body falls but there is still about $0.8 \mu\text{mol}$ of tyrosine left in the fat-body 24h later when the larval cuticle has tanned to a golden brown. In 4–8-day-old larvae the amount of tyrosine in the haemolymph remains fairly constant at $1 \mu\text{g}/\mu\text{l}$, but during the 'rounded-off' stage the concentration doubles, only to fall again within 24h to a value lower than that found earlier in the instar.

Since the content of tyrosine in the fat-body falls at the same time as that in the haemolymph rises, this strongly suggests that the fat-body is the source of some, if not of most, of the tyrosine which is transported via the haemolymph for use in the reactions resulting in the sclerotization of the larval cuticle. Furthermore, the fact that the content of tyrosine in the haemolymph suddenly rises during the 'rounded-off' stage suggests that during this period the rate at which tyrosine is released by the fat-body is faster than the rate at which it is taken up by the epidermal cells. Since these changes are taking place during or just after the critical period of ecdysone release (Shaaya & Karlson, 1965) it may well be that the release of tyrosine from the fat-body is under the control of this hormone.

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Species Differences in the Formation of 4-Amino-3-biphenyl β -D-Glucosiduronate *in vitro*

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Exposure of man or experimental animals to 4-aminobiphenyl generally results in the development of neoplastic disease (Melick, Escue, Naryka, Mezera & Wheeler, 1955; Walpole, Williams & Roberts, 1954; Bonser, 1952), the site of tumour formation depending on the species. In man, dog and rabbit tumours of the urinary bladder develop

whereas in rats and mice intestinal tumours or hepatomas are produced. Guinea pigs appear to be refractory to the carcinogenic action of this type of compound.

4-Aminobiphenyl is hydroxylated on the 3- or 4'-position of the nucleus or on the nitrogen atom (Bradshaw & Clayson, 1955; Uehleke, 1963), but the routes leading to the initiation of the carcinogenic process are at present unknown. The failure to find 2-amino-3-biphenyl glucosiduronate as a metabolite of the non-carcinogenic amine 2-aminobiphenyl (Gorrod & Carey, 1970) suggested that the glucosiduronic acid conjugate of 4-amino-3-hydroxybiphenyl may be implicated in carcinogenesis as suggested by Clayson, Jull & Bonser (1958) and Boyland (1963).

Condensation of potassium 3-hydroxy-4-nitrobiphenyl with methyl (tri-*O*-acetyl- α -D-glucopyranosyl bromide)uronate in alcoholic solution gave methyl (4-nitro-3-biphenyl tri-*O*-acetyl- β -D-glucopyranosid)uronate. Reduction of this material with hydrogen in the presence of platinum oxide yielded methyl (4-amino-3-biphenyl tri-*O*-acetyl- β -D-glucosid)uronate, which on methanolysis with sodium methoxide gave sodium (4-amino-3-biphenyl glucosid)uronate. This material was identical in chromatographic properties and response to detection reagents with the compound formed on incubating 4-amino-3-hydroxybiphenyl with UDP-glucuronic acid in the presence of a hepatic microsomal preparation.

4-Amino-3-biphenyl glucuronide was assayed by diazotization followed by coupling with 1-*N*-naphthylethylenediamine in acid media. The coloured pigment formed had an absorption maximum at 572nm and gave a linear relationship up to $0.5 \mu\text{mol}$ under the conditions used.

The 4-amino-3-hydroxybiphenyl-UDP-glucuronate glucuronyltransferase activity was assayed in hepatic microsomes obtained from various species. Activity decreased in the order guinea pig > rabbit > dog > hamster > mouse > rat.

Very little activity was found in the cat and no activity was detected in human foetal liver.

The results do not give a clear correlation between glucuronide-forming ability and susceptibility towards the carcinogenic action of 4-aminobiphenyl. It may be that involvement of β -glucuronidase releasing the 4-amino-3-hydroxybiphenyl from its conjugate will be a further factor in deciding species sensitivity to this compound.

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