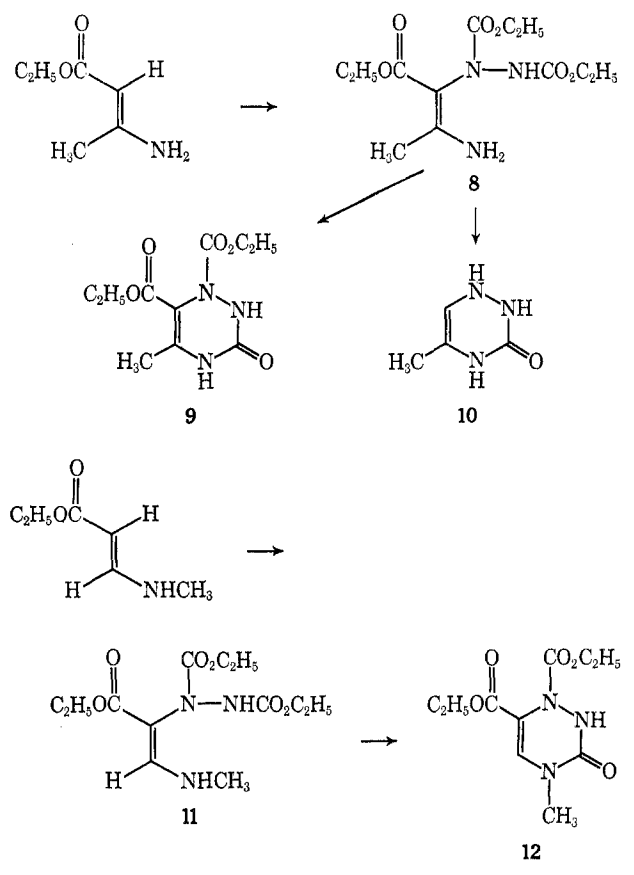
In our previously published total syntheses of fervenulin[4] and MSD-92,[5] a key step was the condensation of a 6-hydrazinopyrimidine with diethyl azodicarboxyl-



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(3) T. K. Liao, F. Baiocchi, and C. C. Cheng, *J. Org. Chem.*, **31**, 900 (1966), and references cited therein.

(4) E. C. Taylor and F. Sowinski, *J. Amer. Chem. Soc.*, **90**, 1374 (1968), and references cited therein.

(5) E. C. Taylor and F. Sowinski, *ibid.*, **91**, 2143 (1969), and references cited therein.

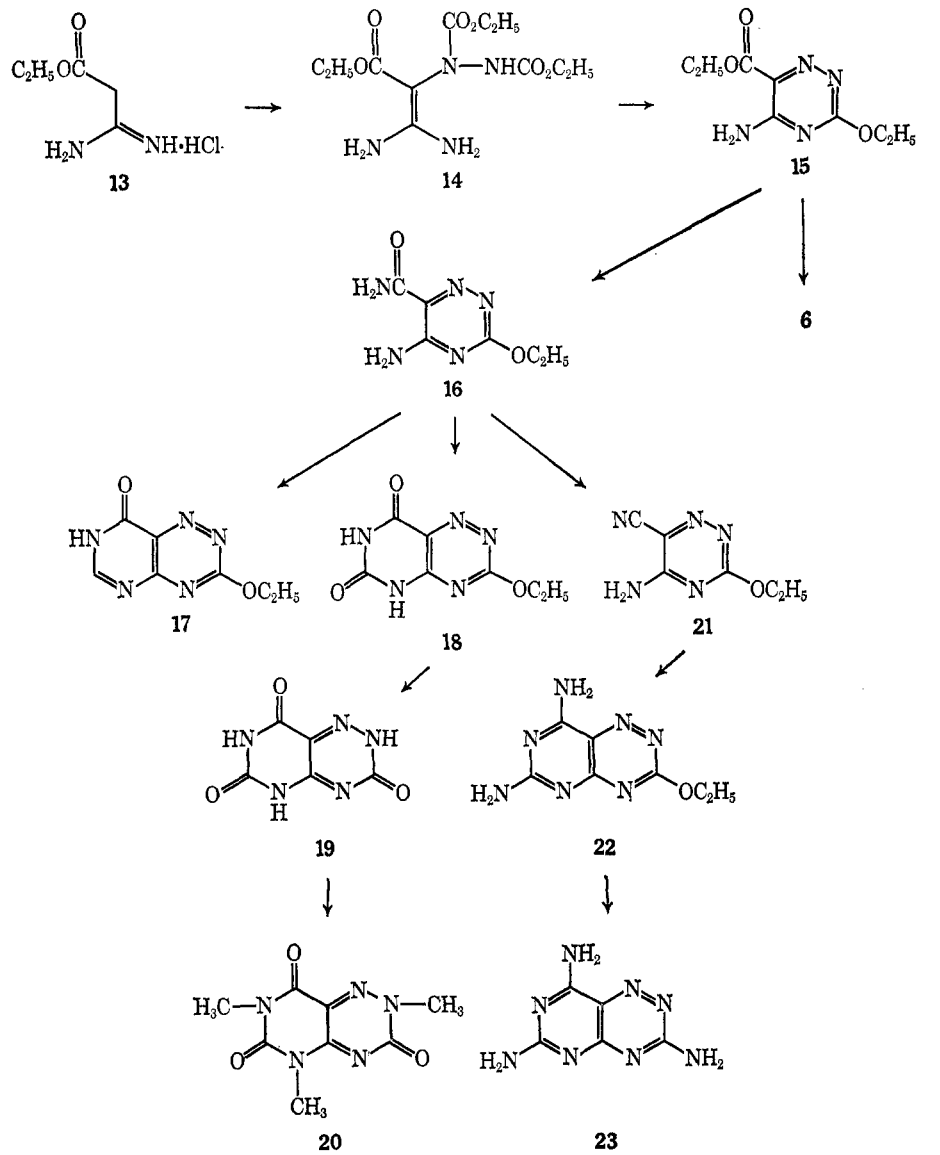
(6) C. Kuchler, W. Kuchler, and L. Heinisch, *Arzneim.-Forsch.*, **16**, 1122 (1966).

(7) C. Temple, Jr., C. L. Kussner, and J. A. Montgomery, *J. Org. Chem.*, **34**, 2102 (1969), and references cited therein; these workers also described the ring cleavage of pyrimido[5,4-*e*]-*as*-triazin-5(6*H*)-one to 6-amino-*as*-triazine-5-carboxamide, and the subsequent recyclization of this latter compound to several 7-azapteridines.

(8) M. E. C. Biffin, D. J. Brown, and T. Sugimoto, *J. Chem. Soc. C*, 139 (1970).

(9) W. Pfleiderer and G. Blankenhorn, *Tetrahedron Lett.*, 4699 (1969).

(10) E. C. Taylor and R. W. Morrison, Jr., *J. Amer. Chem. Soc.*, **87**, 1976 (1965).



ate to give a Michael adduct (**7**, R = $NHNH_2$), which was subsequently modified to form the annelated *as*-triazine ring. The same principle was applied to the synthesis of 6-azapteridines by Michael addition of diethyl azodicarboxylate to 6-aminopyrimidines to give **7** (R = NH_2), followed by suitable ring closure techniques. In both instances, the initial Michael reaction leading to the adducts **7** (R = $NHNH_2$, NH_2) reflects enamine-type reactivity of the 6-hydrazino- (or amino-) 5-unsubstituted pyrimidines. It occurred to us that extrapolation of these reactions to acyclic enamines should provide an entry into *as*-triazines which, with an appropriate choice of substituents, might serve as precursors for subsequent closure of the annelated pyrimidine ring, and thus offer an alternate synthetic pathway to azapteridines.

The potential feasibility of this approach was demonstrated by the following sequence of reactions. Michael addition of diethyl azodicarboxylate to ethyl β-aminocrotonate gave ethyl α-(1,2-dicarbethoxyhydrazino)-β-aminocrotonate (**8**), which was cyclized in quantitative yield with thallium(I) ethoxide[11] in benzene to **9**. Cyclization of **8** could also be effected with aqueous sodium hydroxide, but under these conditions concomitant hydrolysis and decarboxylation of the carbethoxy group occurred to give **10**. In analogous fashion, ethyl β-methylaminoacrylate condensed with diethyl azodicarboxylate to give ethyl α-(1,2-dicarbethoxyhydrazino)-β-methylaminoacrylate (**11**), which was cyclized with thallium(I) ethoxide in benzene to **12**.

Efforts were then directed toward the preparation of an enamine so substituted that the resulting *as*-triazine could be subsequently cyclized to 6-azapteridines. Condensation of diethyl azodicarboxylate with carbethoxyacetamidine hydrochloride (**13**) in the presence of triethylamine gave α-(1,2-dicarbethoxyhydrazino)carbethoxyacetamidine (**14**). Although all attempts to effect cyclization of this intermediate to an *as*-triazine with basic reagents failed, it was found that a combination of bromine with Hünig's base (diisopropylethylamine)[12] smoothly afforded 3-ethoxy-5-amino-6-carbethoxy-*as*-triazine (**15**). The structure of this compound was confirmed by treatment with ethanolic ammonia at 150° to give 3,5-diamino-6-carbamoyl-*as*-triazine (**6**), identical with an authentic sample prepared by the alternate method of Taylor and Morrison.[10]

(11) E. C. Taylor, G. H. Hawks, III, and A. McKillop, *J. Amer. Chem. Soc.*, **90**, 2421 (1968).

(12) N. Finch and C. W. Gemenden, *Tetrahedron Lett.*, 1203 (1969).

Aminolysis of **15** at room temperature led to 3-ethoxy-5-amino-6-carbamoyl-*as*-triazine (**16**), which was then cyclized to a variety of 6-azapteridines. Thus, condensation with diethoxymethyl acetate gave 3-ethoxy-8(7*H*)-pyrimido[4,5-*e*]-*as*-triazinone (**17**), while reaction with diethyl carbonate in the presence of sodium ethoxide gave 3-ethoxy-6,8(5*H*,7*H*)-pyrimido[4,5-e]-*as*-triazinedione (**18**). Acid hydrolysis of this latter compound afforded 3,6,8(2*H*,5*H*,7*H*)-pyrimido[4,5-e]-*as*-triazinetrione (**19**), identical in physical properties with those reported for this same compound prepared independently *via* the pyrimidine annelation route.[13] Furthermore, methylation of **19** with methyl iodide and sodium hydride in anhydrous dimethylformamide gave 2-methylisofervenulone (**20**), identical with an authentic sample.[14]

Dehydration of the *o*-aminoamide **16** with phosphorus oxychloride in pyridine gave 3-ethoxy-5-amino-6-cyano-*as*-triazine (**21**), which was cyclized with guanidine to 3-ethoxy-6,8-diaminopyrimido[4,5-e]-*as*-triazine (**22**).[15] Subsequent treatment with ethanolic ammonia at 150° gave 3,6,8-triaminopyrimido[4,5-e]-*as*-triazine (**23**). This compound is of particular interest because of the known antifolic acid activity of 2,4-diaminopteridines and other condensed 2,4-diaminopyrimidine systems,[16] and the diuretic activity of 2,4,7-triaminopteridines.[17]

Experimental Section[18]

1,6-Dicarbethoxy-5-methyl-1,4-dihydro-*as*-triazin-3(2*H*)-one (9).[19]—To a solution of 6.45 g (0.05 mol) of ethyl β-aminocrotonate in 25 ml of anhydrous benzene was added, all at once, 8.70 g (0.05 mol) of diethyl azodicarboxylate. The reaction mixture was stirred at room temperature for 2.5 hr and evaporated under reduced pressure and the residual viscous oil dissolved in hot ethyl acetate. Cooling and scratching resulted in the crystallization of a colorless solid which was recrystallized from ethanol. The intermediate ethyl α-(1,2-dicarbethoxyhydrazino)-β-aminocrotonate (**8**) thus obtained weighed 9.10 g (60%) and melted at 92–96°.

To a suspension of 3.03 g (0.01 mol) of **8** in 15 ml of anhydrous ether was added a solution of 7.50 g (0.03 mol) of thallium(I) ethoxide[11] in 25 ml of anhydrous benzene. The mixture was refluxed for 2 hr, cooled, and neutralized with 1.80 g (0.03 mol) of glacial acetic acid. The thallium(I) acetate which separated was collected by filtration and washed thoroughly with methylene chloride, and the combined filtrate and washings were concentrated under reduced pressure. The residual solid was recrystallized from benzene–pentane to give 2.55 g (99%), mp 166°.

Anal. Calcd for $C_{10}H_{15}N_3O_5$: C, 46.69; H, 5.88; N, 16.34. Found: C, 46.71; H, 5.84; N, 16.31.

5-Methyl-1,4-dihydro-*as*-triazin-3(2*H*)-one (10).[19]—A solution of 10.00 g (0.34 mol) of ethyl α-(1,2-dicarbethoxyhydrazino)-β-aminocrotonate (**8**) (see above) in 30 ml of 5 *N* sodium hydroxide and 30 ml of ethanol was boiled (no reflux condenser) for approximately 2 hr, during which time most of the ethanol had evaporated. The resulting aqueous solution was cooled to room temperature, allowed to stand overnight, and then neutralized with glacial acetic acid. Constant extraction with hot chloroform, followed by evaporation of the extracts, gave a white solid which was recrystallized from ethanol to yield 1.98 g (52%), mp 222–224°.

Anal. Calcd for $C_4H_7N_3O$: C, 42.47; H, 6.24; N, 37.15. Found: C, 42.08; H, 6.11; N, 36.74.

Ethyl α-(1,2-Dicarbethoxyhydrazino)-β-methylaminoacrylate (11).—To a solution of 1.50 g (0.0116 mol) of ethyl β-methylaminoacrylate in 5 ml of anhydrous benzene was added, in one portion, 2.04 g (0.0116 mol) of diethyl azodicarboxylate. The reaction mixture was stirred at room temperature for 5 hr, the excess solvent removed by evaporation under reduced pressure, and approximately 10 ml of pentane added to the residual oil. Scratching and cooling induced the crystallization of a cream-colored solid which, upon recrystallization from a mixture of benzene and pentane, gave 2.68 g (77%), mp 90–92°.

Anal. Calcd for $C_{12}H_{21}N_3O_6$: C, 47.52; H, 6.98; N, 13.86. Found: C, 47.38; H, 6.92; N, 13.99.

1,6-Dicarbethoxy-4-methyl-1,4-dihydro-*as*-triazin-3(2*H*)-one (12).—Treatment of 2.55 g (0.0084 mol) of **11** with 6.30 g (0.0242 mol) of thallium(I) ethoxide in benzene, as described above for the conversion of **8** to **9**, gave colorless crystals which were recrystallized from a mixture of chloroform and pentane to give 1.61 g (75%), mp 172–173°.

Anal. Calcd for $C_{10}H_{15}N_3O_5$: C, 46.69; H, 5.58; N, 16.34. Found: C, 46.64; H, 5.71; N, 16.08.

α-(1,2-Dicarbethoxyhydrazino)carbethoxyacetamidine (14).—To a suspension of 12.48 g (0.075 mol) of carbethoxyacetamidine hydrochloride (**13**)[20] in 45 ml of anhydrous benzene was added, in one portion, 13.08 g (0.075 mol) of diethyl azodicarboxylate, followed by dropwise addition of 7.56 g (0.075 mol) of triethylamine in 10 ml of anhydrous benzene. The resulting reaction mixture was stirred at room temperature for 3 hr, the triethylamine hydrochloride collected by filtration, and the filtrate concentrated under reduced pressure. The residual syrup was chromatographed on 500 g of silica gel (Baker) with ethyl acetate–benzene (1:3). The combined eluents were evaporated under reduced pressure and the residual gum recrystallized from chloroform–pentane to give 13.40 g (59%) of a colorless product, mp 110–111°.

Anal. Calcd for $C_{11}H_{20}N_4O_6$: C, 43.41; H, 6.63; N, 18.41. Found: C, 43.61; H, 6.55; N, 18.26.

3-Ethoxy-5-amino-6-carbethoxy-*as*-triazine (15).—To a solution of 15.20 g (0.05 mol) of **14** in 125 ml of methylene chloride, cooled in an ice–salt bath to 0°, was added 14.10 g (0.11 mol) of Hünig's base,[12] followed by dropwise addition of 8.90 g (0.055 mol) of bromine in 25 ml of methylene chloride. Stirring was continued at 0° for 1 hr and the resulting solution then extracted with three 150-ml portions of ice–water. The methylene chloride was dried over anhydrous magnesium sulfate, concentrated under reduced pressure, and the residual syrup chromatographed on 350 g of silica gel (Baker) using ethyl acetate–chloroform (1:10). The combined eluents were concentrated under reduced pressure and the residual solid recrystallized from benzene–pentane to give 4.45 g (42%), mp 127–128°.

Anal. Calcd for $C_8H_{12}N_4O_3$: C, 45.28; H, 5.70; N, 26.40. Found: C, 45.53; H, 5.44; N, 26.30.

3,5-Diamino-6-carbamoyl-*as*-triazine (6).—A solution of 0.20 g of **15** in 10 ml of ethanol saturated with dry ammonia was heated in a sealed tube at 150° (oil bath temperature) for 24 hr. The reaction mixture was evaporated to dryness under reduced pressure, the residue suspended in boiling water, and 1 *N* hydrochloric acid added until complete solution resulted. The resulting solution was decolorized with Norite and the pH of the filtrate then adjusted to neutrality with 5% ammonium hydroxide. The white solid which separated on cooling was collected by filtration to give 90 mg (62%), mp <350°. The product was identical with an authentic sample of 3,5-diamino-6-carbamoyl-*as*-triazine prepared independently.[10]

3-Ethoxy-5-amino-6-carbamoyl-*as*-triazine (16).—A solution of 2.50 g of **15** in 100 ml of anhydrous ethanol saturated with dry ammonia was stirred at room temperature for 24 hr. The reaction mixture was concentrated under reduced pressure to approximately 25 ml, cooled, and the white solid which separated collected by filtration, washed with ethanol, and recrystallized from aqueous dimethylformamide to yield 1.83 g (85%), mp 228–229°.

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(14) F. Sowinski, Ph.D. Thesis, Princeton University, 1971.

(15) E. C. Taylor and A. McKillop, "The Chemistry of Cyclic Enaminonitriles and *o*-Aminonitriles," Interscience, New York, N. Y., 1970.

(16) (a) "Experimental Chemotherapy," Vol. IV, Part I, R. J. Schnitzer and F. Hawking, Eds., Academic Press, New York, N. Y., 1966; (b) L. F. Larionov, "Cancer Chemotherapy," Pergamon Press, Oxford, 1965; (c) F. E. Knock, "Anticancer Agents," Charles C Thomas Publisher, Springfield, Ill., 1966.

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(19) We are indebted to Dr. G. W. McClay for this preparation.

(20) S. M. McElvain and B. E. Tate, *J. Amer. Chem. Soc.*, **73**, 2760 (1951).

Anal. Calcd for $C_6H_9N_5O_2$: C, 39.34; H, 4.95; N, 38.24. Found: C, 39.46; H, 4.97; N, 38.23.

3-Ethoxy-8(7*H*)-pyrimido[4,5-*e*]-*as*-triazinone (17).—A suspension of 0.50 g of **16** in 10 ml of diethoxymethyl acetate[21] was heated with stirring for 4 hr in an oil bath maintained at 100°. The resulting red solution was evaporated to dryness under reduced pressure and the residue triturated with 20 ml of chloroform and filtered. Recrystallization of the resulting solid from isopropyl alcohol then gave 0.37 g (69%), mp 204–205° dec: uv $\lambda_{max}^{C_2H_5OH}$ 321, 251 (sh), 244, 221 nm (ϵ 7700, 6500, 7300, 15,600); nmr (DMSO-d_6–TMS) δ 8.34 (s, 1 H).

Anal. Calcd for $C_7H_7N_5O_2$: C, 43.52; H, 3.65; N, 36.26. Found: C, 43.27; H, 3.88; N, 36.07.

3-Ethoxy-6,8(5*H*,7*H*)-pyrimido[4,5-*e*]-*as*-triazinedione (18).—To a suspension of 0.40 g (2.16 mmol) of **16** in ethanolic sodium ethoxide (prepared from 0.21 g of sodium and 15 ml of anhydrous ethanol) was added 1.05 g (8.80 mmol) of diethyl carbonate. The reaction mixture was heated under reflux for 5 hr and cooled, and the precipitated solid collected by filtration and washed well with ethanol. It was then dissolved in 10 ml of water and the resulting solution acidified with glacial acetic acid, cooled immediately, and filtered. The collected solid was recrystallized from ethanol to give 0.35 g (78%), mp 247–248°: uv $\lambda_{max}^{C_2H_5OH}$ 310, 232 nm (ϵ 12,000, 12,100).

Anal. Calcd for $C_7H_7N_5O_3$: C, 40.19; H, 3.37; N, 33.48. Found: C, 40.45; H, 3.41; N, 33.75.

3,6,8(2*H*,5*H*,7*H*)-Pyrimido[4,5-*e*]-*as*-triazinetrione (19).—A suspension of 0.10 g of **18** in 2 ml of 2 *N* hydrochloric acid was heated under reflux for 1 hr. Solution was initially achieved, but as heating progressed a solid gradually precipitated from the hot solution. The reaction mixture was cooled and filtered, and the collected solid recrystallized from water to give 0.08 g (92%), mp >300°.

Anal. Calcd for $C_6H_3N_5O_3$: C, 33.16; H, 1.67; N, 38.67. Found: C, 33.04; H, 2.07; N, 38.46.

The spectral data for this compound were essentially identical with those previously reported: uv $\lambda_{max}^{H_2O}$ 313 nm (ϵ 5500) [lit.[18] 306 nm (ϵ 5600)].

2,5,7-Trimethyl-3,6,8(2*H*,5*H*,7*H*)-pyrimido[4,5-*e*]-*as*-triazinetrione (2-Methylisofervenulone) (20).—To an ice-cooled suspension of 50 mg (0.28 mmol) of **19** in 1 ml of anhydrous dimethylformamide was added 60 mg (1.25 mmol) of a 50% dispersion of sodium hydride in mineral oil. The mixture was stirred at ice bath temperature for 30 min and then 310 mg (2.21 mmol) of methyl iodide added. The resulting mixture was heated under reflux for 30 min and evaporated under reduced pressure, and the residual solid partitioned between 5 ml of chloroform and 1 ml of water. The chloroform layer was separated, dried over anhydrous magnesium sulfate, and concentrated to a viscous oil under reduced pressure. Chromatography on 5 g of silica gel (Merck, 0.05–0.2 mm), using ethyl acetate as the eluting solvent, yielded 25 mg (30%), mp 184–185° dec, identical in all respects with an authentic sample.[14]

3-Ethoxy-5-amino-6-cyano-*as*-triazine (21).—To a suspension of 1.00 g of 3-ethoxy-5-amino-6-carbamoyl-*as*-triazine in 10 ml of anhydrous pyridine was added dropwise 2 ml of phosphorus oxychloride. The reaction mixture was warmed to 45–50°, maintained at that temperature for 10 min, and then poured slowly over 100 g of ice. The crude product was collected by filtration and recrystallized from acetonitrile to give 0.45 g (50%), mp 225° dec.

Anal. Calcd for $C_6H_7N_5O$: C, 43.63; H, 4.27; N, 42.41. Found: C, 43.87; H, 4.41; N, 42.57.

3-Ethoxy-6,8-diaminopyrimido[4,5-*e*]-*as*-triazine (22).—Guanidine hydrochloride (0.27 g, 2.82 mmol) was dissolved in ethanolic sodium ethoxide (prepared from 0.065 g, 2.82 mmol, of sodium and 20 ml of anhydrous ethanol), the precipitated sodium chloride removed by filtration, and 0.30 g (1.87 mmol) of **21** added to the filtrate. The reaction mixture was stirred at room temperature for 10 min and then heated under reflux for 20 min and cooled, and the precipitated yellow solid was collected by filtration and washed well with water followed by ethanol. Recrystallization from ethanolic dimethylformamide gave 0.29 g (76%), mp >300°: uv $\lambda_{max}^{CH_3OH}$ 347, 325 (sh), 253 nm (ϵ 9700, 7500, 19,800).

Anal. Calcd for $C_7H_9N_7O$: C, 40.57; H, 4.38; N, 47.32. Found: C, 40.39; H, 4.33; N, 47.08.

3,6,8-Triaminopyrimido[4,5-*e*]-*as*-triazine (23).—A suspension of 0.15 g of **22** in 15 ml of anhydrous ethanol saturated with dry ammonia was heated in a sealed tube for 24 hr at 150° (oil bath temperature). The reaction mixture was then filtered and the collected yellow solid recrystallized from ethanolic dimethylformamide to give 0.11 g (85%), mp >300°.

The compound was most satisfactorily analyzed as its ditosyl salt, prepared by addition of 150 mg of *p*-toluenesulfonic acid to a suspension of 50 mg of the free base in 2 ml of boiling ethanol. The analytical sample of the ditosyl salt was prepared by recrystallization from methanol–ether, mp 278–279° dec: uv $\lambda_{max}^{CH_3OH}$ 343, 257 nm (ϵ 16,200, 28,400).

Anal. Calcd for $C_{19}H_{22}N_8O_6S_2$: C, 43.66; H, 4.24; N, 21.45. Found: C, 43.66; H, 4.22; N, 21.62.

Registry No.—**6**, 1501-48-0; **8**, 26154-44-9; **9**, 26154-45-0; **10**, 26154-46-1; **11**, 26154-47-2; **12**, 26154-48-3; **14**, 26154-49-4; **15**, 26154-50-7; **16**, 26154-51-8; **17**, 26154-52-9; **18**, 26154-53-0; **19**, 26154-54-1; **20**, 26154-55-2; **21**, 26154-56-3; **22**, 26154-57-4; **23**, 26154-58-5; **23** (ditosyl), 26154-59-6.

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