Table II

Compound	Approximate LD_{50}, mg./kg. i.p., mouse[a]	Pseudocholinesterase I_{50}, M	Other actions[b]
I	56	$5 \times 10^{-}$[illegible]	Depressant
II	56	8×10^{-5}	
III	56	$1 \times 10^{-}$[illegible]	
IV	562		Depressant
V	1000	$2 \times 10^{-}$[illegible]	Stimulant
VI	75	3×10^{-5}	Stimulant
VII	56	$3 \times 10^{-}$[illegible]	
VIII	56	3×10^{-5}	
IX	56	$5 \times 10^{-}$[illegible]	
X	42	2×10^{-4}	Stimulant
XI	21	$3 \times 10^{-}$[illegible]	

[a] Toxicity was determined in groups of 4 albino mice per dose utilizing doses decreasing in 0.5 log intervals from 1000 mg./kg. (intraperitoneal route). The LD_{50} was estimated by the method of Spearman and Karber and the 95% confidence intervals are approximately ±0.3 log units. Observations of the effects of these compounds on behavior were carried out simultaneously with the determination of toxicity. [b] All compounds caused convulsions in lethal doses; the observations in this column apply to effects seen at nonlethal doses.

acetate IIa in 75 ml. of ethanol was added a solution of 2.4 g. (0.06 mole) of sodium hydroxide in 15 ml. of water. The solution was stirred under reflux for 2 hr. and at room temperature overnight. The solvent was removed under reduced pressure and the residue was taken up in methylene chloride and a small amount of water. The water layer was separated and the organic layer was dried, filtered, and evaporated. The residue was distilled to give 8.8 g. (90.7%) of a colorless viscous oil, b.p. 132–134° (0.30–0.35 mm.). A sample of liquid, b.p. 134° (0.35 mm.), $n^{25}D$ 1.5265, was submitted for analysis: ν^{max} 3420 (OH stretching) and 1665 $cm.^{-1}$ (amide carbonyl).

Anal. Calcd. for $C_{11}H_{17}NO_2$: C, 67.66; H, 8.78; N, 7.17. Found: C, 67.33; H, 8.89; N, 6.94.

Alternate Preparation of 1-(2-Morpholinoethyl)-1,3-dihydro-3,5,7-trimethyl-2H-azepin-2-one Hydrochloride *via* **IIc.**—An ice-cold solution of 4.9 g. (0.025 mole) of IIc in 35 ml. of pyridine was treated with 5.35 g. (0.028 mole) of *p*-toluenesulfonyl chloride in several portions. The resulting solution was stored at 5° for 2 days. Ice water (300 ml.) was added and, after standing at 5° for 2 hr., the aqueous solution was extracted with ether. The combined ether layers were washed with cold N hydrochloric acid, dried, filtered, and evaporated. The residual oil, which did not crystallize, was used without further purification.

The crude tosylate from this was dissolved in 10 ml. of morpholine and the solution was allowed to stand overnight at room temperature. Crystals had separated; ether was added, and the precipitated solid was separated by filtration. The filtrate was evaporated under reduced pressure and the residual oil was converted to its hydrochloride, 1.34 g. (20.3%, over-all), m.p. 219–223° dec. The infrared spectrum of this material was identical with that of the sample prepared by the alternate route. A mixture melting point showed no depression.

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Synthesis of Unsymmetrical Diphenylalkenes[1]

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Bis-(*p*-methoxyphenyl)-alkenes have been prepared by action of the Grignard reagent from *p*-bromoanisole on the esters of carboxylic acids. In the same manner using cycloalkanecarboxylic acids some bis-(*p*-methoxyphenyl)-cycloalkylidene methanes have also been prepared. Their demethylation by potassium hydroxide triethyleneglycol was in several cases accompanied by the formation of the ethane derivatives. Ultraviolet absorption data are discussed.

The present tendency in steroid hormone research is to look for substances with a more specific action on different tissues or organs.[3] Surprisingly little has been done in this respect with synthetic estrogens belonging to the stilbestrol–hexestrol series. This is probably due to the fact that interest in these substances was particularly high at a time when it was necessary to discover inexpensive and potent estrogenic substances.

An investigation of the specific action of hexestrol and stilbestrol analogs on different biological receptors was therefore decided on in our Institutions. Several results have already been presented.[4,5] This report concerns the synthesis of diphenols, their ethers, and esters belonging to the class of *unsym*-diphenylethylene.

Some substances from this series have already been found to possess the ability to interfere with the vaginal estrus reaction induced by estradiol benzoate.[6] Their relations to diethylstilbestrol and trianisylethylene, in terms of spatial structure, have also been discussed.[7]

All eighteen compounds were synthesized by the same method, by reaction of *p*-methoxyphenylmagnesium bromide with esters. The methyl or ethyl esters, which are all known compounds, were prepared from the corresponding acids by way of the acid chlorides. The following were not commercially available: 2,3-dimethylbutanoic acid,[8] 2-ethylpentanoic acid,[9] 2-methyl-4-pentenoic acid,[10] 2-methylhexanoic acid,[9] 2-butylhexanoic acid,[11] cycloheptanecarboxylic acid,[12] and cyclooctanecarboxylic acid.[12] Of these, the two cyclic

(1) For the previous paper in this series see J. F. Miquel, *Bull. Soc. Chim. France*, 239 (1962).

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(3) R. I. Dorfman, "Methods in Hormone Research II," Academic Press, New York, N. Y., 1962.

(4) E. H. Bárány, P. Morsing, W. Müller, G. Ställberg, and E. Stenhagen, *Acta Soc. Med. Upsalien.*, **60**, 68 (1955).

(5) L. Juhlin and J. F. Miquel, *Acta Endocrin.*, **36**, 87 (1961).

(6) J. F. Miquel, E. H. Bárány, and W. Müller, *Arch. Intern. Pharmacodyn.*, **117**, 262 (1958).

(7) J. F. Miquel, *Tetrahedron*, **8**, 205 (1960).

(8) P. A. Levene and L. W. Bass, *J. Biol. Chem.*, **70**, 211 (1926).

(9) M. R. Raseth, *Bull. Soc. Chim. France*, [3] **33**, 681 (1905).

(10) J. Cologne and R. Dumesnil, *ibid.*, 634 (1952).

(11) P. A. Levene and L. H. Cretcher, *J. Biol. Chem.*, **33**, 505 (1918).

(12) L. Ruzicka, P. Barman, and V. Prelog, *Helv. Chim. Acta*, **34**, 401 (1951).

acids were prepared by the reaction of carbon dioxide with the corresponding Grignard reagent, according to the method of Ruzicka.[12] The other acids were prepared by alkylating, hydrolyzing, and monodecarboxylating the malonic ester.[13] The yield of dimethoxyphenylethylene was between 50 and 80%, calculated on the basis of the esters used. In no case was isolation of the intermediate carbinol attempted, because, as is well known, the electron-donating properties of the methoxy group in the *para* position cause a strong tendency to dehydration in the carbon atom on which the benzene rings are fixed.[1,14] These dimethyl ethers are described in Table I.

Demethylation was effected by heating while stirring the methyl ethers in triethyleneglycol containing an excess of potassium hydroxide. The free phenols thus obtained were immediately acetylated to facilitate the purification (Table II). The diacetates were then hydrolyzed to free phenols (Table III). Before demethylation, the purity of the methoxy compounds was always checked by paper chromatography. Despite this, in several cases acetates and free phenols were found to be mixed with another substance rather similar in chemical and physical behavior. It was laborious to separate both substances in a spectroscopically pure condition by selective crystallization, whereas this could be done easily by paper chromatography.

Figure 1 shows a chromatogram on which the spot in position 1 was obtained with a pure sample of the diphenylethylene (XVIb) (R_f 0.43). The spot in position 2 was obtained with a pure sample of its ethane analog prepared by condensation of phenol with cyclohexanecarboxaldehyde (R_f 0.35). In position 3 a mixture, resulting from a demethylation of XVIa followed by acetylation and saponification, was used. Two spots were found at the same R_f as XVIb and its ethane analog. An elution of these spots and an analysis of the solution in the spectrophotometer gave absorption curves identical with those from the parent compounds. Both spots in position 4 were obtained when solutions of the ethylene and ethane were mixed in a 9:1 ratio. Spots produced by the ethylene compound could be detected directly with Hanovia Chromatolite (2537 Å.), whereas ethane spots were detected by the Folin–Ciocalteus reagent.

The chromatographic analysis of the methoxy and the acetoxy compounds was made on paraffin-impregnated Whatman No. 1 paper with methanol–water in varying proportions according to Larsson.[15] The analysis of the hydroxy compounds was made on Whatman No. 1 paper impregnated with formamide, with toluene as the mobile phase according to Zaffaroni.[16] Figure 2 shows the ultraviolet spectra of a pure specimen of compound XVIc (curve C) and its ethane analog (curve D). The band characteristic of *unsym*-diphenylethylene at 247 mμ in the diacetyl derivative has disappeared on curve D, which only shows the absorption of benzenoid compounds. Curves A and B, respectively, represent the dimethyl ether (XVIa) and the free phenol (XVIb).

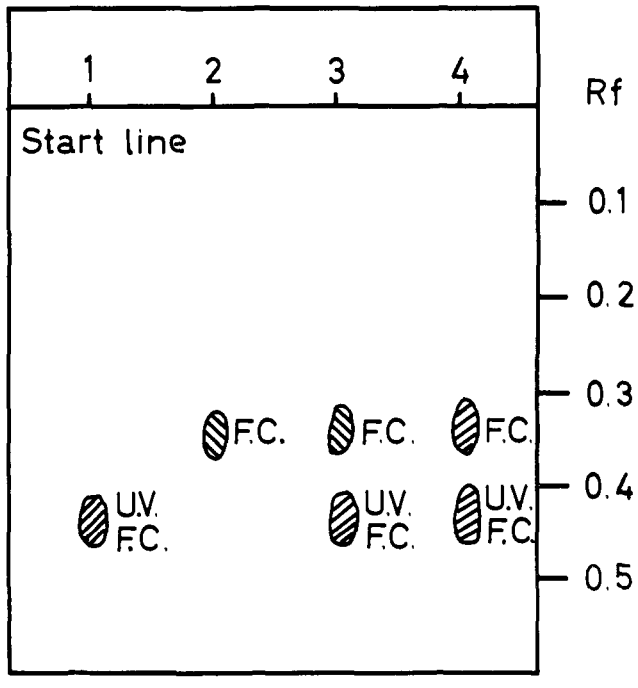


Fig. 1.—Paper chromatogram of ethylene and ethane derivatives. 1. Compound XVIb (10 γ). 2. Ethane derivative of compound XVIb (10 γ). 3. Mixture from the demethylation of XVIa (10 γ). 4. Mixture of XVIb (9 γ) and its ethane derivative (1 γ). U. V., ultraviolet absorption under ultraviolet light; F.C., blue spot with Folin–Ciocalteus reagent.

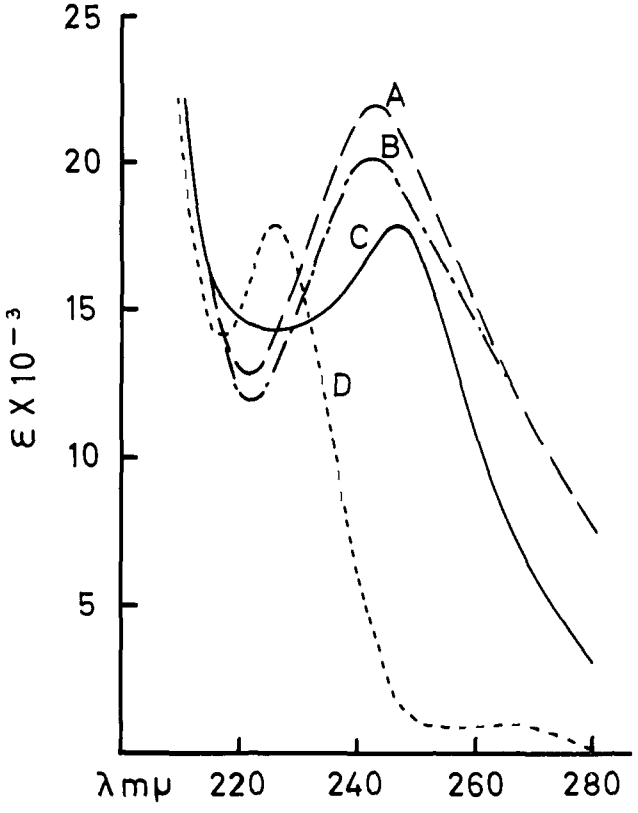


Fig. 2.—Ultraviolet spectra in ethanol of XVIa, XVIb, XVIc; D, ethane derivative of XVIc.

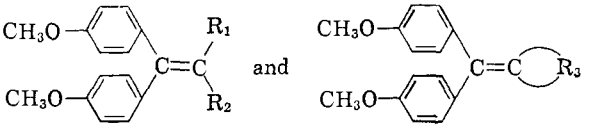
The amount of ethane analog which was formed through what could be a special kind of reduction during the course of the demethylation was not the same for each compound prepared. Compound Ia, which does not possess any substituent on the β-carbon atom,

(13) M. Conrad and C. A. Bischoff, *Ann.*, **204**, 143 (1880).
(14) M. Fetizon, M. Golfier, and A. Rassat, *Compt. Rend.*, **252**, 130 (1961).
(15) H. Larsson, *J. Chromatog.*, **11**, 331 (1963).
(16) A. Zaffaroni, *Recent Progr. Hormone Res.*, **8**, 51 (1953).

TABLE I

CHEMICAL DATA OF BIS-(*p*-METHOXYPHENYL)-ALKENES PREPARED (Ia–XVIIIa)

All the compounds are recrystallized from ethanol



Number	Name	R_1	R_2	Empirical formula
Ia[a]	1,1-Bis-(*p*-methoxyphenyl)-ethene	H	H	$C_{16}H_{16}O_2$
IIa[a]	1,1-Bis-(*p*-methoxyphenyl)-propene-1	CH_3	H	$C_{17}H_{18}O_2$
IIIa[b]	1,1-Bis-(*p*-methoxyphenyl)-butene-1	C_2H_5	H	$C_{18}H_{20}O_2$
IVa[a]	1,1-Bis-(*p*-methoxyphenyl)-2-methyl-1-propene	CH_3	CH_3	$C_{18}H_{20}O_2$
Va[c]	1,1-Bis-(*p*-methoxyphenyl)-2-methyl-1-butene	C_2H_5	CH_3	$C_{19}H_{22}O_2$
VIa	1,1-Bis-(*p*-methoxyphenyl)-2-methyl-1-pentene	n-C_3H_7	CH_3	$C_{20}H_{24}O_2$
VIIa	1,1-Bis-(*p*-methoxyphenyl)-2,3-dimethyl-1-butene	i-C_3H_7	CH_3	$C_{20}H_{24}O_2$
VIIIa	1,1-Bis-(*p*-methoxyphenyl)-2-methyl-1,4-pentadiene	CH_2—CH=CH_2	CH_3	$C_{20}H_{22}O_2$
IXa	1,1-Bis-(*p*-methoxyphenyl)-2-methyl-1-hexene	n-C_4H_9	CH_3	$C_{21}H_{26}O_2$
Xa[d]	1,1-Bis-(*p*-methoxyphenyl)-2-ethyl-1-butene	C_2H_5	C_2H_5	$C_{20}H_{24}O_2$
XIa	1,1-Bis-(*p*-methoxyphenyl)-2-ethyl-1-pentene	n-C_3H_7	C_2H_5	$C_{21}H_{26}O_2$
XIIa	1,1-Bis-(*p*-methoxyphenyl)-2-*n*-propyl-1-pentene	n-C_3H_7	n-C_3H_7	$C_{22}H_{28}O_2$
XIIIa	1,1-Bis-(*p*-methoxyphenyl)-2-*n*-butyl-1-hexene	n-C_4H_9	n-C_4H_9	$C_{24}H_{32}O_2$
			R_3	
XIVa	Bis-(*p*-methoxyphenyl)-cyclobutylidenemethane		$(CH_2)_3$	$C_{19}H_{20}O_2$
XVa[e]	Bis-(*p*-methoxyphenyl)-cyclopentylidenemethane		$(CH_2)_4$	$C_{20}H_{22}O_2$
XVIa[f]	Bis-(*p*-methoxyphenyl)-cyclohexylidenemethane		$(CH_2)_5$	$C_{21}H_{24}O_2$
XVIIa	Bis-(*p*-methoxyphenyl)-cycloheptylidenemethane		$(CH_2)_6$	$C_{22}H_{26}O_2$
XVIIIa	Bis-(*p*-methoxyphenyl)-cyclooctylidenemethane		$(CH_2)_7$	$C_{23}H_{28}O_2$

[a] Prepared earlier by the same method. See P. Pfeiffer and R. Wizinger, *Ann.*, **461**, 132 (1928). [b] S. Skraup and F. Nieten, *Ber.*, **57**, 1294 (1924), obtained a different compound with m.p. 96° in an abnormal Friedel–Crafts reaction. [c] Prepared earlier by S. H. Zaheer, B. Singh, B. Bhushan, I. K. Kacker, K. Ramachandran, and N. S. Rao, *J. Chem. Soc.*, 1706 (1955). [d] Prepared earlier by Z.

was very sensitive to this reduction. The only product identified in the 40% yield of free phenol obtained during the demethylation was found to be the ethane analog. Its identity was established by comparison with an authentic specimen obtained following the procedure of Harden and Reid.[17]

The demethylation of monosubstituted β-derivatives (IIa and IIIa) produced about 10–15% of reduced compounds from the isolated phenols. Disubstituted β-derivatives (IVa–XIIIa) were practically not reduced during demethylation. We were not able to show the presence of ethane compounds by studies of their ultraviolet absorption or by chromatographic analysis.

In the case of cycloalkylidene derivatives the same tendency to reduction was found. The amounts of ethane formed during the course of the demethylation decreased with increasing size of the ring. The reduction was quite considerable for the cyclobutane and cyclopentane compounds, but negligible for the cycloheptane and cyclooctane derivatives. We have also tried demethylations with potassium hydroxide and ethanol at 200°, but always obtained about the same amounts of hydrogenated compounds. A similar addition of hydrogen atoms to a double bond during the course of a demethylation has already been noticed during the treatment of anethole with potassium hydroxide and alcohol in an autoclave.[18] Propylphenol was isolated among the products of the reaction; hexestrol which was formed in this reaction also required the addition of two atoms of hydrogen.

We have not been able to find in the literature a study of these reductions. They may be similar to already well known reactions, such as disproportionations of the Cannizzaro reaction type or they may be a Meerwein–Ponndorf–Verley type reduction. The last is also in agreement with results reported by Behun and Levine,[19] who reduced benzophenone to benzhydrol with ethanolic potassium hydroxide.

Another phenomenon encountered during demethylation was the displacement of the allyl double bond in compound VIIIa, the diphenol VIIIb and the diacetate VIIIc now having a more stable conjugated butadiene structure. This could be proved by ultraviolet absorption. VIIIa has a band at 242 mμ (ϵ 2.07 × 10^{-4}). After demethylation and acetylation, the band is at 282 mμ (ϵ 2.03 × 10^{-4}). This is the region where an aromatically substituted butadiene with great steric hindrance to planarity is supposed to absorb. 1,4-Di-*p*-anisyl-2,3-dimethylbutadiene, which has an identical number of phenyl and methyl groups, possesses its absorption band at 298 mμ (ϵ 3.37 × 10^{-4}). The absorption at longer wave length and the higher intensity in the 1,4-dianisyl compound are in good agreement with the symmetrical structure and the less twisted position of the two phenyl groups.[1]

(17) W. C. Harden and E. E. Reid, *J. Am. Chem. Soc.*, **54**, 4325 (1932).

(18) N. R. Campbell, E. C. Dodds, and W. Lawson, *Nature*, **142**, 1121 (1938).

(19) J. D. Behun and R. Levine, *J. Am. Chem. Soc.*, **81**, 5666 (1959).

Yield, %	B.p., °C., uncor.	B.p., Pressure, mm.	$n^{25}D$	M.p., °C. cor.	% Calcd. C	% Calcd. H	% Found C	% Found H	$\lambda_{max.}$ mμ	$\epsilon \times 10^4$
50				142–143					248	2.34
81				100–101	80.28	7.13	80.0	7.29	245	2.14
									260	1.78
62	137–138	0.05		39-40	80.56	7.51	80.3	7.43	244	2.10
									260	1.83
74	190-192	3		66–67	80.56	7.51	80.3	7.65	242	2.11
62	173–174	1		49–50	80.81	7.85	80.7	7.93	242	2.08
62	171–173	0.8	1.5733		81.04	8.16	80.5	8.06	242	2.08
72	167–168	.7	1.5733		81.04	8.16	80.9	8.21	242	2.07
73	143	.05	1.5854		81.60	7.53	81.6	7.65	242	2.07
70	184	.8	1.5681		81.25	8.44	81.0	8.46	242	2.07
73	170–175	.05		92–93	81.04	8.16	80.5	8.05	241	1.95
68	176	.7	1.5656		81.25	8.44	81.1	8.36	241	1.98
71	185–190	1.5		62–63	81.44	8.70	81.3	8.62	241	2.02
64	186–187	0.5	1.5513		81.77	9.15	81.8	9.20	241	2.02
70	173–175	.05		116–117	81.39	7.19	81.1	7.16	246	1.93
									264	2.07
69	182–188	.3		64–65	81.60	7.53	81.4	7.53	245	1.88
									255	1.79
66	184–188	.1		109–110	81.78	7.84	81.2	7.70	242	2.08
58	225–230	.05		89–90	81.95	8.13	82.1	8.16	241	1.98
74	190–200	.05		68–70	82.10	8.39	81.9	8.39	241	1.88

Földi and I. Demjén, *Ber.*, **74**, 930 (1941). [e] When prepared by action of *p*-methoxyphenylcyclopentyl ketone on *p*-methoxyphenylmagnesium bromide, the crystals obtained had a m.p. of 75–76°. From both substances demethylation and acetylation afforded an identical diacetate. [f] Prepared earlier by Fetizon, *et al.*[14]

The n.m.r. spectra (60 Mc., CCl_4, TMS = 0) also make it obvious that VIIIc, in contrast to VIIIa, has a butadiene structure from the following τ values and coupling constants: VIIIc doublet centered at 8.30 (J = 6 c.p.s., $CH_3-\underset{|}{C}H=$), 8.15 τ ($CH_3-\underset{|}{C}=$). VIIIa 8.30 ($CH_3-\underset{|}{C}=$), double centered at 7 τ, broad, possibly due to the long range coupling with ethylene CH_2[20] (J = 5 c.p.s., $=\underset{|}{C}-CH_2-CH=CH_2$).

In a previous study[6] the band at 240–250 mμ, characteristic of *unsym*-dianisylethylenes, was considered to be analogous to the band described by Braude[21] for dibenzyldiphenylmethane and substituted stilbenes, and was called band E. Some reservation was made, however, regarding the application of Braude's theory to this class of compounds. With the present compounds and their spectra available, a more detailed discussion of the origin of the absorption band in the *unsym*-diphenylethylene system is possible. Starting from substance Ia, which is nonsubstituted on the β-ethylenic carbon, the absorption band is slightly displaced from 248 to 245 mμ for β-monosubstituted compounds and further on to 242 and 241 mμ when two substituents are present in the β-position. This shift is followed by a regular decrease in the intensity, falling gradually from ϵ 2.34 × 10^{-4} to 2.00 × 10^{-4}. Simultaneously the shoulder which was present at 260–280 mμ in the nonsubstituted and monosubstituted substances disappears in the disubstituted compounds.

In the cycloalkylidene compounds, cyclobutylidene and cyclopentylidene have their maximum absorptions at 246 and 245 mμ, whereas the higher homologs have their band displaced to 242–241 mμ. A new band, at 264 and 255 mμ for XIVa and XVa, respectively, which could have the same origin as the shoulder found in compounds Ia and IIa, disappears when the size of the cycloalkyl ring increases. The shift of the E′ band (from 248 to 241 mμ for the diethers) is small, but it falls outside any limits of experimental error. There is a regular tendency for the hypochromic shift to follow the presence and the size of substituents on the β-ethylenic carbon atom. Furthermore, the ultraviolet absorption band of the diacetyl esters in Table II shows a bathochromic shift as compared with the corresponding dimethyl ethers, when the β-substituents are not too large. This is particularly remarkable in the cycloalkylidene series: cyclobutylidene methane (XIVc) absorbs at 258 mμ [$\Delta\lambda_{max}$ 12 mμ from the dimethyl ether (XIVa)] and cyclooctylidene methane (XVIIIc) absorbs at 242 mμ [$\Delta\lambda_{max}$ 1 mμ from the dimethyl ether (XVIIIa)]. These results suggest that the band studied in this paper is still sensitive to steric and electronic influences, but these influences decrease as the substituents in the β-position become larger. Braude attributed the 240 mμ band in substituted *trans*-stilbenes to a "slightly displaced E′ band mainly due to the dibenzyl chromophore." In the present case the

(20) R. A. Hoffman and S. Gronowitz, *Arkiv Kemi*, **16**, 471 (1961).
(21) E. A. Braude, *J. Chem. Soc.*, 1902 (1949).

TABLE II

MELTING POINTS, ANALYTICAL DATA, AND ULTRAVIOLET ABSORPTION OF ACETYLATED PHENOLS FROM TABLE III. ALL THE COMPOUNDS WERE RECRYSTALLIZED FROM ETHANOL

Number	Empirical formula	Yield %	M.p., °C (corr.)	% Calcd. C	% Calcd. H	% Found C	% Found H	Ultraviolet $\lambda_{max.}$, mμ	Ultraviolet $\varepsilon \times 10^{-3}$
Ie	$C_{15}H_{16}O$								
IIe	$C_{19}H_{18}O_2$	46	95–96	73.53	5.85	73.3	5.84	250	1.79
IIIe	$C_{20}H_{20}O_2$	41	64–65	74.05	6.22	73.8	6.29	251	1.67
IVe[a]	$C_{20}H_{20}O_4$	57	136–137	74.05	6.22	74.1	6.28	246	1.68
Ve	$C_{21}H_{22}O_4$	61	92–93	74.53	6.55	74.5	6.55	241	3.63
VIe	$C_{22}H_{24}O_4$	58	74–75	74.97	6.86	74.5	6.84	245	1.661
VIIe[b]	$C_{23}H_{26}O_4$	58	125–126	74.97	6.86	74.8	6.84	243	1.57
VIIIe[c]	$C_{22}H_{22}O_4$	41	144–146	75.41	6.33	75.6	6.47	282	2.03
IXe	$C_{21}H_{20}O_4$	58	84–85	75.38	7.15	74.9	7.09	245	1.61
Xe[d]	$C_{22}H_{22}O_4$	61	110–111	74.97	6.86	74.9	6.82	242	1.58
XIe	$C_{23}H_{26}O_4$	54	71–72	75.38	7.15	75.1	7.23	242	1.50
XIIe[e]	$C_{23}H_{28}O_4$	57	59–60	75.76	7.12	76.2	7.61	242	1.52
XIIIe	$C_{26}H_{32}O_4$	50	46	76.44	7.90	75.8	7.87	243	1.55
XIVe	$C_{21}H_{20}O_3$	33	130–141	74.98	5.99	75.0	6.16	258	1.84
XVe	$C_{22}H_{22}O_4$	49	128–129	75.41	6.33	75.2	6.38	253	1.61
XVIe	$C_{23}H_{24}O_4$	38	137–138	75.80	6.64	75.7	6.60	247	1.70
XVIIe	$C_{24}H_{26}O_4$	36	104–105	76.16	6.93	76.2	6.98	243	1.51
XVIIIe	$C_{25}H_{28}O_4$	49	111–113	76.50	7.19	76.1	7.09	242	1.56

[a] Dipropionate, m.p. 94–95°; Calcd. for $C_{24}H_{28}O_4$: C, 75.0; H, 6.86. Found: C, 74.9; H, 7.01. [b] Dipropionate, m.p. 96–97°; Calcd. for $C_{24}H_{28}O_4$: C, 75.8; H, 7.42. Found: C, 75.6; H, 7.39. [c] In VIIIe the radical corresponding to E, in VIIIa, $CH_2CH=CH_2$ is changed to $-CH=CHCH_3$. [d] Dipropionate, m.p. 89–90°; Calcd. for $C_{24}H_{28}O_4$: C, 75.8; H, 7.42. Found: C, 75.7; H, 7.12. [e] Dipropionate, m.p. 70–71°; Calcd. for $C_{26}H_{32}O_4$: C, 76.4; H, 7.90. Found: C, 76.3; H, 7.94.

TABLE III

MELTING POINTS AND ANALYTICAL DATA OF THE FREE PHENOLS OF COMPOUNDS Ia–XVIIIa

Number	Empirical formula	Recrystallization solvents[a]	M.p., °C (corr.)	% Calcd. C	% Calcd. H	% Found C	% Found H
Ib[b]	$C_6H_{12}O_2$						
IIb	$C_{15}H_{14}O_2$	(MC)	141–142	79.62	6.23	79.5	5.97
IIIb	$C_{16}H_{16}O_2$	(MC)	134–136	79.97	6.71	79.1	6.82
IVb	$C_{16}H_{16}O_2$	(T) + (M)	188–189	79.97	6.71	79.1	6.91
Vb	$C_{17}H_{18}O_2$	(T) + (M)	179–180	80.28	7.13	79.9	7.20
VIb	$C_{17}H_{18}O_2$	(T) + (M)	173–174	80.56	7.51	80.5	7.68
VIIb	$C_{17}H_{18}O_2$	(M) + (W)	191–192	80.56	7.51	80.1	7.61
VIIIb[c]	$C_{17}H_{18}O_2$	(M) + (W)	160–162	81.17	6.84	81.7	6.79
IXb	$C_{19}H_{22}O_2$	(T)	146–147	80.81	7.86	80.1	7.92
Xb[d]	$C_{17}H_{18}O_2$	(M) + (W)	179–180	80.56	7.51	80.1	7.61
XIb	$C_{19}H_{22}O_2$	(B)	174–175	80.81	7.85	80.1	7.82
XIIb	$C_{20}H_{24}O_2$	(M) + (W)	195–196	81.01	8.16	81.01	8.06
XIIIb	$C_{23}H_{30}O_2$	(CT)	149–150	81.11	8.70	80.7	8.62
XIVb	$C_{17}H_{16}O_2$	(M) + (W)	190–192	80.92	6.39	81.2	6.51
XVb	$C_{18}H_{18}O_2$	(M) + (W)	197–199	81.17	6.81	80.5	6.80
XVIb	$C_{18}H_{20}O_2$	(M) + (W)	235–236	81.39	7.19	81.1	7.20
XVIIb	$C_{19}H_{22}O_2$	(M) + (W)	202–203	81.60	7.53	81.2	7.49
XVIIIb	$C_{20}H_{24}O_2$	(M) + (W)	190–191	81.78	7.81	81.2	7.89

[a] MC = methylene chloride, T = toluene, M = methanol, W = water, B = benzene, CT = carbon tetrachloride. [b] The only product we obtained here was the saturated compound. [c] In VIIIb the radical corresponding to E, in VIIIa, $CH_2CH=CH_2$ is changed to $CH=CHCH_3$. [d] Prepared earlier by Z. Földi, see ref. 17.

same band is mainly due to the diphenylmethane chromophore for the substances with the maximum steric hindrance to planarity with an increasing participation of the π-electrons from the two rings for the substances with less steric hindrance.

Pharmacology.—The compounds discussed in this paper have been submitted to the Allen–Doisy reaction and Rubin's uterine growth assays. The Allen–Doisy assay has been carried out under the same conditions as in a previous study using two subcutaneous injections of the substance dissolved in olive oil daily.[5] The ED_{50} values found are reported in Table IV, where it can be seen that the acetoxy compounds studied have invariably a low activity. The most active compounds are Xc and XIVc with ED_{50} of 40 and 50 γ, respectively. Hydroxy compounds have about the same activity (XVb, XVIIb), whereas the methoxy compounds have been found practically inactive.

Rubin's uterus test was carried out according to a previous study.[1] The activity is expressed as a ratio E/C, where E is the average uterus weight in the group of animals injected during three days with the compound to be studied and C the same for the group receiving three daily injections of 0.025 γ of estradiol-17β benzoate. Differences in average body weight between experimental and control groups were taken into account, using the relation between uterus dry weight and body weight shown in Fig. 3. The results are shown in Fig. 4 and 5, where they are compared with four classical estrogens, tri-*p*-anisylchloroethylene, diethylstilbestrol, estradiol-17β benzoate, and hexestrol.

TABLE IV

Compounds	Mice subcutaneously γ	No. of animals
Diethylstilbestrol	0.1	180
Estradiol-17β benzoate	.1	>1000
IIc	>1000	140
VIc	150	100
VIIIc	250	100
Xc	40	180
XIVc	50	160
XVc	140	300
XVIc	140	460
XVIIc	140	180
XVIIIc	360	180
XVb	150	140
XVIb	100	60

The pituitary gonadotrophin-inhibiting properties have also been studied. These results and a discussion of the activity ratio on different receptors will be published elsewhere.

Experimental

General.—Melting points were determined in a Tottoli melting point apparatus and are corrected. Ultraviolet absorption spectra were taken in ethanol with a Zeiss Model PMQII spectrophotometer using 1-cm. matched quartz cuvettes. Nuclear magnetic resonance spectra were obtained using a Varian Associates HR-60 system with samples in 5-mm. o.d. tubes., and resonance frequencies were determined relative to the internal standard, tetramethylsilane.

Bis-(*p*-methoxyphenyl)-alkenes (Ia-XVIIIa, Table I).—To a Grignard solution prepared from 1.1 moles of magnesium turnings, 1.0 mole of *p*-methoxyphenyl bromide, and 200 ml. of dry ether in a three-necked flask equipped with a reflux condenser, a dropping funnel, and a sealed stirrer, was added a solution of 0.45 mole of the ester dissolved in 100 ml. of dry ether during a period of 30 min. After boiling for 1 hr. with vigorous stirring, the Grignard complex was decomposed with 5 *N* sulfuric acid. The ether layer was separated, washed with saturated sodium chloride solution, and dried over anhydrous sodium sulfate. After removing the ether, the oil obtained was distilled and purified by redistillation or recrystallization. Yield, melting points, and analytical data of the eighteen compounds are shown in Table I.

Bis-(*p*-acetoxyphenyl)-alkenes (IIc—XVIIIc, Table II).—The following materials, 0.35 mole of a bis-(*p*-methoxyphenyl)-alkene, 100 g. of potassium hydroxide pellets, and 400 ml. of triethyleneglycol, were mixed in a three-necked flask equipped with a 2-dm. high glass tube, a sealed stirrer, and a contact thermometer adjusted to 210°. The mixture was heated by an electric heating mantle and was kept at 210° for 3 hr. After cooling to room temperature, the dark brown mixture was poured into 1000 ml. of water, extracted with ether, and acidified with 5 *N* hydrochloric acid. The dark brown oil which separated was taken up in ether, washed with saturated sodium chloride solution to neutral reaction, and dried over anhydrous sodium sulfate. After removing the ether, 250 ml. of acetic anhydride and a trace of concentrated sulfuric acid were added to the residue. The reaction mixture, which immediately became warm, was heated for 30 min. on a steam bath. After cooling, the mixture was poured into water and the acetylated compound, which had separated as an oil, was taken up in ether and washed twice with saturated sodium chloride solution. The solvents were driven off and the bis-(*p*-acetoxyphenyl)-alkene was distilled and recrystallized from ethanol. Yields, melting points, and analytical data of the seventeen compounds are shown in Table II.

Bis-(*p*-hydroxyphenyl)-alkenes (IIb-XVIIIb, Table III).—On heating, 0.015 mole of the pure bis-(*p*-acetoxyphenyl)-alkene was saponified on the steam bath with 50 ml. of 20% potassium hydroxide in methanol for 30 min. The free phenol was precipitated with 2 *N* hydrochloric acid, filtered, washed with water, and recrystallized. Melting points and analytical data are to be found in Table III.

Bis-(*p*-acetoxyphenyl)-cyclohexylmethane.—A quantity of 33.7 g. (0.3 mole) of cyclohexanecarboxaldehyde, prepared according to Wood and Comley,[22] was mixed in a flask with 113 g. (1.2 moles) of phenol. The mixture was stirred for 30 min. while dry hydrogen chloride was passed through it. During this time the temperature rose to 72°. The gray mass thus obtained was left

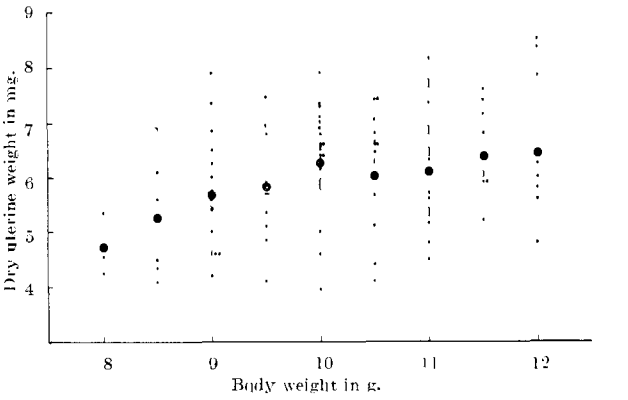


Fig. 3.—Dry uterine weight from NMRI mice after three daily injections of 0.025 γ of estradiol-17β benzoate in function of the body weight.

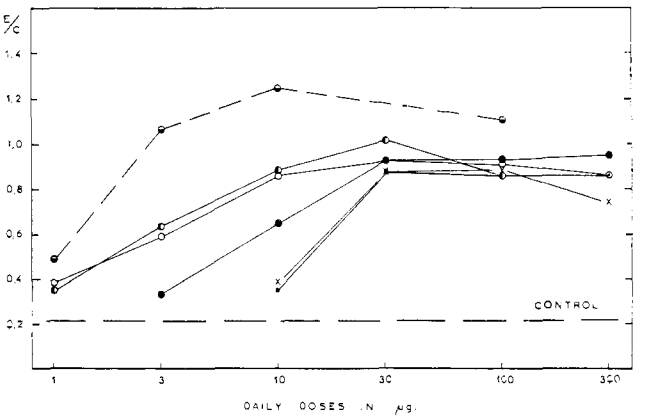


Fig. 4.—Uterus growth curves. *E/C* is the ratio of the uterus mean weight from the animals in the experimental group receiving during three days the compound to be studied and the uterus mean weight from a control group receiving 0.025 γ of estradiol-17β benzoate during three days, mean body weight of the experimental group being the same as mean body weight of the control group (see Fig. 3): ○, compound XIVc; •, compound XVIIc; ◐, compound XVc; ×, compound XVIIIc; ●, compound XVIc; ◒, tri-*p*-anisylchloroethylene.

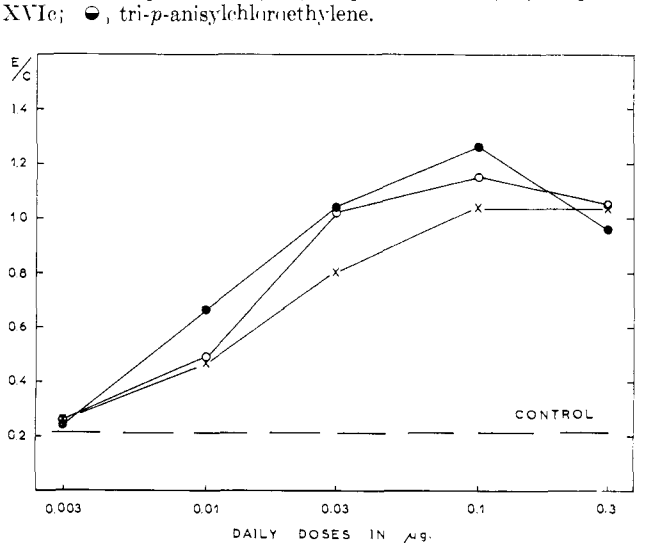


Fig. 5.—Same experiment as Fig. 4: ●, diethylstilbestrol; ○, estradiol-17β benzoate; ×, hexestrol.

(22) C. E. Wood and M. A. Comley, *J. Soc. Chem. Ind.*, **42**, 429 T (1923).

at room temperature for 5 days and was then refluxed with 250 ml. of acetic anhydride for 1 hr. The resulting clear solution was poured into 1000 ml. of water and the precipitate obtained was taken up in ether, washed with saturated sodium chloride solution, and dried over anhydrous sodium sulfate. After removing the ether the residue was distilled at 185–190° (0.1 mm.) and recrystallized twice from ethanol, m.p. 138–139°, yield 79 g. (72%), λ_{max} 227 (ϵ 1.77 × 10^4).

Anal. Calcd. for $C_{23}H_{26}O_4$: C, 75.38; H, 7.15. Found: C, 75.41; H, 7.17.

Bis-(p-hydroxyphenyl)-cyclohexylmethane.—A quantity of 18.2 g. (0.05 mole) of bis-(p-acetoxyphenyl)-cyclohexylmethane was boiled with 200 ml. of a 20% methanolic solution of potassium hydroxide for 30 min. After cooling, 200 ml. of water was added and the solution neutralized with 2 N HCl. The resulting crystalline mass was recrystallized twice from 50% ethanol, m.p. 226–228°.

Anal. Calcd. for $C_{19}H_{22}O_2$: C, 80.81; H, 7.85. Found: C, 81.0; H, 7.67.

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Synthesis and Antibacterial Activity of Symmetrical Bis-quaternaries Derived from β-Ionone and Related Compounds

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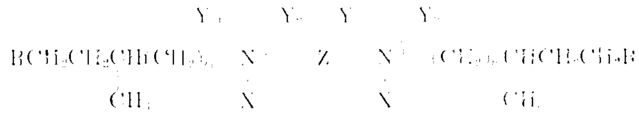
A series of new symmetrical bis-quaternary compounds, derived from alkyl substituted cyclohexenyl-, alkyl substituted cyclohexyl-, alkyl substituted cyclopentyl-, and branched aliphatic amines, have been prepared for evaluation as antimicrobial agents. Most of the compounds showed a strong *in vitro* and local *in vivo* antibacterial activity and, in addition, those derived from tetrahydroionone exhibited a limited systemic antistaphylococcal effect in mice.

Symmetrical bisquaternary compounds, whose cationic groups are derived from amines containing long chain alkyl, alkoxyalkyl, and thioalkyl moieties and are bridged by an alkylene chain, are known to exhibit germicidal activity.[3] Antibacterial activity has also been described for a series of α,ω-bis(2,2′-dipyridylamino)alkanes,[4] as well as for decamethylenebis(4-aminoquinaldinium chloride) and analogs.[5]

This paper describes the preparation and antimicrobial activity of a series of symmetrical bis-quaternary compounds of the general formula I, in which R can be an alkyl substituted cyclohexenyl, an alkyl substituted cyclohexyl, an alkyl substituted cyclopentyl, or a branched aliphatic group, and m = 0 or 1, Y_1 = alkyl, Y_2 = Y_1 or substituted alkyl, and Z represents alkylene, alkenylene, alkynylene, p-xylylene, or an aliphatic chain containing an ether, hydroxy, ester, or amide function. The new compounds were obtained by several methods, using mainly β-ionone and related compounds as starting materials.



I

Most of the bis-quaternaries exhibited, as expected, a strong *in vitro* antibacterial activity, often accompanied by good local antimicrobial activity. Unexpectedly, some of the compounds also had a limited systemic antistaphylococcal effect in mice, and it was most interesting to find that the structural requirements for this *in vivo* activity were very specific. Only those compounds of structure I, in which R was 2,2,6-trimethylcyclohexyl, m was 0 or 1, Y_1 and Y_2 were methyl, and Z ranged from ethylene to decamethylene, showed this systemic activity. One of these compounds, N,N′-bis[1-methyl-3-(2,2,6-trimethylcyclohexyl)propyl]-N,N′-dimethyl-1,6-hexanediamine bis(methochloride)[7] (II), has been tested clinically and found to be useful as a topical antimicrobial agent.

Chemistry. The new symmetrical bisquaternaries were synthesized by the methods outlined in the flow diagram for the preparation of II. Essentially, two methods were used. In one approach (method E), two equivalents of the appropriate monotertiary amine were treated in refluxing acetonitrile with an α,ω-dihalide. In the alternate route (method F), the appropriate ditertiary amine was treated with a quaternizing agent such as methyl bromide in acetone at room temperature, or methyl chloride in methanol under pressure at 100°. In two instances, the appropriate disecondary amine was directly alkylated and quaternized (method H), using methyl chloride or ethyl bromide in the presence of anhydrous sodium carbonate. Finally, a few of the bis-quaternary bromides or iodides were converted to the corresponding chlorides by treatment with freshly precipitated silver chloride (method G). The bis-quaternaries thus obtained were usually hygroscopic, and were often obtained in various hydrated crystalline forms.

The mono- and diamines (Tables I and II), used as intermediates in the preparation of the bis-quaternaries, were all new compounds, except for a few that were recently described.[8] The tertiary mono- and diamines

(1) Department of Chemical Research.

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(3) P. L. de Benneville, "Medicinal Chemistry," Vol. III, F. F. Blicke and R. H. Cox, Ed., John Wiley and Sons, Inc., New York, N. Y., 1956, pp. 114 and 122.

(4) H. H. Gadebusch and C. J. Cavallito, *Antibiot. Chemotherapy*, **7**, 549 (1957).

(5) M. Baldis, H. O. J. Collier, W. C. Austin, M. D. Potter, and E. P. Taylor, *J. Pharm. Pharmacol.*, **8**, 110 (1956).

(7) Triclobisonium chloride, Triburon®.

(8) See footnotes k (Table I) and footnote l (Table II).