Elution of the peroxidase activity from a calibrated column of Sephadex G-200, equilibrated with 0.1M-NaCl in 0.02M-phosphate buffer, pH 7.0, indicated that the molecular weight of the enzyme is about 220 000.

The purified enzyme did not display a Soret band, but was strongly inhibited by cyanide and azide, as are the activities of most haemoproteins. The inhibition with cyanide was competitive with hydrogen peroxide, as is the case with conventional haem-containing peroxidases.

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Metabolism of Sodium Oestrone [³⁵S]Sulphate in the Rat

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Earlier studies on the metabolic fate of orally administered and injected sodium oestrone [³⁵S]-sulphate in the rat (Hanahan & Everett, 1950; Davis *et al.* 1950; Lewison, Levi, Jones, Jones & Silberstein, 1951) have shown that the ester undergoes appreciable desulphation with the rapid excretion of inorganic sulphate via the kidneys. Subsequently several workers have reported that mammalian liver, placental and kidney preparations are able to desulphate oestrone sulphate, the responsible enzyme being localized mainly in the microsomal fraction of the tissue cell (Pulkkinen & Paunio, 1963; French & Warren, 1967; Zuckerman & Hagerman, 1969). However, the site of desulphation of injected oestrone sulphate in the mammalian system has never been established with certainty, and a reinvestigation of the metabolism of oestrone sulphate by using new and improved techniques was therefore undertaken.

Free-ranging male and female rats receiving oestrone [³⁵S]sulphate either orally or by intravenous or intraperitoneal injection excreted approx.

70% of the radioactivity in the urine as inorganic sulphate within 48 h. The remainder of the dose was recovered mainly as ester-bound sulphate in the faeces (10–20%) and the urine (5%), although this material was not identified. In antibiotic-treated animals a similar excretion pattern was observed when the ester was administered intraperitoneally or intravenously. However, when given orally to treated animals, only 25% of the dose appeared in the urine as inorganic sulphate, the major portion being recovered in the faeces as unidentified ester sulphate (40%).

Administration of the ester to normal animals with bile-duct and ureter cannulae resulted in excretion of most of the radioactivity in the urine within 12 h. Relatively small amounts (4%) of the dose appeared in the bile, although this was shown by chromatography to consist of at least six components including inorganic sulphate oestrone sulphate and oestradiol 3-sulphate.

The organ distribution of administered oestrone [³⁵S]sulphate was examined by whole-body radioautography by the method of Curtis, Powell & Dodgson (1967). It was observed that in young male and female rats receiving the labelled ester intraperitoneally ³⁵S readily accumulated in the liver within 30 min. Subsequently ³⁵S was detected in kidney bladder and gut.

These collective findings suggest that desulphation of the ester *in vivo* is an intracellular process and that the liver is the principal organ where this occurs. This was supported by the observed rapid liberation of ³⁵SO₄²⁻ that occurred when isolated rat liver was perfused with blood containing oestrone [³⁵S]sulphate.

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