siphoned off and the precipitate was further dehydrated with ethanol. It was then filtered on a büchner funnel under dental dam to exclude moisture and dried over calcium chloride in a vacuum desiccator. The sample was then exposed to 65% R.H. for several days to displace adsorbed ethanol. Degradations of β-amylase-limit dextrin (*p*H, 6.5–6.6) were carried out in the same fashion.

β-Amylase-limit Dextrins.—The conversions were carried out on 4% solutions of the modified amylopectins at *p*H 4.7, unbuffered. After 45 hours, the solutions were heated to inactivate the enzyme and the dextrins were precipitated with 1.5 volumes of ethanol. The precipitated gums were washed several times with 60% ethanol, dissolved in water and reprecipitated in flocculent form in 8 volumes of ethanol. The dextrins were isolated and humidified in the same fashion as the modified amylopectins.

Acknowledgment.—We wish to thank Mr. J. C. Rankin for the periodate oxidations, Mrs. Marjorie Austin for some of the enzyme preparations and Mr. W. L. Deatherage for the iodine-sorption measurement.

PEORIA, ILLINOIS

[CONTRIBUTION FROM THE RESEARCH LABORATORIES, CHEMICAL DIVISION, MERCK & CO., INC.]

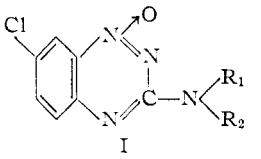
Benzotriazines. II. Synthesis of 3-Amino-7-halo-1,2,4-benzotriazine-1-oxides

BY F. J. WOLF, R. M. WILSON, JR., K. PFISTER, 3RD, AND M. TISHLER

RECEIVED MARCH 22, 1954

A series of 7-halo-1,2,4-benzotriazine-1-oxides bearing variously substituted 3-amino radicals has been prepared and examined for antimalarial activity.

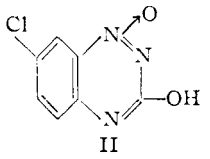
The antimalarial activity of substituted 3-amino-1,2,4-benzotriazines has been described.[1] Since the most active compounds were those with a halogen in position 7, and preliminary experiments indicated that activity was retained by replacement or modification of the amino group in position 3, a series of 7-halogen benzotriazines (I) containing substituted amino groups in this position was



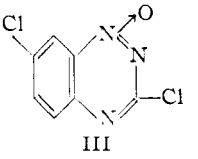
I

prepared and tested for antimalarial activity. In general, the resulting compounds were found to be somewhat less active than the unsubstituted amine. The most active compounds, in which R_1 is benzyl or methyl and R_2 is hydrogen, have about one-tenth the activity of the parent substance. Side chains giving enhanced activity in the plasmoquin and atebrin series were not efficacious.

Although various methods for the preparation of 1,2,4-benzotriazines have been described, none appeared suitable for the preparation of substituted 3-amino derivatives. It appeared probable that 3,7-dichloro-1,2,4-benzotriazine-1-oxide (III) could be readily aminated yielding the desired compounds. However, some difficulty was encountered in obtaining this dichloro compound. When refluxed with thionyl chloride, 3-hydroxy-1,2,4-benzotriazine-1-oxide (II) is recovered unchanged. On refluxing with phosphorus oxychloride or mixtures of phosphorus oxychloride and phosphorus pentachloride, a low yield (about 20%) of the desired 3-chloro compound is obtained. However, when phosphorus oxychloride and dimethylaniline[2] are used, an excellent yield of the dichloro compound is obtained.

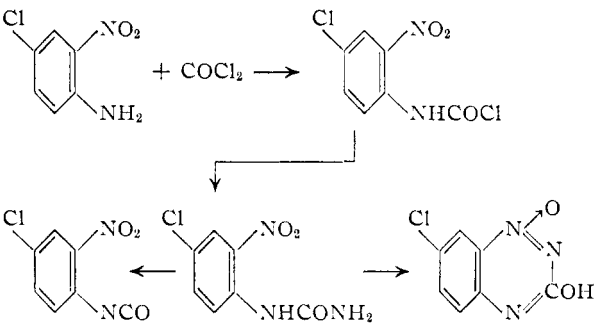


II



III

In order to provide sufficient quantities of the hydroxy compound, which had previously been prepared in 50% yield by diazotization of the corresponding amine, a new method of synthesis was developed utilizing conditions for ring closure similar to those described by Arndt[3] for the preparation of other benzotriazine compounds. The requisite ureide was obtained by treatment of the corresponding nitroaniline with phosgene and then ammonia. Depending on the conditions used, the ureide could be converted to the corresponding isocyanate or to the required 3-hydroxy-1,2,4-benzotriazine-1-oxide, ring closure being the favored reaction at high concentration of sodium hydroxide.



The chloro compound reacted readily with a variety of amines using temperatures of 60–70° for the amination. These amines are bright yellow in color and although lower melting than the parent compound were obtained in the crystalline state with one exception. The amines used were either available or prepared by standard literature procedures.

It is of considerable interest that when 7-halo-3-amino-1,2,4-benzotriazine-1-oxide or the desoxy compound was refluxed with benzylamine, 7-halo-

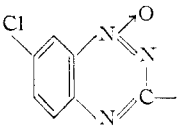
(1) For the previous publication in this series, see F. J. Wolf, K. Pfister, 3rd, R. M. Wilson, Jr., and C. A. Robinson, THIS JOURNAL, **76**, 3551 (1954).

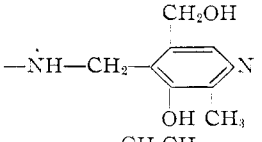
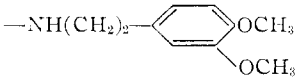
(2) J. Baddiley and A. Topham, *J. Chem. Soc.*, 678 (1944).

(3) F. Arndt, *Ber.*, **46**, 3522 (1913); F. Arndt and B. Eistert, *ibid.*, **60**, 2598 (1927); F. Arndt and T. Tschenscher, *ibid.*, **56**, 1988 (1923).

TABLE I

SUBSTITUTED 3-AMINO-7-CHLORO-1,2,4-BENZOTRIAZINE-1-OXIDES



3-Substituent formula	M.p., °C.	Solvent used for condensation	Yield, %	Analyses[a] Calculated C	H	N	Found C	H	N	Anti-material activity,[b] % in diet
$-NH(CH_2)_3CH_3$	170	CCl_4	70	52.3	5.1	..	52.6	5.3	..	0.5
$-NHCH(CH_3)(CH_2)_5CH_3$	89–90	C_2H_5OH	30	58.3	6.9	18.2	58.9	6.5	18.6	N.A.
$-NHCH(CH_3)(CH_2)_3N(C_2H_5)_2$	[c]	C_2H_5OH	35	56.9	7.2	20.7	57.3	7.1	20.6	0.12
$-NH(CH_2)_4CH(NH_2)COOH$	248	C_2H_5OH	45	48.0	5.0	..	48.1	4.9	..	N.A.
$-N(CH_2CH_2CH_3)_2$	105–106	CCl_4	69	55.6	6.1	20.0	55.9	6.2	19.7	0.12
$-N(CH_2CH_2)_2CH_2$	142	CCl_4	73.5	54.4	5.0	21.2	54.6	5.2	21.0	N.A.
$-N(CH_2CH_2)_2O$	175	CCl_4	63	49.5	4.2	21.0	49.6	4.0	21.0	N.A.
p-$NH-C_6H_4-COOH$	300	C_2H_5OH	75	53.1	2.9	..	52.9	3.0	..	N.A.
p-$NH-C_6H_4-(CH_2)_3COOH$	250–251	C_2H_5OH	74	56.9	4.1	15.8	57.0	4.4	15.6	N.A.
	213–214	C_2H_5OH	62	51.8	4.1	20.1	52.2	4.2	19.9	N.A.
$-NHCH(CH_2CH_3)(CH_2OH)$	138	CCl_4	65	49.2	4.9	20.9	48.9	4.8	20.9	0.06
$-NH-C(=NH)-NH_2$	282	C_2H_5OH	90	40.3	3.0	35.2	40.7	3.1	35.4	N.A.
$-NH(CH_2)_2-C_6H_5$	195–196	CCl_4	51	59.9	4.4	18.6	60.2	4.5	19.0	N.A.
	183–184	CCl_4	54	56.6	4.8	15.4	56.4	4.8	15.5	N.A.
$-NHCH_2-C_6H_5$	186	CCl_4	62	58.6	3.9	19.5	58.8	3.9	19.9	0.03
$-NHC_6H_4OCH_3$-p	211	C_2H_5OH	65	55.5	3.7	18.5	55.8	3.8	18.9	N.A.
p-$NHC_6H_4SO_2C_6H_4-NH_2$-p	293	C_2H_5OH	79	53.4	3.3	16.4	54.0	3.7	16.5	N.A.
$-NH-(CH_2)_{11}CH_3$	140	CCl_4	75	62.5	8.0	15.4	62.9	8.1	15.9	N.A.
$-NH-CH_2-CH_2OH$	186	CCl_4	60	44.9	3.8	23.3	45.0	4.0	23.5	N.A.
$-NH-CH_3$	236	C_2H_5OH	43	45.6	3.4	26.6	45.9	3.4	26.9	0.05
$-NH(CH_2)_2S(CH_2)_2N(C_2H_5)_2$	104–105	CCl_4	52	50.6	6.2	19.7	50.3	6.4	19.6	0.4
$-NH(CH_2)_3N(C_2H_5)_2$	79	CCl_4	58	54.3	6.5	22.6	54.8	6.6	22.5	0.2

[a] The authors are indebted to Mr. R. N. Boos and his associates for performing the microanalyses. [b] Percentage in diet required to give protection. N. A. (not active) indicates failure at 0.5%, the highest level tested. This is test E-3 for blood-induced Gallinaceum malaria in the chick (*cf.* F. Y. Wiselogle, "A Survey of Antimalarial Drugs," 1941–1945, Vol. I, 1946, p. 476). The authors are indebted to Dr. A. O. Seeler and Miss C. Malanga of the Merck Institute for Therapeutic Research for performing the antimalarial tests. [c] This compound was obtained as an oil, b.p. 130° at 3 μ.

3-benzylamino-1,2,4-benzotriazine was formed in good yield. This reaction did not proceed with common alkyl amines or with aryl amines.

The compounds prepared are presented in Table I.

Experimental

4-Chloro-2-nitrophenylurea.—Three hundred and thirty grams of 4-chloro-2-nitroaniline was suspended in six liters of benzene in a 12-liter three-necked flask equipped with a Glas-col heating mantle, stirrer, gas inlet tube and reflux condenser. Addition of phosgene and heating of the flask was begun. The flask was heated until the temperature reached 60°. The time required was two hours and 330 g. of phosgene had been added in this interval. The temperature of 60° was maintained until a total of 1165 g. of phosgene had been added. The total time required was five hours. (The theoretical amount of phosgene is 190 g., but it was found that an excess of phosgene equivalent to six to seven times the theoretical amount was necessary to obtain good yields.) The solution was allowed to stand overnight at room temperature and was then concentrated *in vacuo* until 1000 ml. of benzene had been removed. (The presence of phosgene in the benzene provides a serious disposal problem.) The flask containing the benzene solution was fitted for stirring and cooling and anhydrous ammonia gas was added at such a rate that the temperature remained below 20° and until no more yellow precipitate formed. The time required was three-quarters of an hour. The material was then filtered, washed with 1000 ml. of petroleum ether and dried. In order to remove any urea that formed, the precipitate was washed with five liters of hot water, filtered and dried. (Drying is unnecessary for use in the next step.) The yield was 330 g. (80%), m.p. 204–205°. A sample for analysis was recrystallized from isopropyl alcohol, m.p. 204–206°.

Anal. Calcd. for $C_7H_6N_3O_3Cl$: C, 39.0; H, 2.8; N, 19.5. Found: C, 38.8; H, 2.6; N, 19.5.

4-Chloro-2-nitrophenyl Isocyanate.—Seventy-one grams of 4-chloro-2-nitrophenylurea was suspended in 3600 ml. of a 10% solution of sodium hydroxide. The mixture was warmed on the steam-bath for 60 hours, at which time a clear red solution had formed. The solution was cooled, acidified with 500 ml. of concentrated hydrochloric acid and the resulting yellow flocculent precipitate collected and dried, weight 35 g., m.p. 85–86°, 53% yield. Recrystallization from ethanol yielded material of m.p. 87°.

Anal. Calcd. for $C_7H_3N_2O_3Cl$: N, 14.1. Found: N, 14.2.

7-Chloro-3-hydroxy-1,2,4-benzotriazine-1-oxide.—Three hundred and thirty grams of 4-chloro-2-nitrophenylurea was suspended in nine liters of 30% sodium hydroxide. The mixture was heated at 90–95° with good stirring for one-half hour and then made acid with glacial acetic acid. After cooling the crude 7-chloro-3-hydroxy-1,2,4-benzotriazine-1-oxide was collected. The hydroxy compound was further purified by dissolving in a 5% sodium hydroxide solution, filtering and reprecipitating with concentrated hydrochloric acid. The yellow precipitate was filtered off, washed with water and dried, weight 266 g., m.p. 225–226°, 88% yield. A sample recrystallized from Cellosolve had a melting point of 230–231° which is the same melting point as observed with a sample of 7-chloro-3-hydroxy-1,2,4-benzotriazine-1-oxide prepared by the diazotization of 7-chloro-3-amino-1,2,4-benzotriazine-1-oxide. A mixed melting point of the two samples showed no depression.

3,7-Dichloro-1,2,4-benzotriazine-1-oxide.—Twenty-five grams of 7-chloro-3-hydroxy-1,2,4-benzotriazine-1-oxide was dissolved in 50 ml. of dimethylaniline (free from mono-) and 100 ml. of phosphorus oxychloride and heated to reflux for three-quarters of an hour. The mixture was cooled and quenched in 1500 ml. of an ice–water mixture. The precipitate that formed was slurried with 300 ml. of hot 8 *N* hydrochloric acid to remove any methylanilino compound and then washed with water. The insoluble material consisting of crude 3,7-dichloro-1,2,4-benzotriazine-1-oxide weighed 20.7 g., m.p. 149–152°, 75.4% yield. Neutralizing the acid filtrate gave 4.0 g. of 7-chloro-3-(N-methylanilino)-1,2,4-benzotriazine-1-oxide, m.p. 155° (10% yield).

When dimethylaniline not especially purified was used, 25 g. of the hydroxy compound yielded 8.3 g. of the dichloro compound (32%) and 8.2 g. of methylanilino compound (30%). A sample of 3,7-dichloro-1,2,4-benzotriazine-1-oxide was recrystallized from alcohol, m.p. 153–154°.

Anal. Calcd. for $C_7H_3N_3Cl_2O$: C, 38.9; H, 1.4; N, 19.5. Found: C, 39.3; H, 1.5; N, 20.0.

7-Chloro-3-methoxy-1,2,4-benzotriazine-1-oxide.—To a solution consisting of 6.5 g. of 3,7-dichloro-1,2,4-benzotriazine-1-oxide in 50 ml. of absolute methanol was added 0.7 g. of sodium. The solution was then heated to reflux for 18 hours; on cooling, yellow-white crystals separated. The precipitate was recrystallized from methanol, weight 3.1 g., m.p. 155–156°, 50% yield. A sample was recrystallized from methanol, m.p. 157°.

Anal. Calcd. for $C_8H_6N_3O_2Cl$: C, 45.4; H, 2.9; N, 19.9. Found: C, 45.8; H, 3.2; N, 20.5.

7-Chloro-3-butylamino-1,2,4-benzotriazine-1-oxide.—Replacement of the 3-chloro group by a substituted amine is illustrated by this and the following example which are typical of the methods used for the preparation of compounds in Table I.

A mixture of 10 g. of 3,7-dichloro-1,2,4-benzotriazine-1-oxide and 6.8 g. of *n*-butylamine in 100 ml. of carbon tetrachloride was heated to reflux for 18 hours. At the end of this time the mixture was filtered to remove *n*-butylamine hydrochloride and the filtrate concentrated to dryness *in vacuo*. The residue was dissolved in the minimum quantity of hot absolute ethanol, treated with activated carbon and filtered. On cooling the filtrate, yellow needles, m.p. 170°, were formed, weight 3.7 g., 70% yield.

7-Chloro-3-(*p*-methoxyphenylamino)-1,2,4-benzotriazine 1-oxide.—A solution consisting of 6.6 g. of 3,7-dichloro-1,2,-4-benzotriazine-1-oxide and 7.2 g. of *p*-anisidine in 150 ml. of absolute alcohol was heated to reflux for 18 hours. Upon cooling the alcohol solution, a dark red precipitate was obtained. This material was filtered and recrystallized from Cellosolve. A yield of 6.0 g. (65%), m.p. 210–211°, was obtained.

7-Bromo-3-benzylamino-1,2,4-benzotriazine.—A mixture of 9.7 g. of 7-bromo-3-amino-1,2,4-benzotriazine-1-oxide and 75 ml. of benzylamine was heated to reflux for eight hours. The solution was cooled and poured into 500 ml. of methanol. The resulting precipitate was filtered off and washed with 100 ml. of methanol. The crude product was recrystallized from Cellosolve. A yield of 5.0 g. (40%), m.p. 172–173°, was obtained. Recrystallization from methanol yielded material, m.p. 173–174°.

Anal. Calcd. for $C_{14}H_{11}N_4Br$: C, 53.4; H, 3.5; N, 17.9. Found: C, 53.8; H, 3.7; N, 18.2.

7-Chloro-3-benzylamino-1,2,4-benzotriazine.—This compound was readily obtained by refluxing 7-chloro-3-amino-1,2,4-benzotriazine with benzylamine. The light yellow product was obtained in 78% yield, m.p. 175°.

Anal. Calcd. for $C_{14}H_{11}N_4Cl$: C, 62.2; H, 4.1; N, 20.7. Found: C, 61.9; H, 4.1; N, 21.3.

RAHWAY, N. J.

NOTES

L-Pyrrolidonecarboxylic Acid

BY A. F. BEECHAM

RECEIVED MARCH 31, 1954

King and McMillan,[1] after thermal dehydration of glutamic acid, isolated two compounds, pyrrolidonecarboxylic acid, m.p. 180–183°, and "3,6-diketopiperazine-2,5-dipropionic acid," m.p. 160–161°. This work has been repeated and the two substances examined in more detail. The acid m.p. 180–183° is found to be optically inactive; the acid m.p. 160–161° has $[\alpha]D$ −11.7° (in water) and is shown to be the optically active form of the first by the following considerations: (1) The infrared absorption of the tributylamine salts of the two compounds in chloroform solution is identical. Several well-defined absorption bands are assignable to the salts rather than to tributylamine or the solvent. (2) The ultraviolet absorption curves of the two acids in aqueous solution are featureless but identical. (3) The two acids have identical R_f values on paper chromatograms run with three different solvent systems. (4) The acid m.p. 160–161° can be converted in good yield to the acid m.p. 180–183° by heating at 190°.

The melting point and the specific rotation found for the acid m.p. 160–161° agree with the constants

(1) J. A. King and F. H. McMillan, THIS JOURNAL, **74**, 2859 (1952).