Dose dependent disposition of [^{14}C]-ICI 89,406 in the male rat: Is this due to a capacity limited excretory pathway for the drug into bile?

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The metabolism and excretion of [^{14}C]-ICI 89,406 (1-(2-[^{14}C]-2-cyanophenoxy)-3-β-(3-phenylureido) ethylamino-2-propanol) a β-adrenoreceptor antagonist, has been studied in the male rat and using the isolated perfused rat liver. [^{14}C]-ICI 89,406 was dosed to rats orally (5 or 50 mg/kg), i.p. (5 mg/kg) and i.v. (0.6 mg/kg). The liver preparation was perfused with whole blood and ICI 89,406 (10 mg) introduced into the perfusate as a bolus injection.

Following oral administration of ICI 89,406 to the rat (5 mg/kg) urinary and faecal excretion accounted for 5.8 ± 0.7% (mean ± s.d.; $n = 3$) and 96.4 ± 2.1% of the dose. When the dose was increased to 50 mg/kg urinary and faecal excretion accounted for 14.0 ± 4.0% and 78.4 ± 2.4% of the dose. The proportions of unchanged drug and metabolites in urine were independent of the dose. The unchanged drug accounted for 81.6% of the faecal components after a dose of 5 mg/kg but this was reduced to 47.8% when the dose was increased to 50 mg/kg ICI 89,406.

The proportion of two polar metabolites increased from 6.4% to 23.5% of the faecal radioactivity with increasing dose.

The mean clearance of [^{14}C]-ICI 89,406 from the isolated perfused rat liver was 4.1 ± 1.7 ml/min and the extraction ratio 0.34 ± 0.05. The fraction, F, of an 'hepatically' administered dose reaching the systemic circulation was 0.65 suggesting that ICI 89,406 undergoes a modest degree of 'first pass' metabolism. From in vivo studies F was calculated to be 0.68 from the ratio of unchanged drug excreted in urine after i.p. and i.v. administration. This is in good agreement with the perfused liver studies. Bile contained 52% of the radioactivity introduced into the perfusion system. ICI 89,406 and its glucuronide conjugate accounted for less than 10% of the radiolabelled components. This was also seen when the dose to the perfused liver was reduced to 75 µg.

The dose dependent disposition of ICI 89,406 in the male rat is apparently due to a limited capacity to excrete ICI 89,406 into bile either as a conjugate or unchanged drug. Studies in the perfused rat liver demonstrated that excretion of ICI 89,406 and its conjugate in bile is a minor route of elimination and may not account for the dose dependent disposition of this drug in vivo. ICI 89,406 undergoes conjugation with glucuronicacid thus the dose dependent pathway seen after oral administration of ICI 89,406 may be due to metabolism in the gut wall. As the dose is increased and a greater fraction of the dose is absorbed unchanged then a greater fraction of the dose is available for oxidative metabolism.

Changes in dopamine receptor status after denervation or chronic receptor stimulation

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It has been shown that denervation or stimulation or striatal dopamine (DA) mechanisms can lead to changes in DA receptor status (see Cross, Longden, Owen, Poulter & Waddington, 1978; Quik & Iversen, 1978). It has also been shown that manipulation of DA mechanisms in the extrapyramidal system can influence DA metabolism (as measured by homovanillic acid) in an anatomically distinct area of the mesolimbic system (Costall & Naylor, 1979). Therefore, the present studies were designed to investigate possible changes in extrapyramidal, mesolimbic and cortical

DA receptor status following destruction of the DA pathways to the extrapyramidal and mesolimbic areas, and after chronic striatal stimulation by DA. Changes in haloperidol receptor binding (Leysen, Tollenaere, Koch & Laduron, 1977) and in the activity of DA stimulated adenylate cyclase (Kebabian, Petzold & Greengard, 1972) were determined in the striatum (CP), tuberculum olfactorium (TUO), nucleus accumbens (ACB) and frontal cortex (FC) following denervation of the ascending DA pathways in the lateral hypothalamus (LH) (8 µg/4 µl 6-hydroxydopamine; Costall, Naylor & Pycock, 1975), and haloperidol binding was determined following chronic receptor stimulation caused by repeated intrastriatal DA administration (Costall, Naylor & Pinder, 1974). DA concentrations in the CP, TUO and ACB were also determined.

Male Sprague-Dawley rats (250 ± 25 g) with unilateral LH lesions, and exhibiting a maximum contralateral circling response to apomorphine, were selected

for the biochemical studies. Thirty days after denervation (CP DA depleted by 84%) striatal DA receptor density, as assessed using [^{3}H]-haloperidol binding, was elevated by 34% (range 26–28%, $P < 0.001$); the activity of DA stimulated adenylate cyclase in striatal homogenates was unchanged. A 17% increase in ACB DA receptor density was recorded but this did not achieve significance ($P > 0.05$); no changes were observed in the TUO or FC. The LH lesions caused 77–86% depletions in ACB and TUO DA.

One or three days of treatment with intrastriatal DA (100 µg daily) failed to alter the number of haloperidol binding sites in the CP, TUO, ACB or FC. However, 7 days of treatment caused a 47% decrease (38–58%, $P < 0.001$) in haloperidol binding in CP tissue. Whilst there was also a tendency for the number of haloperidol binding sites to be reduced in both the ACB and TUO after 7 days of intrastriatal DA, this never achieved significance (range 11–31%, $P > 0.05$). The number of binding sites in the FC remained unchanged.

The data indicates a differential response of the receptor population in the striatal and mesolimbic areas to denervation: the number of haloperidol binding sites were significantly increased in the CP but not in the ACB or TUO. Also, the data provides the first evidence that chronic stimulation of DA receptors in the CP by DA can reduce the number of DA receptor sites in this area independent of significant changes in receptor numbers, as indicated by haloperidol binding, in mesolimbic or cortical areas.

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Effects of benzodiazepines and barbiturates in a GABA-dependent animal model: interactions with muscimol in the globus pallidus

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Benzodiazepines can interact with γ-aminobutyric acid (GABA)-mediated neurotransmission (Costa, Guidotti, Mao & Suria, 1975; Haefely, Kulcsar, Moehler, Pieri, Polc & Schaffner, 1975). Their ability to potentiate GABA at postsynaptic receptor sites might underlie some of their pharmacological actions (Haefely, 1977; MacDonald & Barker, 1978). We have investigated the effects of benzodiazepines and bar-

biturates on GABA-mediated neurotransmission in the globus pallidus (GP) using an *in vivo* animal model (Crossman, Lee & Slater, 1977).

Female Sprague-Dawley rats (180–200 g) were fitted with a chronically-implanted electrode for stimulating the neostriatum and a cannula for microinjection into the ipsilateral GP. Electrical stimulation of the neostriatum causes a contralateral head-turn. The time taken for a 90° head-turn is measured. GABA antagonists shorten the response time but the GABA agonist muscimol increases it (Crossman et al., 1977).

Benzodiazepines (administered either i.p. or in GP) resembled GABA agonists and caused slowing of the head-turn. The doses required (i.p.) to increase the response time by 50% (ED$_{50}$) were: chlordiazepoxide 10.8 mg/kg; diazepam 3.7 mg/kg; lorazepam 1.5 mg/kg and clonazepam 0.6 mg/kg. Diazepam no longer slowed the head-turn after twice daily injections of 3 mg/kg for 4 days. The sedation also disappeared after a few days.