The addition of equimolar amounts of 2-hydroxy-oestradiol-17 β , 2-hydroxyoestrone or 2-hydroxy-oestriol to the incubations inhibited the enzymic formation of 3-*O*-methyladrenaline by 96%. Even at an adrenaline/2-hydroxyoestradiol-17 β molar ratio 12:1 the methylation of adrenaline by the cytosol fraction of human liver was inhibited by 93%. Similar results were obtained when noradrenaline was used as substrate. The competitive nature of this inhibition was indicated by kinetic experiments. The methylation of 2-hydroxyoestradiol-17 β to the corresponding 2- or 3-monomethyl ethers (Knuppen & Breuer, 1966) by the catechol *O*-methyltransferase of human liver was not inhibited by adrenaline or noradrenaline. Even a 20-fold molar excess of the catecholamines was without any effect.

When 2-methoxy oestrogens were added to the incubations the methylation of adrenaline to 3-*O*-methyladrenaline was also inhibited; however, this inhibition was a non-competitive one. Oestrone, oestradiol-17 β and oestriol had no effect on the methylation of adrenaline or noradrenaline.

The results obtained indicate that 2-hydroxy oestrogens, which are among the main metabolites of oestrogens in man, may have a regulatory influence on the biological inactivation of vasoconstricting substances.

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Steroid Glucuronyltransferases from the Cytosol Fraction of Human Liver

By G. S. RAO, M. L. RAO and H. BREUER. (*Institut für Klinische Biochemie, Universität Bonn, Bonn, Germany*)

UDP-glucuronate glucuronyltransferases (EC 2.4.1.17) catalyse the transfer of glucuronic acid from UDP-glucuronic acid to a large number of acceptors, resulting in the formation of water-soluble compounds.

The present investigation deals with glucuronyltransferases in the cytosol fraction (158000g) of human liver that conjugate the 16 α -hydroxyl group of oestriol and the 17 β -hydroxyl group of testosterone. Fractionation with ammonium sulphate

and filtration through Sephadex G-200 purified the oestriol-conjugating enzyme 100-fold and the testosterone-conjugating enzyme 50-fold. The two activities could not be separated from one another by conventional methods. To find out whether the two activities are due to one enzyme or to two different enzymes, several studies were carried out with the partially purified enzymes (Sephadex G-200 fraction). (1) Controlled heating (Dixon & Webb, 1964; Linn & Lehman, 1965) inactivated the two enzymic activities at the same rate. (2) Freezing and thawing also inactivated both enzymic activities at the same rate. (3) The oestriol 16 α -glucuronyltransferase had optimum pH 8.0 in tris-HCl buffer, whereas the testosterone 17 β -glucuronyltransferase did not show a well-defined optimum in the different buffers used. (4) The temperature optimum for the oestriol 16 α -glucuronyltransferase was found to be 37°C; the testosterone glucuronyltransferase revealed a plateau between 32°C and 45°C. (5) The activation energy for the glucuronidation of oestriol was 11.1 kcal/mol and that of testosterone 5 kcal/mol. (6) Oestriol did not inhibit the glucuronidation of testosterone and vice versa. (7) The mixed-substrate method (Dixon & Webb, 1964) indicated that the rate of conjugation of the different substrates when incubated together is approximately equal to the sum of the rates of the conjugation when the substrates were incubated separately. (8) The K_m value for oestriol was 11.4 μ M and that for testosterone 6.4 μ M. (9) With oestriol as substrate the K_m for UDP-glucuronic acid was 100 μ M and with testosterone as the substrate 25 μ M.

These results indicate the possible existence of two separate enzymes conjugating oestriol and testosterone. Further studies are required to characterize the two enzymic activities.

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Role of Endogenous Hormones in Uterine Binding of Oestradiol-17 β

By R. J. B. KING, A. W. STEGGLES, J. GORDON and JOANNA MARX. (*Imperial Cancer Research Fund, Lincoln's Inn Fields, London W.C.2, U.K.*)

Cytoplasm from mature uteri contains a 4S oestradiol-17 β receptor that is not present in immature animals or ovariectomized animals (Steggles & King, 1970). This indicated that oestradiol-17 β binding is influenced by endogenous