219. The Isomerisation of Dienœstrol and Some Related Dienes and Indenes.

By Werner Hausmann and A. E. Wilder Smith.

Dienœstrol and some related dienes have been found to undergo cyclisation to the corresponding indenes under mild acid conditions. During the preparation of the dienes from the respective pinacols by dehydration with acetic anhydride–acetyl chloride mixtures the same indenes were also found to be formed in addition to the dienes.

During experiments on the metabolism of dienœstrol (Sahasrabudhe and Wilder Smith, *Biochem. J.*, 1947, **41**, 190; Hausmann and Wilder Smith, *Nature*, 1948, **161**, 892) unaccountable losses of œstrogenic activity were noticed when urines containing free added dienœstrol were heated under reflux in the presence of dilute mineral acids. In order to eliminate the possibility that such losses were due to interference by unknown factors in the urine or to oxidation, an aqueous suspension of dienœstrol, 1N. with respect to hydrochloric acid, was heated under reflux for 1 hour in a stream of nitrogen. From the resultant suspension, an 80% yield of an isomer, m. p. 175—176°, was isolated which proved to be identical with the œstrogen obtained by Adler and Hägglund (*Arkiv Kemi, Min., Geol.*, 1945, **19**, *A*, No. 23) by the action of boron trifluoride on dienœstrol in chloroform solution and which they designated " Indenœstrol A." This indene, 4′ : 6-dihydroxy-2-phenyl-1-methyl-3-ethylindene, has an E.D. 50 of approximately 3 µg. when tested on spayed rats by the technique used at this Institute (Wilder Smith and Williams, *J. Endocrin.*, 1947, **5**, 152). Under the same conditions dienœstrol has an E.D. 50 of about 0·7 µg., so this transformation would account for the observed losses of œstrogenic potency.

It has been found that other dilute mineral acids, such as sulphuric and phosphoric acids, are able to effect the same cyclisation under the same conditions. Also, passage of dry hydrogen chloride into a solution of dienœstrol in glacial acetic acid in the cold, or the addition of a trace of concentrated sulphuric acid to such a solution, gave a good yield of the same isomer.

Similar isomerisations occur in other suitably substituted dienes. For example, treatment of 3 : 4-diphenylhexadiene with dry hydrogen chloride in hot glacial acetic acid solution yielded 2-phenyl-1-methyl-3-ethylindene. As would be expected, the same indene was formed during the preparation of 3 : 4-diphenylhexadiene by dehydration of propiophenone pinacol with a mixture of acetyl chloride and acetic anhydride. Plentl and Bogert (*J. Amer. Chem. Soc.*, 1941, **63**, 989) have synthesised 2-phenyl-3-methyl-1-ethylindene from 2-phenyl-3-methylindan-1-one. They describe it as a bright orange viscous oil distilling under reduced pressure, but do not state a boiling point or pressure. They give, however, a correct analysis. Our material (m. p. 46—47°), on catalytic reduction in the presence of palladium charcoal, showed one reducible double bond per molecule, and on analogy with the cyclisation to the other indenes described it is assumed to be 2-phenyl-3-methyl-1-ethylindene, which would agree with the properties quoted by Plentl and Bogert (*loc. cit.*). 2 : 3-Diphenylbutadiene and acetophenone pinacol yielded 2-phenyl-3-methylindene under similar conditions.

The latter observation would explain the findings of Allen, Eliot, and Bell (*Canadian J. Res.*, 1939, **17**, 75), *viz.*, that a substance, m. p. 69—73°, was formed during the dehydration of acetophenone pinacol, but they did not identify it. 2-Phenyl-3-methylindene melts at 75—76°.

The type of cyclisation reported here would also explain the anomalies observed by Campbell and Chattaway (*Proc. Roy. Soc.*, 1942, *B*, **130**, 435), who dehydrated four pinacols with acetic anhydride–acetyl chloride mixtures and found that the resultant " dienes " only absorbed half or less of the theoretical amounts of hydrogen on catalytic reduction.

Blum-Bergmann (*Ber.*, 1932, **65**, 109; *J.*, 1935, 1020) observed that the dehydration of certain pinacols with dilute mineral acid yielded the corresponding indenes. However, propiophenone pinacol, when heated for 5 hours with 1N-hydrochloric acid, yielded no appreciable amounts of indene or diene.

Ziegler and Sauermilch (*Ber.*, 1930, **63**, 1856) found that certain phenyl-substituted allenes isomerised to the corresponding indenes in the presence of mineral acids. Dienœstrol, containing two *p*-hydroxy-phenolic groups, cyclised more readily than the corresponding hydrocarbon.

Catalytic reduction of indenœstrol A gave two isomeric indanes. The higher-melting isomer, m. p. 198—199° (decomp.), as described by Adler and Hägglund (*loc. cit.*), was comparatively inactive biologically (E.D. 50 about 200 μg.). The lower-melting *isomer* (m. p. 74°) showed about the same biological activity as the original indene (E.D. 50 about 3 μg.) and has, as far as we are aware, not been described before.

EXPERIMENTAL.

Cyclisation of Dienœstrol in Aqueous Medium.—Dienœstrol (2 g.), on being heated under reflux for 1 hour in 1N-hydrochloric acid in a stream of nitrogen, became greenish-yellow and then deposited a white precipitate after about 20 minutes. After being cooled to 0°, the product (1·88 g.) was filtered off, washed with water until neutral, treated with charcoal and acetone, and crystallised from acetone–benzene to yield a white microcrystalline powder (1·23 g.), m. p. 175—176° (Adler and Hägglund, *loc. cit.*, quote 175°). The powder readily aquired electrostatic charges which made it difficult to handle (Found : C, 80·9; H, 6·9. Calc. for $C_{18}H_{18}O_2$: C, 81·2; H, 6·8%).

The same indene was obtained in excellent yield and purity when 1N-sulphuric or -phosphoric acid was employed under the same conditions. Treatment with water only or with dilute acetic acid did not effect any cyclisation.

Cyclisation of Dienœstrol in a Non-aqueous Medium.—Dienœstrol (5 g.) in glacial acetic acid (200 ml.) was cooled to 30°, and a vigorous current of dry hydrogen chloride passed through the solution during 20 minutes. The mixture became dark green. After 40 minutes' keeping at 0°, the solvent was evaporated in a stream of nitrogen under reduced pressure, and the viscous oily residue solidified on cooling. Three crystallisations from acetone–benzene yielded the indene as before, m. p. 175—176° (3·3 g.).

A similar experiment using one drop of concentrated sulphuric acid instead of hydrogen chloride gave the same product in 82% yield. Concentrated (90%) phosphoric acid under the same anhydrous conditions in the cold left dienœstrol unchanged, but cyclisation occurred readily on 1 hour's refluxing (62% yield). Refluxing dienœstrol for 1 hour in glacial acetic acid effected no noticeable cyclisation.

Characterisation of 4′ : 6-*Dihydroxy*-2-*phenyl*-1-*methyl*-3-*ethylindene* (*Indenœstrol A*).—On warming, the indene dissolved in 2N-aqueous sodium carbonate to give a yellow solution. It could not be recovered quantitatively by acidification and ether extraction. No addition compound with maleic anhydride could be formed under the conditions required for reaction in the case of the isomeric dienœstrol (Sahasrabudhe and Wilder Smith, *loc. cit.*).

Reduction to the Indanes.—4′ : 6-Dihydroxy-2-phenyl-1-methyl-3-ethylindene (200 mg.) in absolute ethanol (10 ml.) was catalytically reduced (at 760 mm. and 20°) with hydrogen and 2% palladium–charcoal (100 mg.), 1 mole of hydrogen (18·5 ml.) being absorbed in 2 hours. Filtration and the addition of water to the filtrate yielded a product (150 mg.) as colourless needles, m. p. 198—199° (decomp.), after three crystallisations from benzene–light petroleum (b. p. 60—80°) (Found : C, 80·7; H, 7·5.

Calc. for $C_{18}H_{20}O_2$: C, 80·6; H, 7·5%) (Adler and Hägglund, *loc. cit.*, quote m. p. 196—197°). The E.D. 50 was about 200 µg. in spayed rats.

From the mother-liquor an isomeric *indane* crystallised in long, colourless needles, m. p. 74°, on the addition of more light petroleum (b. p. 60—80°) (Found : C, 80·6; H, 7·6. $C_{18}H_{20}O_2$ requires C, 80·6; H, 7·5%). The E.D. 50 in spayed rats was about 3 µg.

Diacetate. The indene (200 mg.), acetic anhydride (400 mg.), and pyridine (3 ml.) were heated on the steam-bath for 2 hours. After working up and recrystallising from benzene–light petroleum (b. p. 60—80°), the diacetate was obtained as colourless needles (120 mg., 60%), m. p. 150—151° (Adler and Hägglund, *loc. cit.*, give for their indenœstrol diacetate m. p. 150—151°) (Found : C, 75·4; H, 6·4. Calc. for $C_{22}H_{22}O_4$: C, 75·4; H, 6·3%).

Dibenzoate. 4′ : 6-*Dibenzoyloxy*-2-*phenyl*-1-*methyl*-3-*ethylindene* was prepared from the indene, benzoyl chloride, and pyridine, and after three crystallisations from benzene–light petroleum (b. p. 60—80°) was obtained as colourless needles, m. p. 176—177° (Found : C, 81·1; H, 5·7. $C_{32}H_{26}O_4$ requires C, 81·0; H, 5·5%).

The corresponding *di*-p-*nitrobenzoyloxy*-derivative was prepared from the indene, *p*-nitrobenzoyl chloride, and pyridine, and after three crystallisations a yellowish microcrystalline powder, m. p. 201—203°, was obtained (Found : N, 5·0. $C_{32}H_{24}O_8N_2$ requires N, 5·0%).

Cyclisation of 2 : 3-*Diphenylbutadiene.*—The butadiene (3·7 g.), m. p. 45—47° (Allen *et al.*, *loc. cit.*), in glacial acetic acid solution (50 ml.) was treated for 5 minutes with a rapid stream of dry hydrogen chloride and then heated under reflux for 5 hours; a green colour developed almost immediately. After removal of the solvent under reduced pressure, the residue was distilled to give two fractions : (1) b. p. 116—130°/0·3 mm. (1·55 g.), containing chiefly unchanged butadiene; (2) b. p. 130—135°/0·3 mm. (1·27 g.), m. p. 76—77°, after two crystallisations from methanol (Found : C, 93·0; H, 6·9. Calc. for $C_{16}H_{14}$: C, 93·2; H, 6·8%). Hydrogenation at 760 mm. and 20° in ethanol in the presence of 2% palladium–charcoal showed the presence of one reducible double bond per molecule. Blum-Bergmann (*loc. cit.*) and Allen *et al.* (*loc. cit.*) quote m. p. 76—78° and 75—76°, respectively.

2-*Phenyl*-3-*methylindene from Acetophenone Pinacol.*—The pinacol, b. p. 150°/0·1 mm. (32·8 g.), acetyl chloride (81 ml.), and acetic anhydride (120 ml.) were heated under reflux for 2 hours, and the solvents removed under reduced pressure. To the resultant oil, 5 vols. of hot methanol were added, and the solution filtered after being heated under reflux for 5 minutes. On cooling, some crystals and a tar separated which were filtered off, treated with charcoal and crystallised twice from methanol. White needles, m. p. 76—77° (3·9 g.), were obtained which showed no m. p. depression with 2-phenyl-3-methylindene. From the mother-liquors a somewhat viscous yellow oil was obtained, b. p. 124—129°/0·05 mm. (10·1 g.), which yielded a further 2·3 g. of the same indene on crystallisation from methanol.

2-*Phenyl*-3-*methylindane.*—2-Phenyl-3-methylindene (1·5 g.) in ethanol (50 ml.) in the presence of 2% palladium–charcoal (100 mg.) at 760 mm. Hg and 20° absorbed 169 ml. of hydrogen in 2 hours (theory for 1 mole, 163 ml.). After removal of the catalyst and the solvent, the residue was distilled to give 2-*phenyl*-3-*methylindane* as a colourless oil, b. p. 106°/0·4 mm. (1·23 g.) (Found: C, 92·0; H, 7·7. $C_{16}H_{16}$ requires C, 92·3; H, 7·7%).

Cyclisation of 3 : 4-*Diphenylhexadiene.*—The hexadiene, m. p. 100—101° (8·2 g.), was suspended in glacial acetic acid (70 ml.) and saturated with dry hydrogen chloride during 15 minutes, then heated under reflux for 1 hour. After resaturation with hydrogen chloride and refluxing for 1 hour, the solvent was removed under reduced pressure. Unchanged diphenylhexadiene was removed by addition of maleic anhydride (10 g.) and xylene (50 ml.) followed by refluxing for 30 minutes. The cooled solution was extracted twice with 50 ml. portions of water, then three times with 50 ml. portions of 2N-sodium hydroxide; the solvent was removed, and the residue distilled under reduced pressure to give a main fraction, b. p. 122—124°/0·05 mm. (5·73 g.), an orange viscous oil. On standing for one month the oil crystallised. After three crystallisations from methanol white plates were obtained, m. p. 46—47° to a colourless oil [Found : C, 92·1; H, 7·8; *M* (Rast), 218. Calc. for $C_{18}H_{18}$: C, 92·3; H, 7·7%; *M*, 234].

The same substance was isolated from the mother-liquors remaining after the preparation of 3 : 4-diphenylhexadiene from propiophenone pinacol : this pinacol (33·2 g.), b. p. 150°/0·05 mm., yielded after 2 hours' refluxing with acetyl chloride (81 ml.) and acetic anhydride (120 ml.), 3 : 4-diphenylhexadiene, m. p. 100—101° (14·0 g.; 49%), and 2-phenyl-3-methyl-1-ethylindene, b. p. 128°/0·1 mm. (8·2 g.; 28%).

2-*Phenyl*-3-*methyl*-1-*ethylindane.*—The indene (2·13 g.) and ethanol (50 ml.) in the presence of 2% palladium–charcoal (100 mg.) at 760 mm. and 20° absorbed 209 ml. of hydrogen (theory for one double bond, 204 ml.) in 4 hours. One removal of the catalyst and the solvent, 2-*phenyl*-3-*methyl*-1-*ethylindane* remained as an oil which was distilled under reduced pressure, b. p. 115—116°/0·3 mm. (1·32 g.), as an almost colourless, somewhat viscous oil (Found: C, 91·2; H, 8·5. $C_{18}H_{20}$ requires C, 91·5; H, 8·5%).

We are very grateful to the Council of the Middlesex Hospital Medical School for the provision of laboratory facilities for this work which was carried out during the tenure of the Countess of Lisburne Memorial Fellowship. One of us (W. H.) wishes to thank the Swiss Gnehm Stiftung for a scholarship held for a part of the period during which this research was carried out, and Professor E. C. Dodds, F.R.S., for a grant.

MIDDLESEX HOSPITAL MEDICAL SCHOOL,
LONDON, W. 1.

[*Received, October 4th*, 1948.]