ml of MeOH was added 1.6 g (0.01 mole) of 1-naphthaldehyde and 3 drops of piperidine. After 30-hr refluxing, solvent was evapd off, and the residue was washed with Et_2O and then recrystd from hot 1:1 water–MeOH to yield 2.8 g (75%) of IXa·HI, mp 278–281°, dec. *Anal.* ($C_{18}H_{17}IN_2$) C, H.

Treatment of the HI salt at room temp with 10% aq NaOH yielded the base; recrystd from hot CH_3CN, mp 200–203°. Treatment of the base with HI regenerated the original salt (same mp and ir spectrum).

1-Hydroxyethyl-2-imino-4-(1-naphthylethenyl)-1,2-dihydropyridine (IXb) and HBr Salt. A soln of 5.3 g (0.05 mole) of 2-amino-4-methylpyridine and 6.2 g (0.05 mole) of bromoethanol in 30 ml of *i*-PrOH was refluxed overnight. Crude product sepd on cooling and was recrystd from hot MeCN to yield 9.2 g (80%) of 1-hydroxyethyl-2-imino-4-methyl-1,2-dihydropyridine·HBr, mp 132–134°. This was treated with 1-naphthaldehyde and the product recrystd as described for IXa, mp 214–217°. *Anal.* ($C_{19}H_{19}BrN_2O$) C, H.

Treatment of the HBr salt with 10% aq NaOH at room temp yielded the base; recrystd from hot *i*-PrOH, mp 162–163. *Anal.* ($C_{19}H_{18}N_2O$) C, H, N.

Acknowledgments.—The choline acetyltransferase and AChE inhibition studies at Montefiore Hospital and Medical Center were supported in part by U. S. Public Health Service Grants MHI3331 and NBO5541. The research at the University of North Carolina was supported in part by U. S. Public Health Service Grant NBO7932. Compound VIII was prepared by M. L. Edwards while supported by a fellowship from Bristol Laboratories, Syracuse, New York. We are indebted to Pfizer Medical Research Laboratories, Groton, Conn., for a sample of 1-(phenylvinyl)pyridinium bromide.

Inhibitors of Blood Platelet Aggregation. 1. Biphenylyloxyalkylamines

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Received July 28, 1970

Certain biphenylyloxyalkylamines (**2**) inhibit ADP-induced platelet aggregation *in vitro*. The most active compounds, 5-(4-chloro-2-phenylphenoxy)-*N*-ethyl-*N*-(2-hydroxyethyl)amylamine·HCl (**14**) and the homologous butylamine·HCl (**11**), afford 50% inhibition of ADP-induced platelet aggregation at 4.8×10^{-5} *M* and 7.1×10^{-5} *M*, respectively. The structural requirements for activity are rather general. Thus, a compound having a large lipophilic moiety separated from a basic N by 2–10 elements will usually inhibit, to some degree, this type of platelet aggregation. The activity of these compounds appears to be related to that exhibited by "membrane-active" drugs such as the tricyclic antidepressants and certain antihistamines. The mechanism of action appears to be mediated through a nonspecific adsorption of these lipophilic amines onto the platelet membranes, causing a general disruption of membrane function.

One of the earliest events in the formation of an intravascular thrombus is the aggregation of blood platelets.[1] This intravascular aggregation has been described as a pathological exaggeration of the normal role of platelets in hemostasis and repair.[2] It is generally conceded that an agent which would reduce abnormally high platelet adhesiveness to, but not below, that level required for adequate hemostasis may be useful in the treatment and prophylaxis of thrombotic diseases.

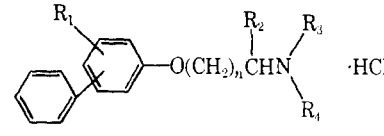
The role of adenosine diphosphate (ADP) as an important and biologically omnipresent initiator of platelet aggregation has been well documented.[3] Thus, agents that will inhibit ADP-induced platelet aggregation are of interest as potential drugs.

A number of lipophilic amines have been reported to inhibit ADP-induced platelet aggregation *in vitro*. Among these are tricyclic antidepressants,[4] antihistamines,[4c,5] and phenothiazine antipsychotic agents.[5a,c,6]

We explored the structure–activity relationship of a series of biphenylyloxyalkylamines. The general structure (**2**) of this series can be derived from **1** by moving the side chain to one of the rings and eliminating the methylene bridge. Two methods were used to prepare

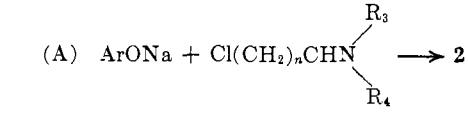
$(C_6H_5)_2CHO(CH_2)_2N(CH_3)_2$

1

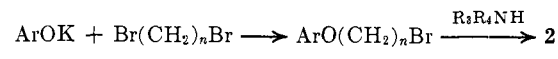


2

the compounds listed in Tables I and II.



(A) $ArONa + Cl(CH_2)_nCHNR_3R_4 \longrightarrow$ **2**



(B) $ArOK + Br(CH_2)_nBr \longrightarrow ArO(CH_2)_nBr \xrightarrow{R_3R_4NH}$ **2**

Both methods involve a Williamson ether synthesis for coupling phenols and side chains. In method A the amine moiety was introduced using an aminoalkyl halide, whereas in method B an ω-bromoalkoxybiphenyl was prepared and used to alkylate the appropriate amine.

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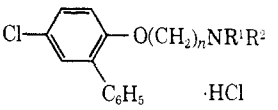
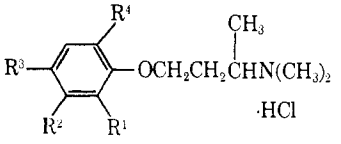
TABLE I



No.	n	R^1	R^2	Method	Mp, °C[a]	Formula	Anal.	Rp[b]
3	2	CH_3	CH_3	A	133–135	$C_{16}H_{19}BrClNO$[c]	C, H, N	0.1
4	3	H	H	B	174–175	$C_{15}H_{17}Cl_2NO$	C, H, N	0.5
5	3	CH_3	CH_3	A	173–174	$C_{17}H_{21}Cl_2NO$	C, H	0.9
6	3	C_2H_5	C_2H_5	B	139–140	$C_{19}H_{25}Cl_2NO$	C, H, N	0.8
7	3	H	C_2H_5	B	130–132	$C_{17}H_{21}Cl_2NO$	C, H	0.4
8	3	CH_3	$(CH_2)_2OH$	B	136–137	$C_{18}H_{23}Cl_2NO_2$	C, H[d]	0.7
9	3	C_2H_5	$(CH_2)_2OH$	B	120–122	$C_{19}H_{25}Cl_2NO_2$	C, H, N	0.5
10	4	C_2H_5	C_2H_5	B	105–106	$C_{20}H_{27}Cl_2NO$	C, H	1.1
11	4	C_2H_5	$(CH_2)_2OH$	B	114–116	$C_{20}H_{27}Cl_2NO_2$	C, H	1.4
12	5	CH_3	CH_3	B	105–106	$C_{19}H_{25}Cl_2NO$	C, H, N	1.2
13	5	C_2H_5	C_2H_5	B	125–126	$C_{21}H_{29}Cl_2NO$	C, H	1.2
14	5	C_2H_5	$(CH_2)_2OH$	B	103–105	$C_{21}H_{29}Cl_2NO_2$	C, H	2.1
15	6	CH_3	CH_3	B	120–121	$C_{20}H_{27}Cl_2NO$	C, H, N	0.7
16	10	CH_3	CH_3	B	110–112	$C_{24}H_{35}Cl_2NO$	C, H	0.3

[a] Recrystn solvent EtOH–Et_2O except **16** which was recrystd from EtOAc–Et_2O. [b] Rp is the relative potency of the compd compared with **17**, which affords 50% inhibition of ADP-induced platelet aggregation at 1×10^{-4} *M*. For comparison, adenosine afforded 50% inhibition at 3×10^{-7} *M* and methapyrilene·HCl at 5×10^{-4} *M*.[5a] [c] HBr salt. [d] H: calcd 6.24; found 6.72.

TABLE II



No.	R^1	R^2	R^3	R^4	Mp, °C[a]	Formula	*Anal.*	Rp
17	Ph	H	Cl	H	146–148	$C_{18}H_{23}Cl_2NO$	C, H	1.0
18	Ph	H	H	H	142–143	$C_{18}H_{24}ClNO$	C, H, N	0.6
19	H	Ph	H	H	86–87	$C_{18}H_{24}ClNO$	C, H	0.6
20	H	H	Ph	H	160–163	$C_{18}H_{24}ClNO$	C, H	0.8
21	Cl	H	Ph	H	150–151	$C_{18}H_{23}Cl_2NO$	C, H, N	0.8
22	Ph	H	H	Cl	134–136	$C_{18}H_{23}Cl_2NO$	C, H	0.6
23	Ph	H	NO_2	H	170–172	$C_{18}H_{23}ClN_2O_3$	C, H, N	0.5
24	Ph	H	NH_2	H	229–231	$C_{18}H_{26}Cl_2N_2O$[b]	C, H, N	0.3

[a] All compds listed in Table II except **24** were prepared by method A. Recrystn solvent EtOH–Et_2O. [b] Dihydrochloride salt.

The compounds prepared during the course of this investigation were tested for their *in vitro* activity as inhibitors of ADP-induced platelet aggregation by utilizing a procedure reported previously.[5a] In addition to inhibiting ADP-induced platelet aggregation, these compounds also inhibit collagen-, thrombin-, and epinephrine-induced aggregation. Among the most interesting members of this series are **10–14** (Table I) and **17** (Table II). They inhibit ADP-induced aggregation by 50% at concentrations ranging from 1×10^{-4} *M* to 5×10^{-5} *M*. None of these compounds are as effective as adenosine which affords 50% inhibition at 3×10^{-7} *M*. However, the activity of these compounds is comparable to that reported for the tricyclic antidepressants,[4] the antihistamines,[4c,5] and the phenothiazines.[5a,c,6]

Inhibitory activity increases with increasing chain length to a maximum at a 5-C separation between the O and N atoms (**10–14**). Significant activity is still seen, however, with a 10-C separation (**16**). Critical comparison of the data in Table I and Table II reveals that many rather large changes in structure are accompanied by relatively small changes in potency. This seems to indicate a nonspecific mechanism of action and probably reflects the effect of adsorption of these lipophilic amines onto the platelet membrane. This adsorption would be expected to disrupt membrane function in proportion to the amount of amine bound.

Experimental Section[7]

Biological Methods and Materials. Glassware.—All glassware coming into contact with the whole blood or PRP was freshly siliconized using a 3.8% soln of General Electric SC 87 silicone in pet ether (bp 30–60°). It was necessary to carefully remove old silicone from all glassware before washing and resiliconization. This is easily accomplished by allowing the glassware to soak overnight in a mixture of 70 g of NaOH, 700 ml of Me_2CO, and 700 ml of H_2O.

Platelet-Rich Plasma (PRP).—New Zealand White rabbits were anesthetized with Na pentobarbital. The carotid artery was carefully exposed and cannulated with a 15-cm piece of siliconized PE-200 tubing. The first 3- to 5-ml portion of blood was discarded, and two 40-ml centrifuge tubes (in an ice bath) were filled. The blood was centrifuged at 0–3° and 100*g* for 20 min. The supernatant PRP was removed with a pipette, stored at 0° in a centrifuge tube, and used within 3 hr.

(7) Melting points were determined on a Mel-Temp melting point apparatus and are uncorrected. Where analyses are indicated only by symbols of the elements, analytical results for those elements were within ±0.4% of the theoretical values. All α,ω-dibromoalkanes, all phenols with the exception of 4-nitro-2-phenylphenol, and all dialkylaminoalkylchloride hydrochlorides with the exception of 3-chloro-*N*,*N*,1-trimethylpropylamine·HCl were obtained from commercial sources. The identity of compds prepared in the course of this work was confirmed by nmr analysis and, where appropriate, by anal. titration.

Platelet Aggregation.—The rate and extent of platelet aggregation were measured by the optical density method of Born[3b] and by using instrumentation described by Mustard, *et al.*[8]

A cuvette containing 1.0 ml of PRP and (for controls) 0.3 ml of imidazole buffer (pH 7.4) or (for test compounds) 0.3 ml of a soln of the compd in imidazole buffer was placed in the aggregometer and allowed to warm to 37° during 2 min. At this point, the PRP was challenged with 0.1 ml of a soln of ADP in the imidazole buffer.

The sensitivity of platelets to aggregating agents varies from preparation to preparation. Therefore, the amt of ADP used to challenge a particular PRP preparation was adjusted to afford a barely maximal response. This usually required a final ADP concn of $3.5–4.5 \times 10^{-7}$ *M*. In addition, an arbitrarily chosen compd (**17**) was tested in every PRP preparation and used as a reference standard. The activity of each compd was calcd relative to the activity of this reference compd. The per cent inhibition of aggregation by a test compd was calcd by dividing the max deflection in the optical density curve during aggregation in the presence of the compd by that observed in the control, then multiplying by 100. The molar concds of each compd required to inhibit aggregation by between 20 and 50% and by between 50 and 80% were determined and plotted *vs.* the observed per cent inhibition (semilog graph paper).[9] The molar concn of the test compd which would inhibit aggregation by 50% was read from the graph and divided by the molar concn of **17** necessary to inhibit aggregation by 50%. The result is the relative potency (Rp) value reported for the compds listed in Table I and Table II.

Chemical Methods and Materials. 4-Nitro-2-phenylphenol (25).—HNO_3 (2.95 moles, 186 ml, ρ 1.42) was added dropwise during 3 hr to a soln of 2-phenylphenol (500 g, 2.95 moles) in 2 l. of AcOH. The temp was kept between 15 and 18°. The mixture was stirred overnight at room temp and was poured into 10 l. of ice–H_2O. The pptd crude product was extd into 2 l. of Et_2O, washed with H_2O, dried (Na_2SO_4), filtered, and evapd. The residue was dissolved in 1 l. of C_6H_6 and poured over a column of 500 g of Florisil (100–200 mesh, 7.5 × 45 cm). The column was eluted with C_6H_6. The fractions eluting from 1000 ml to 5000 ml were combined, and the C_6H_6 was distd (vac). The residue was recrystd twice from C_6H_6, affording 160 g of **17** (yellow solid), mp 125–127° (lit.[10] mp 125–126°). *Anal.* ($C_{12}H_9NO_3$) C, H, N.

3-Chloro-*N*,*N*,1-trimethylpropylamine·HCl (26).—A soln of 3-dimethylaminobutanol[11] (647 g, 5.53 moles) in 2.4 l. of $CHCl_3$ was chilled in a salt–ice bath and satd with HCl (g). A soln of $SOCl_2$ (1450 ml) in 800 ml of $CHCl_3$ was added slowly with stirring and cooling. The mixture was allowed to warm to room temp and stir overnight; it was then refluxed for 2 hr and evapd to dryness. Abs EtOH (500 ml) was added to and distd (vac) from the residue 4 times. One recrystn from EtOH–Et_2O afforded **26** (972 g of colorless needles), mp 144–146°. *Anal.* ($C_6H_{13}Cl_2N$) C, H, N.

3-(2-Phenyl-4-aminophenoxy)-*N*,*N*,1-trimethylpropylamine·2HCl (24).—A soln of **23** (3.5 g, 0.01 mole) in 200 ml of abs EtOH and 0.5 g of 5% Pd–C was shaken under 3.6 kg/cm² of H_2 until the theoretical amount had been consumed (*ca.* 1 hr). The reaction mixture was filtered and evapd under reduced pressure, and the residue was dissolved in 10 ml of abs EtOH. The EtOH soln was treated with HCl (g). The ppt was collected by filtration and recrystd twice from EtOH affording **24** (1.8 g of tan solid), mp 220–231°. *Anal.* ($C_{18}H_{26}Cl_2N_2O$) C, H, N.

Method A. This general procedure involves the reaction of a dialkylaminoalkyl chloride with a sodium phenoxide in EtOH. The following example illustrates the method.

3-(4-Chloro-2-phenylphenoxy)-*N*,*N*,1-trimethylpropylamine·HCl (17).—A soln of **26** (206 g, 1.2 moles) in 120 ml of ice–H_2O was treated with 120 ml of cold 50% NaOH. The free base was extracted into 1 l. of Et_2O, dried (Na_2SO_4), and filtered; the solvent was distd (vac, gentle warming). The residue was added to a soln of 4-chloro-2-phenylphenol (204 g, 1.0 mole) and the NaOEt from 23 g of Na in 2 l. of EtOH. The mixture was refluxed overnight, cooled, filtered, and evapd *in vacuo*. The residue was dissolved in 2 l. of 3 *N* HCl, washed with Et_2O, and made alkaline with 50% NaOH. The crude **17** was extd into 2 l. of Et_2O, washed with four 500-ml portions of H_2O, dried (Na_2SO_4), and filtered. The HCl salt was formed by passing in HCl (g) until pptn was complete. Two recryst from EtOH–Et_2O afforded **17**: mp 146–148°; pK_a 8.6; neut equiv 351. *Anal.* ($C_{18}H_{23}Cl_2NO$) C, H.

Method B. ω-Bromoalkoxybiphenyls.—These intermediates were prepd from the appropriate potassium phenoxide and dibromoalkane. The following example illustrates the procedure.

2-(10-Bromodecyloxy)-5-chlorobiphenyl (27).—A soln of 4-chloro-2-phenylphenol (40.8 g, 0.2 mole), KOH (13 g), and 1,10-dibromodecane (141 g, 0.47 mole) in 1 l. of MeOH was refluxed overnight. The mixture was cooled and filtered, and the solvent distd (vac). The residue was dissolved in 1 l. of Et_2O, washed with several portions of dil NaOH and then with H_2O until the washes were neutral. The organic phase was dried (Na_2SO_4) and filtered and the solvent distd (vac). Distn of the residue afforded 41 g of **27** (viscous yellow oil); bp 230–235° (0.1 mm). *Anal.* ($C_{22}H_{28}BrClO$) C, H.

The ω-bromopropyl, -Bu, -Am, and -Hex ethers of 4-chloro-2-phenylphenol were prepd in the same way; but the distn was stopped after excess dibromoalkane had been removed. The crude intermediates were used to alkylate a variety of amines. The following example illustrates the procedure.

10-(4-Chloro-2-phenylphenoxy)-*N*,*N*-dimethyldecylamine (16).—A soln of the bromide **27** (6 g) and Me_2NH (50 ml) in abs EtOH (200 ml) was kept 6 hr at 110° under pressure. The solvent and excess amine were distd and the residue was dissolved in 500 ml of dil HCl. The aq soln was washed with Et_2O and made alkaline with 50% NaOH. The product was extd into 1 l. of Et_2O, washed with several portions of H_2O, dried (Na_2SO_4), filtered, and treated with HCl (g), until pptn of the HCl salt was complete. The crude product was recrystd from EtOAc–Et_2O, affording **16** (4.1 g): mp 110–112°; pK_a 9.3: neut equiv 440. *Anal.* ($C_{24}H_{40}Cl_2NO$) C, H.

Acknowledgments.—The authors are indebted to Mr. Charles Ashbrook and Mr. Larry White for preparation of some of the compounds, to Mr. George Maciak for microanalyses, to Mr. Jack Campbell for high-pressure reactions, and to Mr. D. O. Woolf for spectral and titration data. We thank the Dow Chemical Company, Midland, Mich., for a generous sample of 4-chloro-2-phenylphenol.

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(9) In most cases at least 1 additional point (usually between 0 and 30% and/or between 70 and 100%) was determined. In some cases only the 2 primary points were used. However, this method was shown by repeat experiments to be capable of reproducing the dose–response curves of members of this series.

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