Letters to the Editor

3-Epimerisation of digoxigenin in the rat

SIR,—It has been demonstrated in several species, including the rat and man, that the metabolism of certain cardiac glycosides involves the stepwise removal of the attached sugar molecules (Lauterbach & Repke, 1960; Wright, 1962). It has also been observed (Herrmann & Repke, 1964) that the resulting genins undergo further metabolism when incubated with liver slices and among the metabolites was a component which could not be separated from the corresponding 3-epi-genin when chromatographed on several systems of paper and thin-layer chromatography. However, Herrmann & Repke (1964) were unable to separate 3-epi-digitoxigenin from 17βH-digitoxigenin and since the corresponding 17βH-isomers were not used as reference materials in the examination of the metabolites of the other genins studied, further evidence in support of metabolic 3-epimerisation has been sought.

Digoxigenin-[12-^{3}H] (2·19 μc/mg) was administered by the peritoneal route to 6 rats and the bile collected through the cannulated bile ducts for 10 hr. The dose received by each animal was equivalent to 3 μg/g body weight. Extraction of the bile with chloroform removed 15·4% of the total radioactivity present in the bile. The radioactivity not extracted by chloroform was shown to be present only in compounds of greater polarity than either digoxigenin or 3-epi-digoxigenin. The chloroform extract was separated by thin-layer chromatography into 3 components. Two of these components behaved on several systems of chromatography as digoxigenin and 3-epidigoxigenin. The ratio of these 2 components was approximately 1 : 10 respectively.

The area corresponding to 3-epi-digoxigenin was eluted from a thin-layer chromatogram of the chloroform extract. The activity of the eluate was $4{\cdot}07 \times 10^5$ dpm. To this was added unlabelled 3-epi-digoxigenin (88 mg) The recovered 3-epi-digoxigenin was recrystallised 5 times from methanol-ethyl acetate. The specific activities of the 5 yields were 4·68, 4·56, 4·35, 4·46, $4{\cdot}40 \times 10^3$ dpm./mg. The calculated activity assuming no separation was $4{\cdot}64 \times 10^3$ dpm/mg. The possibility was considered that the recovered activity was due to the presence of labelled digoxigenin which may have failed to separate from the 3-epi-digoxigenin during the course of the above re-crystallisation. In a control experiment a mixture of ^{3}H-digoxigenin (1·07 mg) and 3-epi-digoxigenin (128 mg) was repeatedly crystallised from methanol-ethyl acetate. The activity of the recovered material fell steadily, 73% of the calculated activity being lost after six re-crystallisations.

It may be concluded that in the whole rat digoxigenin undergoes rapid metabolism comparable with that observed with rat liver slices and that 3-epimerisation constitutes a major pathway in the detoxification of digoxigenin as postulated by Herrmann & Repke (1964).

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