#### **Short Communications**

# The Structure of Prostaglandin E, $F_1$ and $F_2$

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The smooth muscle stimulating activity discovered in sperm and prostate glands by Goldblatt <sup>1</sup> and von Euler <sup>2</sup> (and earlier refs.) is to a large extent due to a compound recently isolated by Bergström and Sjövall <sup>3</sup> and named prostaglandin E. Its composition was found to be  $C_{20}H_{34}O_5$  and more recently we have <sup>4</sup> shown that it contains one carboxyl, one carbonyl (cyclopentanone), 2 hydroxyls and one trans double bond. Reduction of the carbonyl with borohydride yielded two isomeric trihydroxy acids PGF<sub>1</sub> and PGF<sub>2</sub>. We had earlier also isolated the former from sheep prostate glands <sup>5</sup>.

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The following degradations have led to the elucidation of the structure of these compounds. Most identifications have been by non-classical means, *i.e.* by masspectrometry <sup>6</sup> (MS) of fractions isolated by gas chromatography (GLC) and the deductions from the masspectrograms have been checked through synthesis and comparison with the MS of the synthetic compounds.

Extensive oxidation with chromic acid in acetic acid at 50° yielded suberic acid in high yield with the consumption of 30 equivalents of oxidant and the formation of 4-5 mole carbon dioxide. Oxidative ozonolysis of the methyl ester acetate yielded a-acetoxy heptanoic acid from all three compounds.

Sodium hydroxide (0.5 N) at room temperature transformed PGE into a compound absorbing at 278 m $\mu$  with the loss of one hydroxyl. Oxidative ozonolysis of the methyl ester acetate of this compound

yielded suberic acid, succinic acid and ahydroxyheptanoic acid, i.e. 19 of the 20 carbon atoms had been accounted for.

Catalytic reduction of the methyl ester of PGE in ethanol gave a dihydrocompound that on treatment with alkali yielded a compound absorbing at 237 m $\mu$  with elimination of one hydroxyl. Oxidative ozonolysis yielded suberic acid and 4-keto-7-hydroxydodecanoic acid, *i.e.* all 20 carbon atoms were accounted for.

Finally catalytic reduction with platinum in acetic acid of PGE yielded among other products compound (I) that was isolated by partition chromatography, identified through GLC and MS and compared with a synthetic specimen. These data support formula II for PGE. *i.e.* 2-(6-carboxyhexyl)-3-(3-hydroxyocten-1-yl)-4-hydroxy-cyclopentanone, or in I.U.P.A.C. notation • A5(C<sub>8</sub>EIQ3) 1 (7C<sub>7</sub>X) 2EQ3Q5.

The facile dehydration of PGE with alkali is indicative of a  $\beta$ -hydroxy ketone. A tertiary  $\beta$ -hydroxyl was ruled out by the ease of acylation of the two hydroxyls in dihydro-PGE and also by NMR-data

I HOOC 
$$(CH_2)_6 \cdot C = C \cdot (CH_2)_7 \cdot CH_3$$

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kindly supplied by Dr. G. Slomp, The Upjohn Company, Kalamazoo, Mich., USA. After the elimination of the secondary  $\beta$ -hydroxyl in PGE the double bond migrates into the position indicated in (III).

The two isomeric compounds PGF<sub>1</sub> and PGF<sub>2</sub> must thus have structure IV and only differ in the steric position of the hydroxyl formed by reduction of the carbonyl. Compounds of this type do not seem to have been found in nature earlier. A cyclopentenolone nucleus occurs in the "pyrethrins" present in *Pyrethrium* flowers, cf. pyrethrolone (V).

It does not appear unlikely that the prostaglandins are representatives of a group of hormonal compounds of general importance. Their high biological activity makes pharmacological exploration of the activity of similar compounds of interest.

A full report of this work will be published in this journal.

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### Studies on Ester Sulphates

## 13. On the Enzymic Synthesis of Steroid Disulphates \*

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Pormation of monosulphates of certain steroids in microsome-free liver extracts has been reported by several workers 1-4. Evidence of the occurrence in rat liver supernatant fluid of two different steroid sulphokinases was presented by Nose and Lipmann in 1958 5. One of these enzymes, dehydroepiandrosterone sulphokinase, was also shown to react with pregnenolone and androsterone, both of which have a hydroxyl group in the 3-position. The other enzyme was capable of sulphurylating the phenolic hydroxyl group of oestrone. On the other hand, no results with bearing on the enzymic in vitro formation of disulphates of steroids have so far been reported in the literature. In the present paper, an account is given of certain findings indicating the in vitro synthesis of steroid disulphates.

Androst-5-ene-3β-17β-diol \*\*, in a final concentration ranging from 0.016 to 0.083 mM, was incubated for 120 min at 37.5°C in open test tubes, in a medium containing the following constituents: (1) 50 μl of a buffer solution containing equal parts of 0.3 M KH<sub>2</sub>PO<sub>4</sub> (pH 6.8), 0.03 M K<sub>2</sub>SO<sub>4</sub> and 0.005 M MgCl<sub>3</sub>; (2) 20 μl of 0.02 M ATP disodium salt\*\*\*; (3) 50 μl of microsome-free supernatant fluid of female rat liver, homogenized in 2-3 volumes of 0.15 M KCl containing 0.001 M EDTA (pH 7.0), centrifuged at 105 000 g for 60 min, and diluted to contain 10 μg of protein per μl; (4) 0.1 mC of carrier-free <sup>26</sup>S-labelled sulphate † Final volume of the incubation mixture: 120 μl. The steroid was added to the empty

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<sup>\*\*\*</sup> Obtained from Sigma Chemical Company, St. Louis, U.S.A.

<sup>†</sup> Obtained from the Radiochemical Centre, Amersham, England.