Irreversible Enzyme Inhibitors LXXXVI

Hydrophobic Bonding to Dihydrofolic Reductase VIII. Substituted-1-aryl-4,6-diamino-1,2-dihydro-2,2-dimethyl-*s*-triazines

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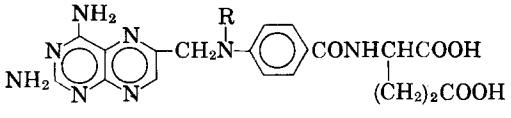
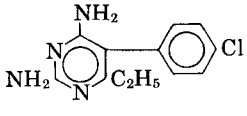
A series of 1-phenyl-4,6-diamino-1,2-dihydro-2,2-dimethyl-*s*-triazines (IV) bearing a phenylalkyl group on the *m*- or *p*-position of the 1-phenyl moiety was synthesized for enzymic evaluation with dihydrofolic reductase. The largest increment in binding was observed with a *m*-phenylbutyl group (XIV) which gave a fortyfold increment in binding; activity was not increased by substitution of one or two chlorines on the terminal phenyl group. The most active compound (XIV) was complexed one-third as well as the potent aminopterin, that is, XIV had 95 per cent of the free energy of binding shown by aminopterin. The potential use of XIV and related compounds for treatment of tumors with an impaired active-transport system for folic acid and aminopterin is discussed.

AMETHOPTERIN (I) has been useful for the treatment of leukemia in children for a number of years (1); unfortunately, only about 20% of the cases respond, and then only a temporary remission occurs. Amethopterin (I) and aminopterin (II) pass through a cell wall by active-transport (2–8), presumably by the same active-transport system used by a cell for folic acid. By use of tritiated amethopterin, Bertino (9) has been able to correlate the 20% of patients responding to amethopterin with ability of the patients' leukemic cells to take up the drug; the 70% showing no response to amethopterin had virtually no uptake of amethopterin in their leukemic cells.



I, R = CH_3
II, R = H



III

Citrovorum factor (5-formyl-tetrahydrofolic acid, CF) is used as an antidote for amethopterin toxicity, since CF can be utilized by a cell in place of tetrahydrofolic acid—the product of the reaction catalyzed by folic reductase which is blocked by amethopterin; presumably CF also enters the cell by the active-transport system used for the uptake of folic acid. It should therefore be theoretically possible to treat those leukemic patients with an impaired leukemic active-transport system with a potent folic reductase inhibitor that enters the cell by passive diffusion; by pre- and post-treatment with the antidotal CF, those cells with an impaired active-transport system would take up less CF and therefore not be protected as well by CF. Folic reductase inhibitors, such as 2,4-diaminopyrimidines of the pyrimethamine type (III), enter cells by passive diffusion (2, 10); presumably 4,6-diamino-1,2-dihydro-*s*-triazines of type VII (Table I) also enter cells by passive diffusion. If an inhibitor of the 4,6-diamino-1,2-dihydro-*s*-triazine type could be found that approached the potency of amethopterin for blocking folic reductase, it would be useful to investigate such a passive transport inhibitor of folic reductase in conjunction with CF treatment. Such an approach has been previously suggested (11); the results of a search for such potent inhibitors is the subject of this paper.

DISCUSSION

Both amethopterin (I) and aminopterin (II) have a strong pH dependence as inhibitors (12, 13), the optimum inhibition being close to pH 6 (13). The far less inhibition at pH 5.5 or 6.5 is dramatic (13), but the difference in inhibition between pH 6.5–7.4 is only nominal. Since few if any cells attain a pH of 6.0, a more reasonable comparison of inhibitors with aminopterin should be made at pH 6.5–7.4; the authors have selected pH 7.4 primarily because almost all previous work was at this pH. Aminopterin (II) (Table I) requires a concentration of 1×10^{-9} *M* for 50% inhibition in our system at pH 7.4.

The most potent compounds of the passive diffusion type observed on the pigeon liver enzyme are the 3-chloro (V) and 3,4-dichloro (VI)-1-phenyl-*s*-

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Previous paper: Baker, B. R., Lourens, G. J., and Jordaan, J. H., *J. Heterocyclic Chem.*, **4**, 39(1967). Previous paper on hydrophobic bonding to dihydrofolic reductase: Baker, B. R., and Ho, B.-T., *J. Pharm. Sci.*, **55**, 470(1966).

triazines (Table I); these two compounds are still $^1/_{10}$ to $^1/_{15}$ so effective as aminopterin (II), but about 7–9 times more effective than the parent 1-phenyl-*s*-triazine (IV). It was noted in earlier studies that substitution of benzyl (VIII) or phenethyl (IX) on the 3-position (14) or benzyl (X) at the 4-position (15) of the 1-phenyl group gave three- to sixfold enhanced binding. Therefore, a study of longer aralkyl groups at the 3-position was initiated first. The phenylpropyl (XIII), phenylbutyl (XIV), and phenylamyl (XV) were all more potent than the phenethyl (IX) group at the 3-position of the 1-phenyl moiety; optimum activity appeared to be at phenylbutyl (XIV), which was fortyfold more potent than the parent 1-phenyl-*s*-triazine (IV) and approached the potency of aminopterin (II) within a factor of three.

That there was some hydrophobic bonding by a 3-alkyl group was indicated by the 3-methyl (XVII) and 3-*n*-butyl (XII) groups, the latter being 3 times as effective as the parent 1-phenyl-*s*-triazine (IV). The presence of this hydrophobic bonding area near the 3-position (15) of the 1-phenyl group was also indicated by evaluation of the more polar anilinomethyl (XVI) and *n*-propoxy (XVIII) groups; the 3-anilinomethyl group (XVI) was about threefold less effective than the 3-phenethyl group (IX), while the 3-*n*-propoxy group (XVIII) was fiftyfold less effective than the 3-*n*-butyl group (XII).

Since phenyl binding to a receptor can sometimes be increased by halogenation (for example, VI *versus* IV) halogen derivatives of the 3-phenylbutyl group of XIV were investigated; no increased binding was observed by substitution of 3,4-dichloro (XIX), 2,4-dichloro (XX), 2,6-dichloro (XXI), or 2-chloro (XXII) substituents, but the binding was within a factor of two of the parent phenylbutyl group (XIV).

The 4-position of the 1-phenyl triazine was then investigated. 4-Phenethyl (XXIII) was as good an inhibitor as 3-phenethyl (IX) and the 2,4-dichlorophenylbutyl group at the 3-position (XX) and 4-position (XXIV) were equivalent. Therefore, XXV was synthesized to see if the ninefold increment in binding seen by introducing a 3-chloro group (V) on on the parent phenyl triazine (IV) would be additive with the 4-phenylbutyl side chain; that this additivity was not achieved can be seen by comparison of XXV and XXVI.

Although the most potent compound (XIV) in this study is less potent than aminopterin (II) by a factor of three, XIV is still sufficiently potent to warrant animal studies on tumor lines with an impaired active-transport system for amethopterin (I) (6). It can be estimated from the concentration of aminopterin required for 50% inhibition at pH 7.4 that $K_i \cong 1 \times 10^{-10}$ *M*, or a free energy of binding of 14 Kcal./mole. Since XIV only binds threefold less than aminopterin, the difference in binding energies is about 0.7 Kcal./mole. Therefore, at pH 7.4 XIV has about 95% of the free energy of binding of aminopterin.

Table I—Inhibition of Dihydrofolic Reductase[a] by

Compd.	R	mμ*M* Concn. for 50% Inhibition[b]
IV	H	110[c]
V	3-Cl	12[c]
VI	3,4-Cl_2	15[d]
VII	4-Cl	440[c]
VIII	3-$CH_2C_6H_5$	19[c]
IX	3-$(CH_2)_2C_6H_5$	24[c]
X	4-$CH_2C_6H_5$	40[e]
XI	4-C_4H_9-*n*	64[e]
XII	3-C_4H_9-*n*	30
XIII	3-$(CH_2)_3C_6H_5$	5.9
XIV	3-$(CH_2)_4C_6H_5$	2.7
XV	3-$(CH_2)_5C_6H_5$	7.1
XVI	3-$CH_2NHC_6H_5$	66
XVII	3-CH_3	78[e]
XVIII	3-OC_3H_7-*n*	1500
XIX	3-$(CH_2)_4C_6H_3Cl_2$-3,4	3.7
XX	3-$(CH_2)_4C_6H_3Cl_2$-2,4	4.9
XXI	3-$(CH_2)_4C_6H_2Cl_2$-2,6	4.4
XXII	3-$(CH_2)_4C_6H_4Cl$-2	6.1
XXIII	4-$(CH_2)_2C_6H_5$	17
XXIV	4-$(CH_2)_4C_6H_3Cl_2$-2,4	5.3
XXV	3-Cl-4-$(CH_2)_4C_6H_5$	9.3
XXVI	4-$(CH_2)_4C_6H_5$	10
II	(Aminopterin)	1.0[f], 0.86[g]

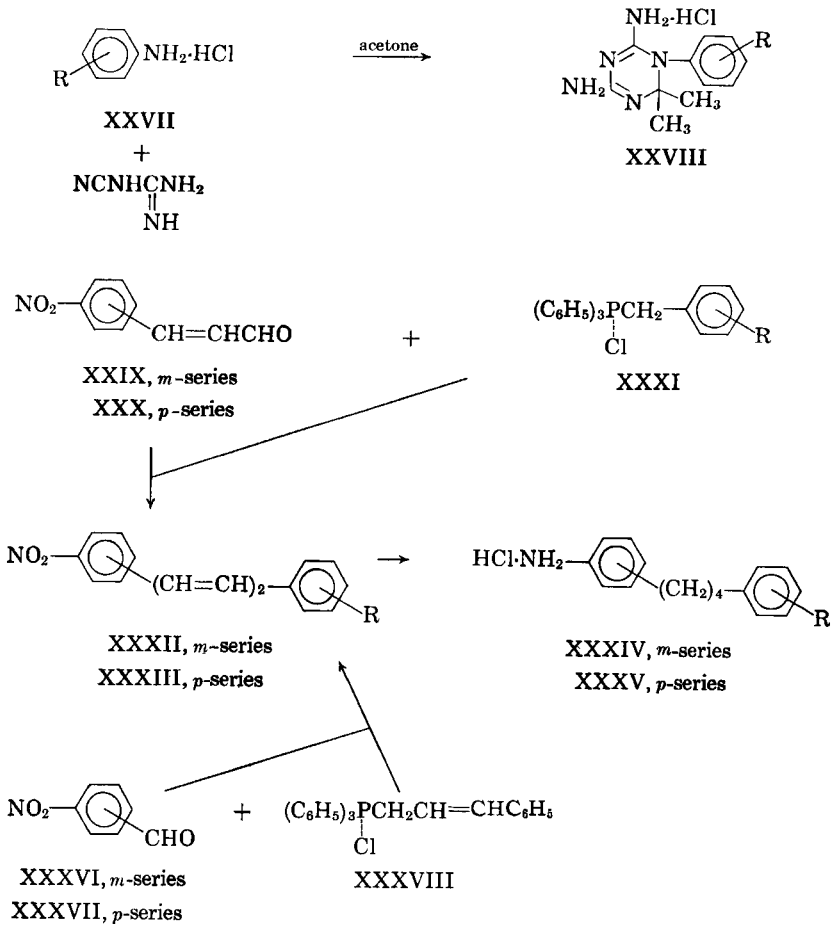
The technical assistance of Maureen Baker, Barbara Baine, and Karen Smith is acknowledged. [a] Dihydrofolic reductase was a 45–90% saturated ammonium sulfate fraction from pigeon liver prepared and assayed with 6 μ*M* dihydrofolate and 12 μ*M* TPNH at pH 7.4 as previously described (21). [b] Inhibitors were dissolved in 1 m*M* hydrochloric acid and dilutions were made with the same solvent. Solutions of some inhibitors at concentrations below 0.1 μ*M* lost appreciable inhibitory activity after 1–2 hr. at room temperature; since more concentrated solutions in the range of 0.1–1.0 m*M* were stable, the loss of inhibitor in highly dilute solution may have been due to adsorption on the glass. Consistent results were obtained by frequent dilution from a master solution of inhibitor at 0.31 m*M* so that solutions below 0.1 μ*M* were not aged for more than 30 min. [c] Data from *Reference 14*. [d] Data from *Reference 22*. [e] Data from *Reference 15*. [f] Data from *Reference 12*. [g] Data from *Reference 23*

CHEMISTRY

Synthesis

All of the substituted-1-phenyl-*s*-triazines (XXVIII) (Tables I and V) were synthesized by the three-component method of Modest (16), where the appropriate arylamine hydrochloride (XXVII) was condensed with cyanoguanidine in acetone. The synthesis of the requisite *m*- or *p*-aralkylanilines could be divided into three classes: (*a*) those where the alkyl was tetramethylene, (*b*) those where the alkyl was trimethylene or pentamethylene, and (*c*) miscellaneous.

The *m*-(XXXIV) and *p*-phenylbutylanilines (XXXV) were prepared by catalytic reduction of the appropriate 1,4-diphenylbutadiene (XXXII or XXXIII); the dienes were synthesized by the Wittig reaction. If the aralkyl group contained no other substituent, as in series *a*, the 1,4-diphenylbutadiene (XXXII*a* and XXXIII*a*) were conveniently synthesized by condensation of the Wittig reagent from cinnamyl chloride (XXXVIII) with the appropriate isomer of nitrobenzaldehyde (XXXVI or XXXVII). When the aralkyl had halogen substituents (series *b–e*), the appropriate benzyl chloride was converted to the Wittig reagent



Series:

a, R = H
b, R = 3,4-Cl_2
c, R = 2,4-Cl_2
d, R = 2,6-Cl_2
e, R = 2-Cl

Scheme I

(XXXI), then condensed with the *m*-isomer (XXIX) or *p*-isomer (XXX) of nitrocinnamaldehyde. In many cases, the 1,4-diphenylbutadienes melted over a wide range; since these crude materials gave good yields of 1,4-diphenylbutanes (XXIV, XXV) on hydrogenation, they were probably mixtures of *cis*- and *trans*-isomers. The yields obtained by the Wittig reaction were far better than those obtained by the older Meerwein and Perkin reactions. (Scheme I.)

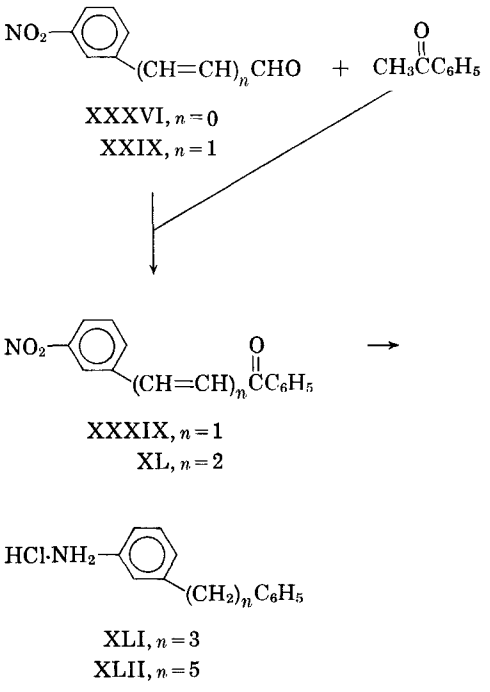
The second class of *m*-aralkylanilines containing an odd number (3 or 5) of methylene groups was synthesized in two steps. Condensation of *m*-nitrobenzaldehyde (XXXVI) or *m*-nitrocinnamaldehyde (XXIX) with acetophenone in aqueous ethanol containing sodium hydroxide gave the chalcones (XXXIX and XL) (17); catalytic reduction of the nitro, vinyl, and carbonyl groups with a palladium charcoal catalyst in glacial acetic acid containing ethanesulfonic acid (14) afforded the required *m*-aralkylanilines (XLI and XLII) in good yield. (Scheme II.)

In the miscellaneous class of 1-phenyl-*s*-triazines were the *m*-(*n*-butyl) (XII), the *p*-phenethyl (XXIII), *m*-anilinomethyl (XVI), and *m*-(*n*-propoxy) (XVIII) derivatives, which were synthesized from the known amines.

Methods

Melting points were determined in capillary tubes on a Mel-Temp block and those below 230° are corrected. Infrared spectra were determined in KBr pellet with a Perkin-Elmer 137B or 337 spectrophotometer. Ultraviolet spectra were determined with a Perkin-Elmer 202 spectrophotometer.

(3,4-Dichlorobenzyl)triphenylphosphonium Chloride (XXXIb)—*Method A*—A solution of 18 Gm. (0.07



Scheme II

mole) of triphenylphosphine and 13.7 Gm. (0.07 mole) of α,3,4-trichlorotoluene in 36 ml. of benzene was refluxed with magnetic stirring for 15 hr.; within 15 min., the product began to separate. The product was collected on a filter and washed with benzene; yield, 21.3 Gm. (66%) of white crystals, m.p. 305–306°. Recrystallization of a sample from absolute ethanol–ether gave white crystals, m.p. 309–310°. $\nu_{max.}$ 1580, 1550 (C=C); 1430, 1105 cm.$^{-1}$ (P-C). (See Table II for additional data.)

1 - (m - Nitrophenyl) - 4 - phenyl - 1,3 - butadiene (XXXIIa)—*Method B*—To a stirred solution of 10.1 Gm. (24 mmoles) of XXXVIII (18) and 3.02 Gm. (20 mmoles) of *m*-nitrobenzaldehyde (XXXVI) in 50 ml. of reagent methanol was added 1.62 Gm. (30 mmoles) of solid sodium methoxide; a yellow solid immediately separated. After being stirred for 20 hr. at ambient temperature protected from moisture, the mixture was filtered. The product was washed successively with methanol, water, and methanol; yield, 2.0 Gm. (40%), m.p. 146–147°. The combined filtrate and washings deposited another 1.46 Gm. (total 69%) of product, m.p. 146–147°, after about 18 hr. at −15°. An analytical sample of unchanged melting point was obtained by recrystallization from methanol as yellow crystals; $\nu_{max.}$ 1600, 1560 (C=C); 1505, 1340 cm.$^{-1}$ (NO_2); $\lambda_{max.}$ (EtOH): 331, 345 mμ (inflection). (See Table III for additional data.)

This compound has been prepared by the Meerwein reaction in 12% yield and melting points of 146° (19) and 145° (20) have been recorded.

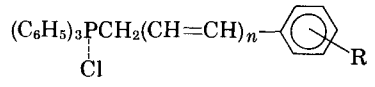
1 - (3,4 - Dichlorophenyl) - 4 - (m - nitrophenyl) - 1,3 - butadiene (XXXIIb)—*Method C*—Condensation of 0.885 Gm. (5 mmoles) of *m*-nitrocinnamaldehyde (XXIX) with 2.74 Gm. (6 mmoles) of XXXI*b* in methanol, as described under *Method B*, gave 0.900 Gm. (56%) of product, m.p. 112–124°, that was suitable for the next step. One crystallization from petroleum ether (b.p. 60–110°) gave 0.451 Gm. (28%) of yellow crystals, m.p. 129–136°. A second recrystallization afforded 0.315 Gm. (20%), m.p. 138–141°. $\nu_{max.}$ 1600, 1560 (C=C); 1520, 1345 cm.$^{-1}$ (NO_2); $\lambda_{max.}$ (EtOH) 338 mμ with shoulders at 325 and 353 mμ.

α-(m-Nitrobenzal)acetophenone (XXXIX)—To a stirred solution of 2.1 Gm. (0.055 mole) of sodium hydroxide in 20 ml. of water was added 10 ml. of ethanol and 5.2 Gm. (0.043 mole) of acetophenone. Then 6.5 Gm. (0.043 mole) of *m*-nitrobenzaldehyde was added in one portion; the temperature was maintained at 20–25° by occasional cooling. After being stirred an additional 2 hr., the thick mixture was kept at 4° for about 18 hr. The product was collected on a filter, then ground in a mortar with water; the solid was collected by filtration. The grinding with water in a mortar was repeated until the final washing was neutral. The product was then washed with cold ethanol; yield, 10.9 Gm. (100%), m.p. 143–144°. Recrystallization from ethanol afforded 7.81 Gm. (72%) of beige-colored crystals, m.p. 145–146°. $\lambda_{max.}$ (EtOH): 294 mμ; $\nu_{max.}$ 1650 (C=O); 1600, 1570 (C=C); 1525, 1340 cm.$^{-1}$ (NO_2).

Anal.—Calcd. for $C_{15}H_{11}NO_3$: C, 71.1; H, 4.38; N, 5.53. Found: C, 71.0; H, 4.16; N, 5.41.

α-(m-Nitrocinnamal)acetophenone (XL)—By condensation of 0.841 Gm. (7 mmoles) of acetophenone with 1.24 Gm. (7 mmoles) of *m*-nitrocinnamalde-

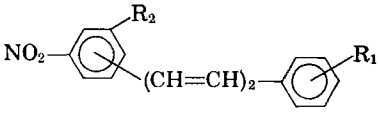
TABLE II—PHYSICAL CONSTANTS OF



Compd.[a]	n	R	M.p., °C.	% Yield	Anal. Calcd. C	Anal. Calcd. H	Anal. Found C	Anal. Found H
XXXI*b*	0	3,4-Cl_2	309–310[b]	66	65.6	4.40	65.3	4.54
XXXI*c*	0	2,4-Cl_2	248–249[c]	73				
XXXI*d*	0	2,6-Cl_2	257–262[d]	46	65.6	4.40	65.4	4.50
XXXI*e*	0	2-Cl	252–253[b]	71	70.9	5.00	70.9	5.06
XXXVIII	1	H	220–222[d,e]	92[d]				

[a] All compounds had infrared spectra in agreement with their assigned structures. [b] Recrystallized from absolute ethanol–ether. [c] Melting point of unrecrystallized product; Keaveney, W. P., and Hennessy, D. J., *J. Org. Chem.*, **27**, 1057(1962) have recorded m.p. 246–249° and a yield of 93%. [d] Recrystallized from methanol–ether. [e] McDonald and Campbell (18) have recorded m.p. 224–226° and a yield of 92%.

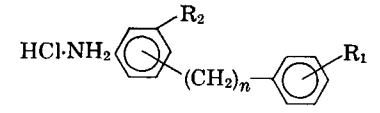
TABLE III—PHYSICAL CONSTANTS OF



Compd.[a]	Isomer	R_1	R_2	Method	% Yield	M. p., °C.	Anal. Calcd. C	Calcd. H	Calcd. N	Found C	Found H	Found N
XXXIIa	*meta*	H	H	*B*	69	146–147[b]	76.5	5.21	5.57	76.4	5.27	5.44
XXXIIb	*meta*	3,4-Cl_2	H	*C*	56[c]	112–124[c] 138–141[d]	60.0	3.46	4.38	60.3[d]	3.62	4.21
XXXIIc	*meta*	2,4-Cl_2	H	*C*	98[c]	110–121[c] 116–123[d]	60.0	3.46	4.38	59.9[d]	3.60	4.13
XXXIId	*meta*	2,6-Cl_2	H	*C*	90[c]	160–180[c] 187–188[e]	60.0	3.46	4.38	60.0[e]	3.61	4.19
XXXIIe	*meta*	2-Cl	H	*C*	31[c]	88– 93[c] 105–107[d]	67.2	4.23	4.90	67.1[d]	4.39	4.71
XXXIIIa	*para*	H	H	*B*	61[c]	128–165[c] 175–176[d,f]						
XXXIIIc	*para*	2,4-Cl_2	H	*C*	87[c]	155–158[c] 143–148[g]	60.0	3.46	4.38	60.3[g]	3.60	4.20
XLIII[h]	*para*	H	Cl	*B*	90[c]	93–138[c] 127–153[e]	67.2	4.23	4.90	67.1[e]	4.29	4.71

[a] All compounds had infrared and ultraviolet spectra in agreement with their assigned structure. [b] See *Method B* under *Experimental*. [c] Yield and melting point of mixed isomers suitable for further transformation. [d] Recrystallized from petroleum ether (b.p. 60–110°). [e] Recrystallized from ethyl acetate. [f] The lower melting isomer, m.p. 79–81°, was recovered from the filtrate in 27% yield. A m.p. of 181–182° and yield of 65% have been recorded for the higher melting isomer (18). [g] Recrystallized from 2-methoxyethanol. [h] The starting 2-chloro-4-nitrobenzaldehyde, m.p. 70–71°, was prepared by oxidation of 2-chloro-4-nitrotoluene in 24% over-all yield for two steps by the method used for synthesis of the isomeric 4-chloro-2-nitrobenzaldehyde; *cf.* Spalding, D. P., Moersch, G. W., Mosher, H. S., and Whitmore, F. C., *J. Am. Chem. Soc.*, **68,** 1596(1946). A lengthy alternate procedure has been previously described by Chardonnens, L., and Heinrich, P., *Helv. Chim. Acta*, **23,** 292 (1940) and Sharadamma, H. S., Kulkarni, S. N., Sattur, P. B., and Nargund, K. S., *J. Karnatak Univ.*, **1,** 61(1956); through *Chem. Abstr.*, **52,** 8083*i*(1958).

TABLE IV—PHYSICAL PROPERTIES OF



Compd.[a]	Isomer	*n*	R_1	R_2	Method	% Yield	M.p., °C.	Anal. Calcd. C	Calcd. H	Calcd. N	Found C	Found H	Found N
XXXIVa	*meta*	4	H	H	*D*	75	127–128[b]	73.4	7.70	5.34	73.2	7.77	5.26
XXXIVb	*meta*	4	3,4-Cl_2	H	*D*	98	Oil[c]						
XXXIVc	*meta*	4	2,4-Cl_2	H	*D*	78	129–130[b]	58.1	5.48	4.24	57.9	5.32	4.11
XXXIVd	*meta*	4	2,6-Cl_2	H	*D*	99	Oil[c]						
XXXIVe	*meta*	4	2-Cl	H	*D*	90	Amorphous[c]						
XXXVa	*para*	4	H	H	*D*	34	165–169[d]	73.4	7.70	5.34	73.4	7.93	5.14
XXXVc	*para*	4	2,4-Cl_2	H	*D*	88	155–156[e]	58.1	5.48	4.24	58.4	5.68	4.18
XLI	*meta*	3	H	H	*E*	89	118–119[b]	72.7	7.32	5.65	72.5	7.27	5.46
XLII	*meta*	5	H	H	*E*	88	137–138[b]	71.7[f]	8.13	4.93	71.9	8.11	5.02
XLIV	*para*	4	H	Cl	*D*	69	158–160[b]	64.9	6.46	4.73	65.0	6.58	4.87
XLV	*para*	2	H	H	*D*[g]	31	48– 49[h]						

[a] All compounds had infrared spectra in agreement with their assigned structures. [b] Recrystallized from isopropyl alcohol–ether. [c] The crude hydrochloride could not be crystallized and was therefore converted directly to the corresponding *s*-triazine in Table V. [d] The free base has been previously described (19). [e] Recrystallized from absolute ethanol–ether. [f] Hemihydrate. [g] The starting 4-nitrostilbene was obtained by the Meerwein method (19). [h] Free base from petroleum ether; a m.p. of 48° has been recorded by von Braun, J., Deutsch, H., and Koscielski, O., *Chem. Ber.*, **46,** 1511(1913). Considerable *p*-aminostilbene, m.p. 139–142°, insoluble in petroleum ether, was obtained as a by-product.

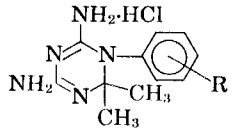
hyde, as described for the preparation of XXXIX, was obtained 1.43 Gm. (51%) of recrystallized product, m.p. 152–154°. For analysis a sample was recrystallized once more from ethanol to give crystals, m.p. 156–159°. $\lambda_{max.}$ (EtOH): 333 mμ; $\nu_{max.}$ 1640 (C=O); 1570 (C=C); 1500, 1335 cm.$^{-1}$ (NO_2).

Anal.—Calcd. for $C_{17}H_{13}NO_3$: C, 73.1; H, 4.69; N, 5.01. Found: C, 72.9; H, 4.65; N, 5.05.

Pfeiffer *et al.* (17) have recorded m.p. 157–158° for this compound.

1 - (p - Aminophenyl) - 4 - (2,4 - dichlorophenyl)-butane Hydrochloride (XXXVc)—*Method D*—A mixture of 2.2 Gm. (6.8 mmoles) of XXXIIIc, 200 ml. of absolute ethanol, and 100 mg. of platinum oxide catalyst was shaken with hydrogn at 2–3 Atm. until reduction was complete (3.5 hr.). The filtered solution was evaporated *in vacuo*. The oily residue was dissolved in 25 ml. of reagent ether, then the solution was treated with hydrogen chloride gas. The product was collected on a filter and washed

TABLE V—PHYSICAL CONSTANTS OF



Compd.[a]	R	% Yield	M.p., °C.	Anal. Calcd. C	Calcd. H	Calcd. N	Found C	Found H	Found N
XII	3-C_4H_9-*n*	39	179–180[b]	58.1	7.81	22.6	57.9	7.81	22.4
XIII	3-$(CH_2)_3C_6H_5$	85	181–182[c]	64.6	7.05	18.8	64.3	7.07	18.6
XIV	3-$(CH_2)_4C_6H_5$	67	175–176[c]	65.4	7.31	18.2	65.1	7.46	18.0
XV	3-$(CH_2)_5C_6H_5$	47	180–181[b]	66.1	7.56	17.5	66.0	7.50	17.3
XVI[d]	3-$CH_2NHC_6H_5 \cdot HCl$	32	181–183*d*[e,f]	53.4[f]	6.60	19.7	53.1	6.60	19.7
XVIII[d]	3-OC_3H_7-*n*	77	184–185[c]	53.9	7.11	22.5	53.7	7.12	22.5
XIX	3-$(CH_2)_4C_6H_3Cl_2$-3,4	18	173–174[g]	55.5	5.76	15.4	55.6	5.99	15.1
XX[h]	3-$(CH_2)_4C_6H_3Cl_2$-2,4	73	174–175[g]	55.5	5.76	15.4	55.3	5.93	15.1
XXI[i]	3-$(CH_2)_4C_6H_3Cl_2$-2,6	39	165–169[c]	52.3[i]	5.91	13.3	52.4	5.83	13.3
XXII	3-$(CH_2)_4C_6H_4Cl$-2	65	176–177[c]	60.0	6.48	16.7	60.3	6.66	16.8
XXIII	4-$(CH_2)_2C_6H_5$	78	200–202[c]	63.8	6.76	19.6	63.8	6.91	19.7
XXIV	4-$(CH_2)_4C_6H_3Cl_2$-2,4	68	181–182[c]	55.5	5.76	15.4	55.6	5.95	15.6
XXV	3-Cl-4-$(CH_2)_4C_6H_5$	85	191–192[c]	60.0	6.48	16.7	60.0	6.50	16.6
XXVI	4-$(CH_2)_4C_6H_5$	58	199–200[j]	65.4	7.31	18.2	65.7	7.51	17.9

[a] All compounds had ultraviolet and infrared spectra in agreement with their assigned structures. [b] Recrystallized from isopropyl alcohol–ether. [c] Recrystallized from absolute ethanol–ether. [d] The starting amine was prepared by catalytic reduction of the corresponding nitro compound. [f] Mono-methanolate. [g] Recrystallized from 0.01 *N* aqueous hydrochloric acid. [h] The ethanesulfonate salt was prepared by condensation of the appropriate aniline, cyanoguanidine with ethanesulfonic acid in 50% yield; white crystals from ethanol–ether, m.p. 174–175°. *Anal.*—Calcd. for $C_{21}H_{25}Cl_2N_5 \cdot C_2H_6SO_3H$: C, 52.3; H, 5.91; N, 13.3. Found: C, 52.0; H, 6.01; N, 13.4. [i] Ethanesulfonic acid salt. [j] Recrystallized from ethanol–petroleum ether (b.p. 30–60°).

with ether; yield, 1.97 Gm. (88%), m.p. 151–153°, that was suitable for further transformation. Recrystallization from absolute ethanol–ether gave nearly white crystals, m.p. 155–156°. $\nu_{max.}$ 2950, 2850, 2610, 1950 (NH, NH^+), 1610, 1575, 1505 $cm.^{-1}$ (NH, C=C). (See Table IV for additional data.)

m-Amino-1,3-diphenylpropane Hydrochloride (XLI)—*Method E*—A mixture of 2.53 Gm. (10 mmoles) of XXXIX, 200 ml. of glacial acetic acid, 2.20 Gm. (20 mmoles) of ethanesulfonic acid, and 0.2 Gm. of 5% palladium-charcoal catalyst was shaken with hydrogen at 2–3 Atm. for 17 hr., during which time reduction ceased. The filtered solution was evaporated *in vacuo*. The residual, oily, ethanesulfonate was dissolved in 40 ml. of water, then the solution was adjusted to pH 8–9 with 10% aqueous sodium hydroxide. The mixture was extracted with four 15-ml. portions of benzene. The combined extracts, dried with magnesium sulfate, were evaporated *in vacuo*. The residue was dissolved in 40 ml. of reagent ether, then the solution was treated with hydrogen chloride gas. The solvent was decanted from the separated gum which was crystallized by trituration with fresh ether; yield, 2.2 Gm. (89%), m.p. 117–118°, that was suitable for further transformation. Recrystallization of a sample from isopropyl alcohol–ether with the aid of decolorizing carbon gave white crystals, m.p. 118–119°; $\nu_{max.}$ 2950, 2850, 2600, 1960 (NH, NH^+); 1620, 1595, 1520 $cm.^{-1}$ (NH, C=C). (See Table IV for additional data.)

4,6-Diamino-1-{m-[4-(2,4-dichlorophenyl)-butyl]phenyl}-1,2-dihydro-2,2-dimethyl-s-triazine Hydrochloride (XX)—*Method F*—A solution of 727 mg. (2.2 mmoles) of XXXIV*c*, 198 mg (2.35 mmoles) of cyanoguanidine, and 3.5 ml. of acetone was refluxed with magnetic stirring for 18 hr.; after about 3 hr., the product began to separate. The product was collected and washed with acetone; yield, 730 mg. (73%), m.p. 180–182°. Recrystallization from 0.01 *N* aqueous hydrochloric acid gave white crystals, m.p. 178–189°. $\lambda_{max.}$ (H_2O): 243 mμ; $\nu_{max.}$ 3450, 3320, 3150 (NH); 1660, 1645, 1600, 1560, 1545, 1520 $cm.^{-1}$ (NH, C=NH^+, C=C, C=N). (See Table V for additional data on this and other compounds prepared by this method.)

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