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Arylaminoheterocycles. III. Arsenicals of Anilinotriazines

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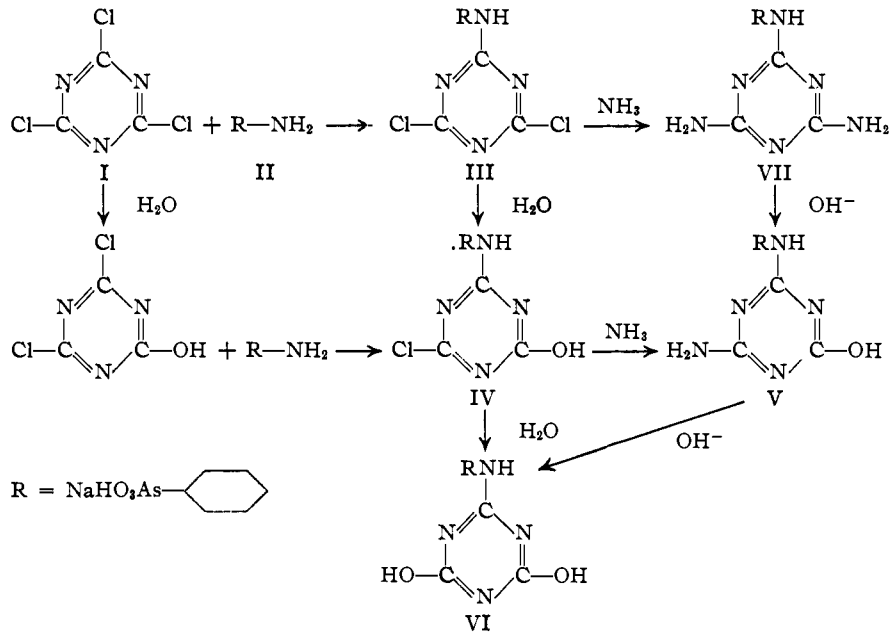
The encouraging results reported in the experimental and clinical study of 2-(4'-arsonoanilino)-4,6-diamino-*s*-triazine (VII) in the treatment of African trypanosomiasis[2] indicated that, like tryparsamide, the compound might have value in neurosyphilis. The possibility was also considered that related compounds might have activity equal to or greater than that of the parent compound.

The method of synthesis originally published[3] for such compounds involved the reaction of an aminobenzenearsonic acid with cyanuryl chloride in alkaline solution followed by reaction with aqueous ammonia under pressure and at elevated temperature. While this procedure gave the desired product in 15-g. lots using highly purified chemicals, an adaptation to larger lots was not successful. Several factors were found to be responsible for this failure. If the cyanuryl chloride was not freshly distilled, a decomposition product was present which also reacted, the resulting mixture not being amenable to the usual procedures of separation. Also, the hydrolysis of the reaction product of cyanuryl chloride (I) and *p*-arsanilic acid (II), 2-(4'-arsonoanilino)-4,6-dichloro-*s*-triazine (III), to the corresponding 4-chloro-6-hydroxy compound (IV) occurred readily. In addition, the pressure reaction with ammonia gave hydrolytic products (V and VI). The physical properties of these compounds are so nearly the same that separation was not feasible.

Because of the difficulties in overcoming the side reaction mentioned above, other synthetic procedures were considered: (1) the cyclization of cyano-, guanidino- and similar derivatives of *p*-arsanilic acid with dicyandiamide, guanidine, biguanidine, cyanogen halides and cyanamide, (2) the reaction of *p*-bromobenzenearsonic acid with melamine, and (3) the condensation of 2-chloro-4,6-diamino-*s*-triazine with *p*-arsanilic acid in the presence of alkaline condensing agents.

The various combinations possible under (1) were tried without success and the details of these preparations will be reported in a separate paper. These failures led to the trial of method (2) which proved to be impractical, since it gave only infinitesimal yields. As was expected, method (3) gave a mixture of 80% of product V and 20% of product VII. A modified technique of condensation was then found to give good results. The principles of the condensation have been reported elsewhere.[4,5] Nearly the theoretical yields of pure product were obtained when *p*-arsanilic acid and 2-chloro-4,6-diamino-*s*-triazine were condensed in the presence of 0.1 to 2 equivalents of acid.



2-Chloro-4,6-diamino-*s*-triazine was first prepared by the method of Liebig[6] and purified by recrystallization from water according to Lemoult.[7] Liebig's preparation gave several products which caused undesired side reactions (principally 2-amino-4,6-dichloro-*s*-triazine, some unreacted cyanuryl chloride and 2-amino-4-chloro-6-hydroxy-*s*-triazine) and recrystallization of one pound of product required 120 gallons of water, so that another method[8] was tried but without success. These methods were modified so as to give a minimum of impurities (less than 5% total). It was then found that digestion with hot water

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(2) Friedheim, *Schw. Med. Woch.*, **5**, 116 (1941).

(3) Friedheim, U. S. Patent 2,295,574 (1942).

(4) Banks, THIS JOURNAL, **66**, 1127 (1944).

(5) Banks, *ibid.*, **66**, 1131 (1944).

(6) Liebig, *Ann.*, **10**, 43 (1834).

(7) Lemoult, *Compt. rend.*, **125**, 822 (1897).

(8) Fierz-David and Matter, *J. Soc. Dyers Colourists*, 426 (1937).

TABLE I[a,b]

INTERMEDIATES

-s-triazine	M. p., °C., cor.	Analyses, % Nitrogen[c] Calcd.	Nitrogen[c] Found	Chlorine[d] Calcd.	Chlorine[d] Found
2-Chloro-4,6-diamino-	300	48.11	48.14	24.4	24.3
2-Chloro-4,6-di-(β-hydroxyethylamino)-	192–194	29.95	29.99	15.2	15.4
2-Chloro-4,6-di-(β-hydroxy-*n*-propylamino)-	195–198	26.77	26.75	13.6	13.9
2-Chloro-4,6-diglycino-	230–235	26.75	26.60	..	..

PENTAVALENT ARSENICALS[e]

	Formula	Arsenic[f] Calcd.	Arsenic[f] Found	Nitrogen[c] Calcd.	Nitrogen[c] Found
2-(4′-Arsonoanilino)-4,6-diamino-	$C_9H_{11}AsN_6O_3$	22.98	23.00	25.78	25.77
monohydrochloride[g]	$C_9H_{11}AsN_6O_3\cdot HCl$	20.70	20.40	23.06	23.20
disodium salt	$C_9H_9AsN_6Na_2O_3$	20.25	20.20	22.72	22.77
sesquisodium salt	$C_{18}H_{19}As_2N_{12}Na_3O_6$	20.86	20.83	23.41	23.34
2-(4′-Arsonoanilino)-4-amino-6-hydroxy-[3]	$C_9H_{10}AsN_5O_4$	22.91	23.02	21.42	21.16
disodium salt	$C_9H_8AsN_5Na_2O_4$	20.18	20.29	18.88	18.86
2-(4′-Arsonoanilino)-4,6-dihydroxy-[3]	$C_9H_9AsN_4O_5$	22.87	22.90	17.08	16.92
disodium salt	$C_9H_7AsN_4Na_2O_5$	20.14	20.02	15.06	14.70
2-(4′-Arsonoanilino)-4,6-di-(β-hydroxyethylamino)-	$C_{13}H_{19}AsN_6O_5$	18.10	18.10	20.30	20.02
sesquisodium salt	$C_{26}H_{35}As_2N_{12}Na_3O_{10}$	16.76	16.82	18.80	18.73
2-(4′-Arsonoanilino)-4,6-di-(β-hydroxy-*n*-propylamino)-	$C_{15}H_{23}AsN_6O_5$	16.95	17.02	19.02	18.70
sesquisodium salt	$C_{20}H_{43}As_2N_{12}Na_3O_{10}$	15.77	15.68	17.69	17.58
2-(4′-Arsonoanilino)-4,6-diglycino-	$C_{13}H_{15}AsN_6O_7$	16.94	16.98	19.01	18.94
tetrasodium salt	$C_{13}H_{11}AsN_6Na_4O_7$	14.13	14.13	15.86	15.78
2-(4′-Arsono-3′-hydroxyanilino)-4,6-diamino-[3]	$C_9H_{11}AsN_6O_4$	21.90	21.78	24.58	24.62
disodium salt	$C_9H_9AsN_6Na_2O_4$	19.41	19.50	21.75	21.78
2-(5′-Arsono-2′-hydroxyanilino)-4,6-diamino-[3]	$C_9H_{11}AsN_6O_4$	21.90	21.83	24.58	24.38
sesquisodium salt	$C_{18}H_{19}As_2N_{12}Na_3O_8$	19.97	20.10	22.40	22.43
2-(5′-Arsono-2′-β-hydroxyethoxyanilino)-4,6-diamino-	$C_{11}H_{15}AsN_6O_5$	19.41	19.55	21.75	21.43
disodium salt	$C_{11}H_{13}AsN_6Na_2O_5$	17.42	17.60	19.53	19.55
2-(4′-Arsonoanilino)-4,6-dichloro-[3]	$C_9H_7AsCl_2N_4O_3$	20.55	20.54	[h]	[h]
2-Amino-4,6-di-(4′-arsonoanilino)-[3]	$C_{15}H_{16}As_2N_6O_6$	28.48	28.43	...	...
tetrasodium salt	$C_{15}H_{12}AsN_6Na_4O_6$	24.44	24.68	13.70	13.58

TRIVALENT ARSENICALS[i]

	Formula	Arsenic Calcd.	Analyses,[f,i] % Found Total	Found Trivalent
2-(4′-Arsenosoanilino)-4,6-diamino-[k]	$C_9H_9AsN_6O$	25.66	25.60	25.63
monohydrate	$C_9H_9AsN_6O\cdot H_2O$	24.20	24.23	24.31
dihydrate	$C_9H_9AsN_6O\cdot 2H_2O$	22.84	22.85	22.88
2-(4′-Dichloroarsenosoanilino)-4,6-diamino-, hydrochloride	$C_9H_9AsCl_2N_6\cdot HCl$	19.57	19.62	19.68
2-[4′-Di-(carboxymethylenethio)-arsenosoanilino]-4,6-diamino-, disodium	$C_{13}H_{13}AsN_6Na_2O_4S_2$	14.92	14.91	14.91[l]
2-[4′-Di-(cysteinyl)-arsenosoanilino]-4,6-diamino-, dihydrochloride	$C_{15}H_{21}AsN_8O_4S_2\cdot 2HCl$	12.72	12.86	[m]
2-[4′-Di-(glutathionyl)-arsenosoanilino]-4,6-diamino-, tetrasodium	$C_{29}H_{37}AsN_{12}Na_4O_{12}S_2$	7.60	7.80	[m]
2-[4′-Di-(2″-carboxyphenylthio)-arsenoanilino]-4,6-diamino-	$C_{23}H_{19}AsN_6O_4S_2$	12.87	12.92	[m]
2-[4′-Arsenosoanilino)-4,6-di-(β-hydroxyethylamino)-	$C_{13}H_{17}AsN_6O_3$	19.63	19.78	19.52
2-(4′-Arsenosoanilino)-4,6-di-(β-hydroxyl-*n*-propylamino)-	$C_{15}H_{21}AsN_6O_3$	18.36	18.49	18.45
2-(4′-Arsenosoanilino)-4,6-diglycino-	$C_{13}H_{13}AsN_6O_5$	18.36	18.42	18.28
2-(5′-Arsenoso-2′-hydroxyanilino)-4,6-diamino-, hydrate[n]	$C_9H_{11}AsN_6O_3$	22.96	22.84	22.86
2-(5′-Arsenoso-2′-β-hydroxyethoxyanilino)-4,6-diamino-	$C_{11}H_{13}AsN_6O_3$	21.26	21.25	21.30
2-Amino-4,6-di-(4′-arsenosoanilino)-	$C_{15}H_{12}As_2N_6O_2$	32.57	32.68	Insol.

[a] All compounds are white. [b] Many thanks are due to Arthur Spang, Leonard Doub, Frances Hummel, Margaret McCarthy Ledyard and Clara Johnston for the adaptation of analytical procedures and the many analyses entailed in this work. [c] Nitrogen was determined by both the micro Dumas and the micro Kjeldahl methods. The micro Kjeldahl method, using potassium sulfate, copper sulfate and selenized granules gave the more consistent results. [d] Chlorine by micro combustion and gravimetric determination as silver chloride. [e] Representative salts are listed for the first compound only except where a specific salt was used in pharmacological testing. [f] All halogen-containing arsenicals were ashed by the peroxide method, all others by the method of Cislak and Hamilton, THIS JOURNAL, **52**, 638 (1930). The arsenic was determined volumetrically with potassium bromate using methyl orange indicator. [g] Calcd.: Cl, 9.79. Found: Cl, 9.67. [h] Calcd.: Cl, 19.45. Found: Cl, 19.18. [i] Representative hydrates and salts are given for only one compound. [j] Analyses of trivalent arsenicals were made in aqueous or dilute propylene glycol solutions, after the addi-

tion of a trace of hydrochloric acid, by titration with standard iodine solution with starch as the indicator. [k] This compound has also been prepared independently by Dr. Friedheim, private communication. Calcd.: N, 28.77. Found: N, 28.67. [l] The amount of iodine required was assumed to be twice the amount required for the oxidation of the arsenic. [m] The analysis was not practical due to side reactions. [n] The hydrate or arsenious acid was found to be the stable product.

TABLE II

TOXICITY AND TRYPANOCIDAL EFFECT

	Compound	LD_{50} Rats I. V., mg./kg.	Trypanocidal effect M. Th. D. mg./kg.	M. C. D. mg./kg.	Th. I.	C. I.
1	Atoxyl[a]	335	100	250	3.4	1.3
2	Tryparsamide[b]	4000	200	1000	20	4
	2-(4′-Arsonoanilino)-R-*s*-triazines					
3	4,6-Diamino-, sesquisodium salt	2000	30	60	67	33
4	4-Amino-6-hydroxy-, disodium salt	4500	50	150	90	30
5	4,6-Dihydroxy-, disodium salt	900	> 60	.....	.....	..
6	4,6-Di-(β-hydroxyethylamino)-, sesquisodium salt	2500+	> 80	.....	....	..
7	4,6-Di-(β-hydroxy-*n*-propylamino)-, sesquisodium salt	3700	>350	.....	...	..
8	4,6-Diglycino-, tetrasodium salt	3000+	>120	.....	...	..
	2-R-4,6-Diamino-*s*-triazines					
9	5′-Arsono-2′-hydroxyanilino-, sesquisodium salt	90	10	40	9	2.3
10	5′-Arsono-2′-β-hydroxyethoxyanilino-, disodium salt	115	>100	.....	...	..
11	4′-Arsono-3′-hydroxyanilino-, disodium salt	120	7.5	15	16	8
12	4′-Arsenosoanilino-[c]	17.5	0.10	0.50	175	35
13	4′-Dichloroarsenosoanilino-, hydrochloride	13.5	0.15	0.40	90	33
14	4′-Di-(carboxymethylenethio)-arsenosoanilino-, disodium	30	0.20	1.00	150	30
15	4′-Di-(cysteinyl)-arsenosoanilino-, dihydrochloride	35	0.15	1.00	233	35
16	4′-Di-(2″-carboxyphenylthio)-arsenosoanilino-	35	0.15	1.00	233	35
17	5′-Arsenoso-2′-hydroxyanilino-[c]	18	2.0	18	9	1

[a] Atoxyl is sodium *p*-aminobenzenearsonate. [b] Tryparsamide is sodium 4-arsonophenylglycinamide. [c] Administered in 50% propylene glycol solution.

had no effect on the product but decomposed all reactive impurities and, by filtering while hot, the impurities were removed with only slight loss of product. In this process the cyanuryl chloride need not be analytically pure.

The condensation of pure 2-chloro-4,6-diamino-*s*-triazine and *p*-arsanilic acid under varying conditions of acid and alkali was investigated. The optimum conditions were then used to condense other triazines with *p*-arsanilic acid and 2-chloro-4,6-diamino-*s*-triazine with other aminobenzenearsonic acids. These arsonic acids were then reduced to the corresponding oxides and several thio derivatives of the oxides prepared. The compounds are listed in Table I and the preliminary pharmacological data are given in Table II. The more active compounds will be reported in detail elsewhere.

It was found that, although the monosodium salts of the arsonic acids did form metastable solutions, the solubility of parent acids in water was so slight that precipitation occurred. The disodium salts were soluble, but highly alkaline. Equimolar quantities of the mono- and disodium salts crystallized in uniform crystals of the sesquisodium salt which proved to be highly soluble and less alkaline than the disodium salts, a condition desirable for intravenous injections.

Experimental

The reactions involved in the arsenicals derived from *p*-arsanilic acid and 2-chloro-4,6-diamino-*s*-triazine are typical and are described in detail. The other compounds in the tables were prepared by appropriate changes in either triazine, arsenical or both.

2-Chloro-4,6-diamino-*s*-triazine.—Cyanuryl chloride (600 g.) was dissolved in hot, dry acetone (360 ml.), filtered to remove any insoluble impurities and poured in a fine stream over crushed ice (2 kg.). The fine suspension and any unmelted ice was filtered off quickly and added to ammonia water (800 ml. of 28%) and ice (400 g.). The reaction mixture was stirred mechanically until the temperature reached 10°, then gaseous ammonia was bubbled through the suspension at such a rate as to cause an elevation in temperature to 40° in two hours. The temperature was maintained at 40° for one hour (water-bath) while additional ammonia gas was introduced, using a total of about 360 g. of the gas. The product was filtered off, digested with boiling water (1200 ml.) for thirty minutes and filtered while hot. This product was generally used as the wet cake but it could be dried at 100° and stored. The yield was 450 g. (over 90%). Material produced in this manner was a stable, white powder, melting above 300°, insoluble in acetone, benzene, alcohol and other organic solvents. Its solubility in water was about 1.2 g. per liter at 100° and 0.6 g. per liter at 20°.

2-Chloro-4,6-diglycino-*s*-triazine.—Cyanuryl chloride (18.5 g.) was suspended in 50 ml. of water at 0° and kept cold externally with an ice-bath. Glycine (22.5 g.) was dissolved in 60 ml. of 5 *N* sodium hydroxide. The alkaline solution was cooled and added dropwise to the cyanuryl chloride suspension with rapid stirring, keeping the temperature below 10°. After the glycine solution was added, the solution was stirred for one hour and then the temperature was increased gradually to 45–50°. The resulting brown solution was kept at this temperature for thirty minutes, cooled and acidified with glacial acetic acid, the cream-colored precipitate filtered off, washed with water and dried *in vacuo* over calcium chloride; yield 16 g. (61%).

2-Chloro-4,6-di-(β-hydroxyethylamino)-*s*-triazine and 2-Chloro-4,6-di-(β-hydroxy-*n*-propylamino-*s*-tri-

azine.—Cyanuryl chloride (9 g.) was dissolved in dry chloroform (110 ml.), the solution filtered and added dropwise to a stirred solution of the amino-alcohol (0.1 mole) in chloroform (14 ml.), keeping the temperature below 5°. After thirty minutes the temperature was allowed to rise gradually to room temperature and then warmed to 45–50° for one hour. The insoluble material was filtered off, and recrystallized from hot water. The white product obtained was dried *in vacuo* over calcium chloride to give 40–50% yields.

2-(4′-Arsonoanilino)-4,6-diamino-*s*-triazine.—Finely ground 2-chloro-4,6-diamino-*s*-triazine (43.8 g.) was suspended in one liter of hot water, hydrochloric acid (5 ml.), octyl alcohol (2 ml.) and 4-aminobenzenearsonic acid (65 g.) added and the whole refluxed for thirty minutes, giving a clear, metastable solution. The solution[9] was decolorized with charcoal and filtered while hot. The filtrate was made strongly acid with hydrochloric acid (100 ml.), precipitating 2-(4′-arsonoanilino)-4,6-diamino-*s*-triazine hydrochloride (the hydrochloride can be recrystallized from the dilute hydrochloric acid). The hydrochloride was filtered, suspended in hot water (1200 ml.), sodium hydroxide added until the solution was neutral to congo red paper and digested for thirty minutes to granulate the product. The product was filtered, washed free of chlorides and dried at 100°. The yield was 95 g. (96%). **Disodium salt.**—The arsonic acid (20 g.) was dissolved in water (100 ml.) with sodium hydroxide (2.1 equivalents), the solution filtered and the salt precipitated by the addition of five volumes of ethyl alcohol. The product was filtered and dried *in vacuo* over phosphorus pentoxide to give the trihydrate. The salt was dehydrated by heating four hours at 135° *in vacuo*. The anhydrous salt hydrated to the tetrahydrate on exposure to air. **Sesquisodium salt.**—The arsonic acid was suspended in water (100 ml.) and sodium hydroxide (1.5 equivalents) added, the solution filtered and precipitated with five volumes of ethyl alcohol. It was filtered and air-dried to give the tetrahydrate which could be partially dehydrated *in vacuo* to the trihydrate and made completely anhydrous by heating for several hours at 135° *in vacuo*.

2-(4′-Arsenosoanilino)-4,6-diamino-*s*-triazine.—2-(4′-Arsonoanilino)-4,6-diamino-*s*-triazine (45.2 g.) was suspended in concentrated hydrochloric acid (560 ml.). Sulfur dioxide (12 g.) was dissolved in concentrated hydrochloric acid (180 ml.) containing hydriodic acid (0.5 ml. of 47%). The sulfur dioxide solution was added slowly to the suspension with stirring. The resulting thick suspension was mixed thoroughly and set aside for forty-eight hours in the ice chest. This mixture was then poured over ice (1200 g.) and sodium carbonate (450 g.) dissolved in water (1200 ml.) added (defoamed with ether). Solid sodium bicarbonate was added until the suspension was neutral and the product filtered and washed by forming a slurry in water (600 ml.). The product was recrystallized from water (about 6 liters), keeping the acidity of the recrystallizing solution below *p*H 6 with acetic acid. The oxide crystallized slowly as fine needles to give a 35 g. (78%) yield of the dihydrate. Drying at room temperature over phosphorus pentoxide *in vacuo* gave the monohydrate. When heated *in vacuo* for sixteen hours at 135°, the anhydrous material was obtained.

2-(4′-Dichloroarsenosoanilino)-4,6-diamino-*s*-triazine.—The corresponding arsenoso compound (20 g.) was suspended in hot water (100 ml.) and concentrated hydrochloric acid added to give a clear solution from which the product crystallized on standing. The yield was 22.3 g. (96%).

Disodium 2-[4′-Di-(carboxymethylenethio]-arsenosoanilino-] - 4,6 - diamino - *s* - triazine.—2 - (4′ - Arsenosoanilino-)-4,6-diamino-*s*-triazine (400 g.) was suspended in water (1300 ml.), ammonia water (400 ml. of 28%) added and then thioglycolic acid (340 g.) stirred in slowly to give a pink solution with the evolution of heat. The solution was filtered and then acidified with hydrochloric acid to *p*H 4. The precipitate was filtered, washed thoroughly with water until all the pink color disappeared. The precipitate was suspended in water and two equivalents of sodium hydroxide added. The solution was filtered and 20 volumes of alcohol and 10 volumes of ether were added. After standing forty hours, the product was filtered and dried *in vacuo* over phosphorus pentoxide. The yield was 505 g. (82%).

2-[4′-Di-(R-thio)-arsenosoanilino]-4,6-diamino-*s*-triazine.—2-(4′-Arsenosoanilino)-4,6-diamino-*s*-triazine (0.01 mole) was dissolved in hot absolute alcohol (30 ml.) and the desired thio compound (0.021 mole) in ethanol (dissolved in hot 75–25 ethanol–water) was added. The resulting solution was heated for a few minutes and then cooled to give crystals of the desired product.

(9) When a tenfold quantity was prepared, the decolorization with charcoal and filtration were omitted, as the hydrochloride of the product started to precipitate during the reaction.

Pharmacology

Toxicity.—Fasting albino male rats (110–130 g.) were given intravenous injections of aqueous solutions of the compounds. A series of four or more graduated dose levels were administered, which resulted in from 0 to 100% mortality in two weeks, using a minimum of five animals per dose level. The test was repeated several times. A total of a hundred or more animals were used for each compound. The dose which was lethal for 50% of the animals (LD_{50}) was calculated from the mortality curve (Table III).

Trypanocidal Effect.—Albino male rats were inoculated intraperitoneally with about four million *Trypanosoma equiperdum* organisms suspended in 0.5 ml. of saline. The inoculum produced in forty-eight hours an infection of about fifty trypanosomes per one thousand red blood cells, at which time graduated doses of the arsenical were given to a series of infected animals. The blood of treated animals was examined for the presence of trypanosomes, by slide preparations and under the dark field microscope, forty-eight hours after the treatment and weekly thereafter for four weeks. Untreated animals died within ninety-six hours after inoculation. The minimal amount of drug which had entirely eliminated the trypanosomes from the peripheral blood stream at the forty-eight hour examination was designated as the minimal therapeutic dose (M. Th. D.) and the minimal amount which prevented a relapse of the infection for four weeks was designated the minimal curative dose (M. C. D.). The therapeutic index (Th. I.) is taken as the ratio LD_{50}/M. Th. D., and the curative index (C. I.) as the ratio, LD_{50}/M. C. D. (Table II).

The results of the toxicity and trypanocidal studies are summarized in Table II. Atoxyl (1) and tryparsamide (2) are included for purposes of comparison. The pentavalent arsenicals (3–11) have toxicities well within the expected range for arsonic acids, the derivatives of -*p*-arsanilic acid being the least toxic (3–8). Substitution in the benzene ring apparently increases the toxicity but does not necessarily destroy the trypanocidal activity (9, 10, 11). Of the simple triazine derivatives of *p*-arsanilic acid, the 4-amino-6-hydroxy (4) is the least toxic, followed by the 4,6-diamino (3), the 4,6-dihydroxy (5) being the most toxic.

However, only the first two have demonstrable trypanocidal activity. Substitution in the amino groups on the triazine ring does not cause any marked change in toxicity but does destroy the activity (6, 7, 8). Of the four pentavalent compounds showing trypanocidal activity (3, 4, 9, 11), the two having substituents in the benzene rings show only moderate activity (9, 11), the better of the two (11) having a curative index twice that of tryparsamide. The two *p*-arsanilic acid derivatives (3, 4) have unusually large curative indices, much larger than any experienced previously by us. Of these two compounds, the 4,6-diamino (3) compound is to be preferred because of ease of preparation and the lower effective dose level.

The trivalent compounds derived from (3) all have unusual activity (12–16) while the trivalent compound (17), having a benzene ring substituent, shows little activity. There is little to choose between these compounds. Of the compounds examined, several are being investigated more extensively to determine if their chronic toxicity will warrant clinical evaluation. The study of effectiveness in rabbit syphilis is also in progress. More detailed pharmacological reports will be made elsewhere.

Summary

1. The condensation of halotriazines with substituted anilines in acid solution has been applied to the synthesis of triazinylarsanilic acids.

2. A number of arsenicals of anilinotriazines have been prepared and studied in experimental trypanosomiasis. Several compounds have such unusual activities as to justify further study and clinical evaluation.

DETROIT, MICHIGAN RECEIVED APRIL 13, 1944

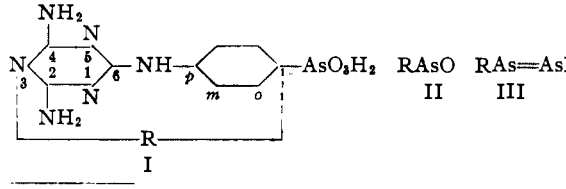
[CONTRIBUTION FROM THE LABORATORIES OF THE UNIVERSITY OF GENEVA, SWITZERLAND]

Trypanocidal and Spirochetocidal Arsenicals Derived from s-Triazine

BY E. A. H. FRIEDHEIM

This study is part of a search for therapeutic agents effective against the causative microorganisms of African sleeping sickness and syphilis. The work was guided by the consideration that it is especially important that such agents be (a) active in stages of these diseases in which the central nervous system is involved, and that (b) they should lack the toxic side effects on the central nervous system, particularly on the optic nerve, which are common to all pentavalent arsenicals used at the present time.

p-(2,4-Dichloro-s-triazinyl-6)-aminophenylarsonic acid, obtained readily from sodium arsanilate and cyanuric chloride, is a very toxic substance of no therapeutic value; however, it acquires considerable trypanocidal activity when at least one of its halogens is replaced by an amino group. In a series of fifteen arsonic acids of this type, the maximum trypanocidal effect combined with minimum toxicity, was found when both halogens were replaced by unsubstituted amino groups, *i. e.*, in *p*-(2,4-diamino-s-triazinyl-6)-aminophenylarsonic acid.[1] The biological effect of the replacement of the chlorine by other substituents is shown in Table II.



I

RAsO II RAs=AsR III

(1) Synonyms: 4-melaminylphenylarsonic acid, melarsen, triazinearsonic acid, 2,4-diamino-6(*p*-arsonoanilino)-s-triazine.

The compounds in this series have been prepared by taking advantage of the rule that in cyanuric chloride the halogens may be readily replaced by amino groups, the first chlorine at 0°, the second at room temperature and the third at 100–120°.[2]

The arsonic acids of this series are white, amorphous powders which remain unchanged when heated to 300°; they discolor and char without melting at higher temperatures ranging from 300–350°.

Reduction with sulfur dioxide, in the presence of hydriodic acid, leads to the corresponding arsine oxide (II) and reduction with hypophosphoric acid–hydriodic acid mixture to the corresponding arseno derivative (III).

I, II and III cure experimental trypanosomiasis and II cures, furthermore, experimental relapsing fever.

The therapeutic index for I, by parenteral administration, was found to be 30–50 for mice and about 10 for rabbits (*T. equiperdum, brucei, gambiense*).[3] This compound does not show the toxic effects of the classical pentavalent aromatic arsenicals on the central nervous system and does not cause in mice the symptom of "waltzing."[4] The therapeutic indices for compounds II and III, by oral administration in mice infected with *T. equiperdum*, were shown to be 20 and 25, respectively. The index for mapharsen in control experiments was found to be 1.3 (see Table III).

(2) H. E. Fierz-David and M. Matter, *J. Soc. Dyers Colourists*, **53**, 424 (1937).

(3) E. A. H. Friedheim, *Ann. Inst. Pasteur, Paris*, **65**, 108 (1940).

(4) E. A. H. Friedheim, *Schweiz. medizin. Wochenschrift*, **71**, 1101 (1941).