642. *Antituberculous Compounds. Part III.* p-*Alkoxy*-N-*aryl*-*benzamidines.*

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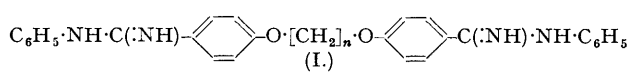
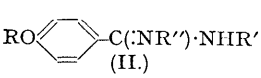
As an extension of experiments on the relation between structure and activity against *Mycobacterium tuberculosis* of *N*-substituted amidines, a series of *p*-alkoxy-*N*-arylbenzamidines and certain analogues have been prepared. Although high activities were observed *in vitro* in several cases, no activity could be demonstrated *in vivo*. The effects, on activity, of homology in the alkoxy-group and of substitution in the *N*-aryl group in *p*-alkoxy-*N*-arylbenzamidines are different from those observed with di-(*p*-*N*-arylamidinophenoxy)alkanes.

In Part II (this vol., p. 2683) evidence was presented that in a homologous series of di-(*p*-*N*-phenylamidinophenoxy)alkanes (I; n = 2, 3, 4, 5, or 6) there is marked *in vitro* activity against *Mycobacterium tuberculosis* in members having an odd number of methylene groups and no activity in members containing an even number of methylene groups. Although the activity was maintained *in vitro* in the presence of serum, no favourable effect on the course of experimental tuberculosis in guinea-pigs could be demonstrated. The work on *N*-substituted monoamidines described in this paper was undertaken in order to accumulate further evidence on the relation between structure and activity and in the hope that *in vivo* activity would ultimately be observed. The effect of homology on activity was studied in a series of *p*-alkoxy-



(I.)



(II.)

N-phenylbenzamidines (II; R = alkyl, R′ = C_6H_5, R″ = H). In addition, an examination was made of the effect of substitution in the *N*-phenyl substituent of the amidine group, of the replacement of the *N*-phenyl group by other ring systems, and of the introduction of a further phenyl residue in the amidine group to give (II; R′ = R″ = C_6H_5, R = alkyl).

The *p*-alkoxyphenyl cyanides described in the Experimental section were obtained by interaction of the appropriate alkyl halide and sodium *p*-cyanophenoxide in ethanol. Preparation of the *p*-alkoxy-*N*-arylbenzamidines by reaction of the cyanide with an arylammonium benzenesulphonate presented no difficulty. Attempts to obtain *p*-butoxy-*NN*-diphenylbenzamidine

by this method failed. In the case of p-*allyloxy*-N-*phenylbenzamidine*, the yield of purified amidine was low (28%) since the period of heating was reduced to avoid a Claisen rearrangement of the p-*allyloxyphenyl cyanide*. Considerable tar formation occurred in the preparation of p-*butoxy*-N-p′-*carbethoxyphenylbenzamidine* (II; R = Bu^n, R′ = *p*-C_6H_4·CO_2Et, R″ = H) from p-*carbethoxyanilinium benzenesulphonate* and p-*butoxyphenyl cyanide*. Hydrolysis of this ester was accomplished, without hydrolysing the amidine group, by boiling it with sodium hydroxide, although the reaction proceeded slowly. p-*Methoxy*- (II; R = CH_3; R′ = R″ = C_6H_5) and p-*butoxy*-NN′-*diphenylbenzamidine* (II; R = Bu^n, R′ = R″ = C_6H_5) were obtained from the corresponding anilides through the imido-chlorides. The aluminium chloride method (Oxley, Partridge, and Short, *J.*, 1947, 1110) was used in the preparation of p-*methoxy*-N-2-*pyridylbenzamidine*. For biological testing, these amidines were obtained in solution as their lactates.

The *in vitro* activities of members of these series are recorded in the Table. Discussion of these results is restricted to certain aspects which appear to have a bearing on the relation between structure and activity against *M. tuberculosis*; a full account will be given elsewhere. In the series of *p*-alkoxy-*N*-phenylbenzamidines (II; R″ = H, R′ = C_6H_5, R = Me, Et, Pr^n, Bu^n, *n*-amyl, or *n*-hexyl) the activity increases with increase in the length of the alkyl chain to an unusually high value for p-*hexyloxy*-N-*phenylbenzamidine*; with the corresponding *octyl* homologue the activity decreases somewhat. Homologation in *p*-alkoxyanilines and 5-amino-2-alkoxypyridines (Friedmann *et al.*, *J. Pharm. Exp. Ther.*, 1947, **89**, 153; *J. Amer. Chem. Soc.*, 1947, **69**, 1204, 1795; Forrest, D'Arcy Hart, and Walker, *Nature*, 1947, **160**, 94) has a similar effect, but the same pronounced difference in activity of the butoxy- and hexyloxy-homologues is not apparent. Similar small, and probably not significant, differences are to be seen in the effect of homologation on the activities of alkylresorcinols (Drea, *J. Bact.*, 1946 **51**, 507) and of alkyl *p*-aminobenzoates (Bloch *et al.*, *Helv. Chim. Acta*, 1947, **30**, 539) against *M. tuberculosis*. Although certain *p*-alkoxy-*N*-phenylbenzamidines resemble the analogous di-(*p*-*N*-phenylamidinophenoxy)alkanes in showing marked activity against *M. tuberculosis*, the relationship of activity to the length of the alkyl chain is quite different.

Benzamidine.	Yield, %.	M. p.	Formula.	Found, N, %.	Required, N, %.	Activity.* In absence of serum.	Activity.* In presence of serum.
1. p-*Methoxy*-N-*phenyl* as *benzenesulphonate*	51	186—187°	$C_{20}H_{20}O_4N_2S$	7·3	7·3	1 (5)	—
2. p-*Ethoxy*-N-*phenyl*-	55	143·5—144·5	$C_{15}H_{16}ON_2$	11·6	11·7	10	—
3. p-*Propoxy*-N-*phenyl*- ...	72	128—129	$C_{16}H_{18}ON_2$	11·1	11·0	10 (100)	—
4. p-*Butoxy*-N-*phenyl*-	90	121—122	$C_{17}H_{20}ON_2$	10·45	10·45	100	50—100
5. p-*Hexyloxy*-N-*phenyl*- ...	90	122—124	$C_{19}H_{24}ON_2$	9·3	9·45	5000	500—1000 (5000)
6. p-*Octyloxy*-N-*phenyl*- ...	90	118—119	$C_{21}H_{28}ON_2$	8·7	8·65	1000	100
7. *p*-Methoxy-*N*-*cyclo*hexyl-	—	—	—	—	—	5	—
8. p-*Methoxy*-NN′-*diphenyl*-	—	—	—	—	—	<1	—
9. p-*Methoxy*-N-2-*pyridyl*-	—	—	—	—	—	1 (5)	—
10. *p*-Methoxy-*N*-2-diphenylyl-	—	—	—	—	—	100	100
11. *p*-Methoxy-*N*-2-(4′-nitrodiphenylyl)-	—	—	—	—	—	50	—
12. p-*Butoxy*-NN′-*diphenyl*-	—	—	—	—	—	1—5	—
13. p-*Butoxy*-N-p′-*chlorophenyl*-	50	151—152	$C_{17}H_{19}ON_2Cl$	9·5	9·3	100	50—100
14. p-*Butoxy*-N-p′-*butoxyphenyl*-	60	149—150	$C_{21}H_{28}O_2N_2$	8·5	8·25	1000	100
15. p-*Butoxy*-N-p′-*carbethoxyphenyl*-	43	138·5—139·5	$C_{20}H_{24}O_3N_2$	8·5	8·25	—	—
16. p-*Butoxy*-N-p′-*carboxyphenyl*-	—	—	—	—	—	—	<1 (5)
17. p-iso*Butoxy*-N-*phenyl*- ...	88	131—132	$C_{17}H_{20}ON_2$	10·6	10·45	100	10—50
18. p-*Allyloxy*-N-*phenyl*- ...	28	115—117	$C_{16}H_{16}ON_2$	11·2	11·1	10	—

* Dilution in thousands at which complete inhibition of the growth of *M. tuberculosis* (human virulent strain) was maintained for 4 weeks in modified Long's medium, using the floating pellicle method. Figures in parentheses represent dilutions at which partial inhibition occurred. Under the same conditions of test *p*-aminosalicylic acid gave a value of 10 in the absence of serum.

1. Leaflets from aqueous ethanol. 2. Leaflets from light petroleum (b. p. 100—120°); the *benzenesulphonate*, prisms from water, had m. p. 168—169° (Found: N, 7·1. $C_{21}H_{22}O_4N_2S$ requires N, 7·05%). 3, 4, 5, 6, 14, 17. Leaflets from light petroleum (b. p. 80—100°). 7. Oxley, Partridge, and Short, *J.*, 1947, 1110. 10, 11. Cymerman and Short, this vol., p. 703. 13. Prisms from ethanol. 15. Needles from light petroleum (b. p. 100—120°). 18. Leaflets from light petroleum (b. p. 80—100°); insoluble in aqueous sodium hydroxide.

In agreement with the difference in activity which was reported in Part II for di-(*p*-*N*-phenylamidinophenoxy)propane and the corresponding unsubstituted diamidine, 5-amidino-2-butoxypyridine, described by Forrest and Walker (*J.*, 1948, 1939), appears to be relatively inactive against *M. tuberculosis* as compared with *p*-butoxy-*N*-phenylbenzamidine. Replacement of *n*-butyl by *iso*butyl in *p*-butoxy-*N*-phenylbenzamidine produces little change in activity, whereas a similar change in 5-amino-2-butoxypyridine causes a sixteen-fold decrease in activity (Friedmann *et al.*, *loc. cit.*). *p*-Allyloxy-*N*-phenylbenzamidine is of about the same low order of activity as the corresponding *propoxy*-derivative.

The introduction of a *p*-chloro-atom into the *N*-phenyl substituent of *p*-butoxy-*N*-phenylbenzamidine causes no change in activity, whereas a *n*-butoxy-group, similarly placed, increases the activity ten-fold. Analogous substitution of chlorine atoms and butoxy-groups in di-(*p*-*N*-phenylamidinophenoxy)propane decreases the activity (Part II, *loc. cit.*). A second phenyl substituent in the amidine group affords compounds of lowered activity in the two examples of this type described here, whereas the *N*-diphenylyl group enhances one-hundred times the activity of the parent compound, p-*methoxy*-N-*phenylbenzamidine*. Replacement of the *N*-phenyl group of the latter by *cyclo*hexyl (II; R = CH_3, R′ = *cyclo*hexyl, R″ = H) and by 2-pyridyl (II; R = CH_3, R′ = 2-pyridyl, R″ = H) produces no outstanding change in activity. p-*Butoxy*-N-p′-*carboxyphenylbenzamidine*, which is almost devoid of activity against *M. tuberculosis*, was prepared in the hope that a compound of high activity but low toxicity would be obtained.

In contrast with di-(*p*-*N*-arylamidinophenoxy)alkanes, the activities *in vitro* of the more active members of this series are decreased by serum, although several of the compounds retain activities of the order of 1 : 100,000. A similar effect of serum on the activity of 2-alkoxy-5-aminopyridines was noted by Forrest, D'Arcy Hart, and Walker (*loc. cit.*). It was not possible to demonstrate any favourable response in experimental tuberculosis in guinea-pigs with any of the compounds described here which show significant activity *in vitro*.

Experimental.

p-*Alkoxyphenyl Cyanides*.—*p*-Cyanophenol, dissolved in a solution of one equivalent of sodium in absolute ethanol, and one equivalent of the appropriate alkyl halide were refluxed for 16—20 hours. The residue left after removing the sodium halide and the solvent was stirred with water and ether, and the aqueous layer was washed with ether. Unchanged *p*-cyanophenol was removed from the combined ethereal solutions with N-sodium hydroxide and, after evaporation of the solvent from the dried solution, the residue was fractionally distilled or crystallised from light petroleum. Thus were obtained: p-*propoxyphenyl cyanide*, leaflets (54%), m. p. 47°, b. p. 121—122°/3 mm. (Found: N, 8·7. $C_{10}H_{11}ON$ requires N, 8·7%); p-*butoxyphenyl cyanide*, prisms (80%), m. p. 35°, b. p. 146—148°/3 mm. (Found: N, 8·25. $C_{11}H_{13}ON$ requires N, 8·0%); p-*hexyloxyphenyl cyanide*, prisms (71%), m. p. 32°, b. p. 155—157°/3 mm. (Found: N, 7·0. $C_{13}H_{17}ON$ requires N, 6·9%); p-*octyloxyphenyl cyanide* (70%), b. p. 171—173°/2 mm. (Found: N, 6·2. $C_{15}H_{21}ON$ requires N, 6·1%); p-*allyloxyphenyl cyanide*, prisms (72%), m. p. 43° (Found: N, 8·8. $C_{10}H_9ON$ requires N, 8·8%); and p-iso*butoxyphenyl cyanide* (35%), b. p. 114—116°/1 mm. (Found: N, 8·3. $C_{11}H_{13}ON$ requires N, 8·0%).

p-*Alkoxy*-N-*arylbenzamidines*.—The *p*-alkoxy-*N*-arylbenzamidines described in the Table were prepared by heating the appropriate *p*-alkoxyphenyl cyanide with an equivalent of an arylammonium benzenesulphonate in the temperature range 180° to 210° for between 1 and 4 hours (Oxley and Short, *J.*, 1946, 147). The experiments were conducted on a 0·035—0·1-g.-mol. scale. The amidine was liberated with aqueous ammonia from a solution of the product in ethanol and purified as the free base, as a salt, or as the free base after separation from non-basic material as the lactate. The yields given are those of purified material.

p-*Methoxy*-N-2-*pyridylbenzamidine*.—*p*-Methoxyphenyl cyanide (13·3 g.) and 2-aminopyridine (9·4 g., 1 mol.) were melted together at 60°; finely powdered aluminium chloride (13·3 g.; 1 mol.) was added during 5 minutes and the mixture was heated, with occasional stirring, at 200° for 20 minutes. The base, liberated on making an aqueous suspension of the cooled product alkaline to Titan-yellow with sodium hydroxide, was collected, together with unchanged cyanide, in chloroform. The solvent was removed from the dried solution by distillation, and a 5N-hydrochloric acid extract of the residue was washed with ether to remove *p*-methoxyphenyl cyanide (5·4 g.), filtered through a layer of kieselguhr, and made alkaline with aqueous ammonia. The precipitate afforded p-*methoxy*-N-2-*pyridylbenzamidine* (5·4 g., 24%) as leaflets, m. p. 107—108°, on crystallisation from light petroleum (b. p. 80—100°) (Found: N, 18·4. $C_{13}H_{13}ON_3$ requires N, 18·5%).

p-*Methoxy*-NN′-*diphenylbenzamidine*.—*p*-Methoxy-*N*-phenylbenzimido-chloride (30 g.) (Wheeler and Johnson, *Amer. Chem. J.*, 1903, **30**, 37) and aniline (12 g., 1·05 mols.) were heated together under reflux in dry benzene (100 c.c.) for 3 hours. The p-*methoxy*-NN′-*diphenylbenzamidinium chloride* (33·5 g., 78%) which separated crystallised from ethanol in needles, m. p. 269—270° (decomp.) (Found: N, 8·25. $C_{20}H_{18}ON_2,HCl$ requires N, 8·25%). The *base*, liberated from the hydrochloride by aqueous ammonia, crystallised from ethanol in prisms, m. p. 133·5° (Found: N, 9·4. $C_{20}H_{18}ON_2$ requires N, 9·3%).

p-*Butoxybenzanilide*.—*p*-Butoxybenzoyl chloride (51·2 g.) (Pierce, Salsbury, and Fredericksen, *J. Amer. Chem. Soc.*, 1942, **64**, 1691) and 2N-sodium hydroxide (70 c.c., 0·58 mol.) were added gradually to a suspension of aniline (22·3 g., 1 mol.) in 2N-sodium hydroxide (50 c.c., 0·42 mol.). Crystallisation

of the solid product from ethanol (800 c.c.) (charcoal) afforded p-*butoxybenzanilide* (52 g., 76%), m. p. 147° (Found: N, 5·4. $C_{17}H_{19}O_2N$ requires N, 5·2%).

p-*Butoxy*-NN'-*diphenylbenzamidine*.—This *compound* was obtained from *p*-butoxybenzanilide, aniline, and phosphorus pentachloride by the method of Hill and Cox (*J. Amer. Chem. Soc.*, 1926, **48**, 3214), in 96% yield, as prisms from ethanol, m. p. 111° (Found: N, 8·2. $C_{23}H_{24}ON_2$ requires N, 8·15%).

p-*Carbethoxyanilinium Benzenesulphonate*.—This *ester*, obtained by interaction of equimolecular quantities of ethyl *p*-aminobenzoate and hydrated benzenesulphonic acid in *iso*propanol, crystallised in prisms, m. p. 194—195° (Found: N, 4·4. $C_{15}H_{17}O_5NS$ requires N, 4·35%).

p-*Butoxy*-N-p'-*carboxyphenylbenzamidine*.—*p*-Butoxy-*N*-*p*'-carbethoxyphenylbenzamidine (3·4 g.) was boiled under reflux for 6 hours with 5N-sodium hydroxide (20 c.c., 10 mols.). Unchanged ester (0·5 g.) was collected and, when the solution was adjusted to pH 4·5—5 by the addition of 5N-hydrochloric acid, p-*butoxy*-N-p'-*carboxyphenylbenzamidine* (2·1 g., 78%) was precipitated; it formed leaflets from ethanol, m. p. 249—250° (decomp.) (Found: N, 9·15. $C_{18}H_{20}O_3N_2$ requires N, 9·0%).

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