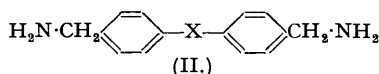
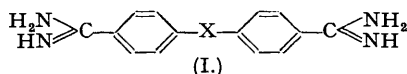


275. Benzylamine Analogues of Chemotherapeutic Diamidines.

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To help trace the origins of the outstanding chemotherapeutic properties of aromatic diamidines of type (I), the simpler bases of type (II) were synthesised for the first time.

THE researches of Ewins and his colleagues (cf. Ashley, Barber, Ewins, Newbery, and Self, *J.*, 1942, 103) established that aromatic diamidines of type (I), especially *c*, *d*, *e*, and *f*, have outstanding antiparasitic properties against protozoa. It is not known (i) whether the anti-protozoal activity of these substances is derived largely from their having two well-ionized and mutually independent basic groups held at a distance from one another, or (ii) whether such activity depends specifically on the amidine group which may conceivably function by assisting adsorption, through the resonance of its ion, or by interfering with a similarly constituted metabolite in the parasite.



(a) X absent; (b) X = CH₂; (c) X = O;
(d) X = O·[CH₂]₃·O; (e) X = O·[CH₂]₆·O; (f) X = CH=CH.

At this stage we propose only to examine hypothesis (i). This cannot be tested simply by replacing each amidine group by an amino-group because the basic strength of the latter would be enormously lowered through interaction with the benzene ring (cf. aniline and methylamine, Table I), leading to an almost complete absence of kations in neutral solution. If, however, the amino-group is insulated from the aromatic ring by a methylene group, the base-weakening resonance is repressed and a high degree of ionization results (cf. benzylamine and methylamine, Table I). That the ionization of amidines is little affected by the presence of an aromatic substituent is now shown for the first time (Table I).

TABLE I.

Ionisation of certain bases under physiological conditions.

Compound.	pK _a , 20° (water).	Source of value.	% ionized pH 7.
Acetamidine	12.52	A	100
Benzamidine	11.6 (M/10)	B	100
Methylamine	10.76	C	100
Aniline	4.65	D	0.5
Benzylamine	9.4	E	99.5

- A. Schwarzenbach and Lutz, *Helv. Chim. Acta*, 1940, **23**, 1162.
 B. Determined potentiometrically by Mr. R. Goldacre (for details of method, see Albert and Goldacre, *J.*, 1946, 706).
 C. Britton and Williams, *J.*, 1935, 796.
 D. Hall and Sprinkle, *J. Amer. Chem. Soc.*, 1932, **54**, 3469.
 E. Carothers, Bickford, and Hurwitz, *ibid.*, 1927, **49**, 2908.

The preparation of the bisbenzylamine compounds (II, *a-f*) was accordingly undertaken to permit of biological comparison with the corresponding diamidines (I), in particular with *c* (Phenamidine), *d* (Propamidine), *e* (Pentamidine), and *f* (Stilbamidine).

The corresponding dinitriles were hydrogenated by the method of Schwoegler and Adkins (*J. Amer. Chem. Soc.*, 1939, **61**, 3499) for aliphatic amines, methanolic ammonia being used in the presence of Raney-nickel catalyst at 110° for 20 minutes under 1200 lb./sq. in. pressure. In this way were prepared *pp'*-bisaminomethyldiphenyl (II, *a*), *pp'*-bisaminomethyldiphenylmethane (II, *b*), characterized as its *dibenzoyl* derivative, *pp'*-bisaminomethyldiphenyl ether (II, *c*), 1 : 3-(*pp'*-bisaminomethyldiphenoxy)propane (II, *d*), 1 : 5-(*pp'*-bisaminomethyldiphenoxy)pentane (II, *e*), and *pp'*-bisaminomethylstilbene (II, *f*).

Before attempting the reduction of 4 : 4'-dicyanostilbene, it was confirmed that these conditions were not severe enough to reduce the double bond in *trans*-4 : 4'-diaminostilbene. The latter was made by reducing the mixture of dinitro- and dinitroso-stilbenes formed by the violent reaction of *p*-nitrotoluene and methanolic sodium hydroxide (Fischer and Hepp, *Ber.*, 1893, **26**, 2232). The mechanism of this reaction was investigated by Green, Davies, and Horsfall (*J.*, 1907, **91**, 2076), but no yields were given. As insufficient details exist in the literature for the safe and successful conduct of this reaction for preparative purposes, a detailed account is given in the experimental section.

The products (II, *a-f*) are white, water-insoluble solids which absorb carbon dioxide rapidly from the air but are stable in the form of their white, readily crystallising salts, of which the diacetates are more soluble than the dihydrochlorides and are to be preferred for biological work. That these substances possess the predicted basic strength may be inferred from the example quoted in Table II. This table also shows that, both here and in the diamidines, the two basic groups are too far apart for the ionization of the first to exert an adverse Coulombic effect on that of the second, such as is seen in trimethylenediamine (pK_a 9.8 and 7.0).

TABLE II.
Ionisation of certain diacid bases.

Compound.	pK_a values, 20° (50% ethanol).	Source of value.	Approx. % ionised in water at pH 7.
"Propamidine" (I, <i>d</i>)	11.6 & 10.8 (M/300)	} B (see Table I)	100 & 100
<i>pp'</i> -Bisaminomethyldiphenyl- methane (II, <i>b</i>)	9.28 & 8.10 (M/160)		100 & 97

Von Braun (*Ber.*, 1937, **70**, 979) claimed to have produced (II, *a*) by the action of liquid ammonia on *pp'*-bisbromomethyldiphenyl; it is possible that his material was contaminated by secondary amines.

Note added in Proof.—Compounds (I, *c-f*) and (II, *a-f*) were kindly compared by Mr. L. G. Goodwin in experimental animal tests against *Trypanosoma rhodesiense*, *T. congolense*, *Plasmodium gallinaceum*, *Leishmania donovani*, and *Entamoeba histolytica*. No activity was shown by any Type (II) compound. We are also grateful to Prof. S. D. Rubbo for testing the same series, *in vitro*, against *Cl. welchii*, *Strept. pyog.*, *Staph. aur.*, *B. coli*, and *B. Proteus*. Only (I, *d* and *e*) showed outstanding activity.

Thus, unless it can be shown that these organisms possess an enzyme specifically destroying Type (II) compounds, hypothesis (i) is no longer tenable.

EXPERIMENTAL.

[The microanalysis of bisaminomethyldiphenyl and its hydrochloride were carried out by one of us (J. A. M.); the remainder were done by Miss J. Fildes, who is hereby thanked.]

General.—The dinitrile (15 g.), Raney-nickel catalyst (*ca.* 5 ml. of sludge), and methanolic ammonia saturated at 0° (150 ml.) were placed in a chilled-steel hydrogenation bomb (unlined) with two ½ in. steel balls. These proportions allow about 7 mols. of ammonia, per cyano-group. Hydrogen was admitted at an initial pressure of 1300 lb./sq. in., and maintained at about 1200 lb. whilst the bomb was heated to 110° in 20 minutes with shaking. Uptake of hydrogen began at about 70°. It was rapid and approximately that required by theory. The autoclave was allowed to cool, and then chilled in ice. It was opened, and the contents filtered quickly with the assistance of some methanolic ammonia, in which the amines dissolve much better than in methanol. Carbon dioxide was excluded as much as possible from this and subsequent operations. The solvent was recovered by using a bath at 140° and applying a vacuum towards the end. The amines (crude yields about 90%) solidified on cooling and sometimes contained colloidal nickel hydroxide which caused foaming when the solids were distilled.

pp'-Bisaminomethyldiphenyl (II, *a*).—This was purified by distillation (b. p. 180°/0.5 mm.) and recrystallisation from 10 parts of toluene. The yield of pure material was 80%. A slightly under-

hydrogenated batch supersaturated badly. The substance is soluble in hot alcohol, benzene, and acetate, and in cold pyridine (Found : C, 79.5; H, 7.5; N, 13.3. Calc. for $C_{14}H_{16}N_2$: C, 79.2; H, N, 13.2%). The compound and its derivatives had the following m. ps. (values of v. Braun, *loc. cit.*, parenthesis) : base, 144—145° (135°); picrate, 232—235° (decomp.) (222°); dibenzoyl derivative, 249—250° (243°); diacetyl derivative, 281—283° (272°). The *dihydrochloride* was formed by crystallisation from hot dilute hydrochloric acid. It is soluble in about 100 parts of cold water and readily precipitated by extra chloride ions (Found : C, 59.15; H, 6.3; Cl, 24.7. $C_{14}H_{16}N_2Cl_2$ requires C, 58.95; H, 6.4; Cl, 24.9%). The crystals developed a blue tint on exposure to sunlight.

pp'-*Bisaminomethyl*diphenylmethane (II, b).—This was purified by distillation at 0.5 mm., followed by dissolution in 5 parts of boiling benzene and treatment with 5 parts of light petroleum (b. p. 60—90°). The white crystals, m. p. 90° (80% yield), were dried in a vacuum over shredded paraffin. The compound is very soluble in alcohol without gradient, sparingly soluble in ether (Found : C, 79.4; H, 7.9; N, 12.2. $C_{18}H_{18}N_2$ requires C, 79.6; H, 8.0; N, 12.4%). The dihydrochloride (m. p. over 350°) is exceptional in this series in that it is difficult to precipitate with chloride ions; it is almost insoluble in alcohol and unaffected by light. The *dibenzoyl* derivative was prepared with benzoyl chloride in 10 volumes of pyridine (yield 90%) as white crystals from chlorobenzene, m. p. 224° (230° corr.), sparingly soluble in benzene, acetone, and alcohol (Found : N, 6.4. $C_{23}H_{26}O_2N_2$ requires N, 6.45%).

pp'-*Bisaminomethyl*diphenyl ether (II, c).—This was distilled (b. p. 186°/0.3 mm.), giving crystals (70% recovery) which began to melt at 56° and were completely molten at 77° (? double m. p.). The product was readily soluble in ethyl and amyl alcohols and in boiling toluene or acetone with gradient; only slightly soluble in ether and in light petroleum (Found : C, 73.0; H, 7.1; N, 12.2. $C_{14}H_{16}ON_2$ requires C, 73.6; H, 7.1; N, 12.3%). The dihydrochloride does not melt below 360°. The diacetate (m. p. 156°, anhydrous) was formed by dissolving the base in 8*N*-acetic acid at 50°, precipitating with acetone, and drying at 110°. It was readily soluble in cold water and hot ethanol.

1 : 3-(*pp'*-*Bisaminomethyl*diphenoxy)propane (II, d).—This was purified by distillation (b. p. 242°/0.5 mm.) and then recrystallised from alcohol, m. p. 90—91° (70% yield). It is sparingly soluble in boiling benzene; moderately so in boiling acetone. The dihydrochloride is sparingly soluble in cold water and readily soluble in hot water; it sinters at *ca.* 290° but does not melt below 360°. The diacetate, m. p. 178°, prepared as above, is readily soluble in cold water, moderately in boiling alcohol, and insoluble in acetone (Found : C, 62.0; H, 7.4; N, 6.85. $C_{17}H_{22}O_2N_2$, $2C_2H_4O_2$ requires C, 62.0; H, 7.4; N, 6.9%).

1 : 5-(*pp'*-*Bisaminomethyl*diphenoxy)pentane (II, e).—This was purified through the acetate without previous distillation. The base (70% yield), m. p. 74—75°, from aqueous ethanol, is readily soluble in pyridine and in methanol and has good gradients in toluene and chlorobenzene. It is very slightly soluble in ether and in light petroleum. The dihydrochloride is soluble in cold water and insoluble in ethanol and acetone. It sinters at *ca.* 270° but does not melt below 360°. The diacetate, m. p. 161°, is very soluble in water, moderately in cold and readily in boiling ethanol, and insoluble in acetone (Found : C, 63.4; H, 7.8; N, 6.4. $C_{19}H_{26}O_2N_2$, $2C_2H_4O_2$ requires C, 63.55; H, 7.9; N, 6.45%).

pp'-*Bisaminomethyl*stilbene (II, f).—This compound was purified by recrystallisation first from chlorobenzene and then from ethanol, giving crystals, m. p. 210—211° (decomp.) on rapid heating (yield 75%). They are soluble in about 27 parts of boiling ethanol, moderately soluble in boiling benzene or pyridine (with gradient), and almost insoluble in acetone, ether, or light petroleum (Found : C, 80.6; H, 7.6; N, 11.7. $C_{14}H_{18}N_2$ requires C, 80.6; H, 7.6; N, 11.8%). The dihydrochloride is moderately soluble in boiling water (with gradient) and insoluble in alcohol or acetone. The diacetate, m. p. 206—207°, solidifies at 210° and melts again at 255° (approx.) and is readily soluble in water, slightly soluble in boiling alcohol, and insoluble in acetone.

trans-pp'-*Diaminostilbene*.—A three-neck flask (capacity 1 l.) was fitted with an efficient mechanical stirrer, a 24-in. water-cooled bulb condenser, and a cork containing a thermometer, the bulb of which was pushed down below the 250 ml. level. The operation was conducted under a good hood.

Sodium hydroxide sticks (125 g.) were broken into coarse grit and refluxed in the flask with methanol (500 ml.) until dissolved (about 90 minutes), heat being supplied by a glycerol-bath at 100°. The flame was extinguished, and the thermometer momentarily withdrawn while molten *p*-nitrotoluene (50 g.) was rapidly added. After 30 seconds a violent reaction set in, the temperature rising to 105° and falling back to 90° when the condensed methanol returned to the flask. The flask contents became deep red. The glycerol-bath was then heated for ½ hour to keep the flask contents at 100°. The thermometer was then withdrawn, and a condenser, arranged for distillation, inserted. Concentrated hydrochloric acid (1 equiv.; 300 ml. of 39% w/v) was added down the upright condenser during 15 minutes with rapid stirring. The bath was then maintained at 120° until 400 ml. of methanol, altogether, had been collected. The product consisted of a mixture of dinitrodibenzyl, dinitrostilbene, and (mainly) dinitrostilbene. These substances were separated by Fischer and Hepp, but it was found more economical to reduce them together, *in situ*, taking advantage of the relative insolubility of diaminostilbene salts.

The temperature of the bath was lowered to 100°, and a solution of stannous chloride crystals (330 g.; 4 mols. per mol. of nitrotoluene) in concentrated hydrochloric acid (330 ml.), warmed to 80°, was added down the upright condenser during 15 minutes. Stirring and heating at 100° (bath temp.) were continued for 2 hours. The flask was then cooled and kept in ice for 2 days.

The precipitate of sodium chloride and diaminostilbene stannichloride was filtered off, pressed well, and extracted successively with 500, 200, and 50 ml. of boiling water, leaving about 1 g. of sludge. The combined filtrates were boiled and treated with sodium hydroxide (*ca.* 350 ml. of 5*N*) until Orange-II paper was reddened. The mixture was heated for ½ hour on the boiling water-bath and again adjusted to redder Orange-II. The precipitate was filtered hot, pressed, and washed until the filtrate ceased to redden phenolphthalein. The cake was dissolved in the minimum (*ca.* 300 ml.) of boiling *n*-hydrochloric acid, 75 ml. of 10*N*-hydrochloric acid were then added, and the whole refrigerated overnight. Next day the diaminostilbene hydrochloride was filtered off, dissolved in boiling water (150 ml.) containing 10*N*-hydrochloric acid (1 ml.), and treated with sodium hydroxide, as above. The cake was collected, pressed hard, washed well, and dried at 120°. The yield was 17.0 g. (45%) of orange powder, m. p. 214°.

This was purified by boiling with 20 parts of chlorobenzene, filtering hot, and cooling in ice for a day (85% recovery; m. p. 225—226°).

Finally, recrystallisation from dilute alcohol removed a trace of orange impurity and gave minute, almost colourless crystals, m. p. 227—228° equal to the highest uncorrected m. p.s in the literature (Calvin and Buckles, *J. Amer. Chem. Soc.*, 1940, **62**, 3324; Klinge, *Ber.*, 1883, **16**, 943).

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