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THE SYNTHESIS OF COUMESTROL FROM A FLAVYLIUM SALT

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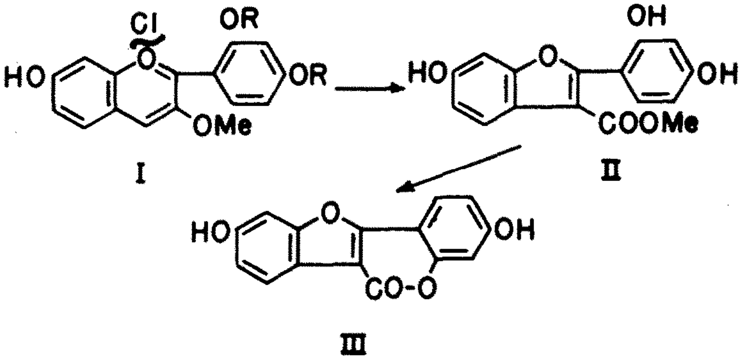
Coumestrol, an estrogenic coumarino-coumarone which occurs in alfalfa and a variety of clover species (1,2), has been synthesised by a process which is both facile and novel (3). Thus, hydrogen peroxide oxidation of 2',4',7-trihydroxy-3-methoxyflavylium chloride (I, R = H) yields the intermediate carbomethoxybenzofuran II which rapidly lactonises on acidification to give coumestrol III (yield, 50%).



The trihydroxyflavylium salt (I, R = H) is readily obtained in high yields by the condensation of 2,4-dihydroxybenzaldehyde and w-methoxy-2,4-dibenzyloxyacetophenone (m.p. 103-104°) in ethereal HCl to give the dibenzylflavylium salt, m.p. 217-218° (I, R = $C_6H_5CH_2$-). Debenzylation of this in acetic acid-concentrated hydrochloric acid then yields (I, R = H), carmine-red needles from aqueous methanolic HCl, $\lambda_{max.}$ 508, 282, 263, 240 mμ (Log ε 4.66, 4.04, 4.02, 4.35, respectively) in ethanol-0.5% HCl, R_f .74 (formic acid /3N HCl, 1:1).

A suspension of the flavylium salt (I, R = H) (10.0 g.) in warm methanol (200 ml.) is diluted with water (100 ml.) and 30% hydrogen peroxide (20 ml.). After 10 minutes the yellow-brown solution is acidified with concentrated sulfuric acid (40 ml.), heated for 15 minutes and allowed to cool to room temperature. Coumestrol precipitates as a fawn colored, crystalline powder (4.18 g.). It migrates as a single, blue fluorescent (in UV light) spot on silicic acid chromatostrips and is essentially pure. Recrystallized for analysis from a large volume of acetone or purified through its diacetate, coumestrol is obtained as a yellow, crystalline powder, m.p. > 350°. It is chromatographically, estrogenically (4) and spectrally [$\lambda_{max.}$ 343, 304, 244 (ethanol), $\lambda_{max.}$ 362, 311, 264, 244 (ethanolic sodium acetate), $\lambda_{max.}$ 387, 320, 281, 260 mμ (sodium ethylate) (5)] identical with an authentic specimen of the natural estrogen.

Calcd. for $C_{15}H_8O_5$: C, 67.1; H, 3.01.
Found: C, 67.0; H, 3.21

The diacetate of the oxidation product crystallizes from acetic acid or tetrahydrofuran-methanol as colorless needles, m.p. 229-230°, undepressed on admixture with the diacetate of natural coumestrol. Chromatographically and spectrally ($\lambda_{max.}$ 342, 327, 296, 236 mμ, ethanol), the acetates are identical.

Calcd. for $C_{18}H_{12}O_7$: C, 64.8; H, 3.43; $2CH_3CO$-, 24.4.
Found: C, 64.7; H, 3.47; CH_3CO-, 24.5 %.

Facile oxidation to 2-arylbenzofurans appears to be characteristic of flavylium salts substituted in the 3-position and a number of compounds structurally similar to coumestrol have been synthesized in this manner. The oxidation occurs very readily in buffered solutions (pH 5-6) but whether this has any biogenetic significance has not yet been established. It is not inconceivable, however, that coumestrol and other 2-arylbenzofurans may be formed in plants by oxidation of corresponding anthocyanins and, consequently, that natural compounds of this class may be more ubiquitous than hitherto recognized. A similar oxidation of anthocyanins may occur in animal organisms since the flavylium salt (I, R = H) also exhibits estrogenic properties and like coumestrol increases the uterine weight of mice.

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