DISTRIBUTION OF [^{14}C] α -CHLOROHYDRIN IN MICE AND RATS

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(Received 25th January 1972, accepted 18th February 1972)

Alpha-chlorohydrin (3-chlor-1,2 propanediol) possesses antifertility properties when administered to male rats, hamsters, guinea-pigs, rams and monkeys (Ericsson, 1968; Ericsson & Norland, 1970). The compound is thought to exert its action on the epididymis, since high doses of α -chlorohydrin cause vascular disturbances in the proximal part of the epididymis (Gunn, Gould & Anderson, 1969; Samojlik & Chang, 1970; Kreider & Dutt, 1971). There is also a time delay between administration of the drug and the onset of inhibition of fertility (Ericsson, 1968; Turner, 1971). Impaired sperm motility which has been observed (Coppola, 1969; Samojlik & Chang, 1970), has been thought to be due to oxygen deficiency in the epididymis following vascular trauma. Coppola (1969) and Gunn *et al.* (1969) suggested that α -chlorohydrin might act as a metabolic antagonist to glycerol in phospholipid metabolism. Erickson & Bennet (1971) observed impaired sperm migration into the Fallopian tubes of rats. Alpha-chlorohydrin has also been shown to act as an alkylating agent *in vitro* (Jones, Davies, Edwards & Jackson, 1969), and the fertility of boar semen is inhibited when the drug is added *in vitro* (Johnson & Pursel, 1972; Crabo, unpublished).

However, the exact mechanism of action of α -chlorohydrin *in vivo* is still not known. The surprising observation that the drug does cause sterility in rats but not in mice (Ericsson, 1968; Samojlik & Chang, 1970) led us to investigate the whole body autoradiography distribution of α -chlorohydrin in rats and mice.

Labelled [^{14}C] α -chlorohydrin, with a specific activity of 2 mCi/mm, was synthesized by New England Nuclear Corporation, Boston, Mass. Six albino male mice (NMR I strain, 25 g body weight) received an intravenous injection of 0.55 mg of the compound and six albino male rats (Sprague Dawley strain, 250 g body weight) were given 5.5 mg by the same route. After survival times of 5, 20 and 60 min, 4 and 24 hr, and 4 days, the animals were anaesthetized with chloroform and frozen in a mixture of hexane–solid CO_2 at -70°C . Sagittal 20- and 60- μm sections were cut from the animals on a heavy microtome, collected on Scotch tape and autoradiographed on X-ray films, according to the method described by Ullberg (1954).

The resulting autoradiograms revealed a marked difference in the distri-

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bution of radioactivity in the epididymis of mice and rats. Twenty-four hours after injection of the labelled compound, the radioactivity in the epididymis of the mouse only slightly exceeded that in the blood (Pl. 1, Fig. 1) while, in the rat, the ^{14}C -concentration in the cauda epididymidis was one of the highest in the body (Pl. 1, Fig. 2). This selective accumulation in the rat epididymis was still more pronounced 4 days after injection and high amounts of radioactivity were also seen in the contents of the ductus deferens (Pl. 1, Fig. 3). No accumulation of ^{14}C was seen in the accessory sexual glands but the radioactivity levels approximated to those found in the blood in both species.

There were high amounts of ^{14}C in the liver and bile of both species shortly after injection but judging from the ^{14}C -concentration in the kidney and urinary bladder 60 min after administration, the kidney seemed to be the main organ of excretion.

In both mice and rats, high amounts of radioactivity were observed in the sebaceous glands 24 hr and 4 days after injection. In the mouse, the radioactivity concentration was also high in the thymus and Harder's gland.

The present investigation showed species differences in drug distribution between mice and rats and thus helps to explain why an antifertility effect is seen in rats but not in mice (Samojlik & Chang, 1970). It was also shown that α -chlorohydrin, or metabolites of α -chlorohydrin, are rapidly secreted into the lumen of the corpus and cauda epididymis. This explains why ligation of the corpus epididymis did not influence the antifertility effect of the drug in the experiments of Turner (1971). Very low radioactivity was seen in rat caput epididymidis where a high dose of the drug has been reported to cause epithelial necrosis (Samojlik & Chang, 1970).

The observed accumulation of radioactivity in the epididymis is in disagreement with the conclusion of Kirton, Ericsson, Miller, Cornette, Forbes & Duncan (1969) that the concentration of the drug in the reproductive fluids follows that of the blood but on a lower level. Their conclusions, however, were based on measurement of α -chlorohydrin in seminal plasma.

The results of the present investigation are not contrary to the suggestion that α -chlorohydrin might act as an alkylating agent in the epididymis (Jones *et al.*, 1969). The delay in time (48 hr) observed between the administration of the drug and the drop in fertility (Turner, 1971) could represent the time necessary for alkylation since the time required for the passage of spermatozoa through the epididymis is apparently not involved. However, this does not exclude other possible mechanisms and it still remains to be shown that metabolites of α -chlorohydrin or its incorporation in phospholipids, as was suggested by

EXPLANATION OF PLATE 1

FIG. 1. Whole body autoradiograph of a male mouse 24 hr after intravenous injection of [^{14}C] α -chlorohydrin. White areas correspond to high amounts of radioactivity. Note the low concentration of radioactivity in the epididymis.

FIG. 2. Part of a whole body autoradiograph of a rat 24 hr after intravenous injection of [^{14}C] α -chlorohydrin. Note the high concentration of radioactivity in corpus and cauda epididymis.

FIG. 3. Detail of a whole body autoradiograph of a rat 4 days after intravenous injection of [^{14}C] α -chlorohydrin. Note the high concentration of radioactivity in the cauda epididymidis and in the lumen of ductus deferens.

Coppola (1969), are not involved. Such investigations are now being carried out.

This investigation was supported in part by the Swedish Medical Research Council and also performed pursuant to Contract No. NIH-70-2056 with the National Institutes of Health, Department of Health, Education and Welfare.

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