**Species differences in the plasma protein binding of desipramine**

SIR,—Species differences have been demonstrated for the plasma protein binding of acidic drugs (Anton, 1960; Sturman & Smith, 1967), but to our knowledge such information is not available for basic drugs. It is generally accepted that only the unbound portion of a drug is available for pharmacological activity. The unbound portion also is available to the liver for metabolism and appears in the ultrafiltrate in the kidney glomeruli. Hence, the degree of protein binding can be of great quantitative significance in the pharmacological (therapeutic as well as toxic) action of drugs. Marked species and strain differences have been reported for the pharmacological effects of one group of basic drugs, the tricyclic antidepressants (Brodie, 1965). The work now reported was undertaken to determine the degree of plasma protein binding of desipramine in various species.

Blood from the various species was collected in tubes containing heparin sodium and centrifuged at 2,000 rev/min. If analyses were not made the same day, the plasma was stored at 4° for 15–24 hr. The plasma was incubated at room temperature (21–24°) with [$^3$H] desipramine (1·1 $\mu$M, 14·7 mc/mmole, chromatographically pure) for 60 min. The degree of protein binding was determined by ultrafiltration at room temperature (Schanker & Morrison, 1965). [$^3$H]Desipramine was determined in the ultrafiltrate and in the plasma by extraction of an alkalinized aliquot with the toluene scintillation mixture. Appropriate corrections were made for the minimal quenching found in the samples.

The species differences in the plasma protein binding of desipramine are as follows.

| Species | Unbound desipramine % |
|---|---|
| Man .. .. .. | 8·6, 10·5, 10·0, 10·8, 9·0 |
| Rat .. .. .. | 10·6, 9·9, 6·0, 6·0, 7·4 |
| Rabbit .. .. .. | 13·8, (14·2), 16·1, 15·3, 16·4, 15·9 |
| Dog .. .. .. | 2·0, 4·0, 0·7, 3·8, 4·0 |
| Cat .. .. .. | 1·5, 1·8, 2·1, 3·2 |

Each value is the mean of at least two duplicate determinations of individual plasma samples for all species except the rat where the values represent results from a pool of 4–6 rats. The values were determined at a desipramine concentration of 1·1 $\mu$M. The value of 14·2 was obtained from plasma from a rabbit 1 hr after intravenously administered desipramine, 10 mg/kg. The desipramine was determined by the method of Hammer & Brodie, 1967. The *in vitro* and *in vivo* values correlate very well.

It is interesting to speculate whether the reported species differences in pharmacological effect of tricyclic antidepressants could be related to the degree of protein binding.

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## The dual mode of action of histamine in the cat isolated tracheal chain

SIR,—In the cat isolated tracheal chain, a fully relaxed preparation *in vitro* (Hawkins & Paton, 1944), histamine antagonizes non-specifically the contractions induced by acetylcholine (Akcasu, 1952) and potassium chloride (Akcasu, 1959). Since in the isolated heart histamine effects have been attributed to a direct interaction with mycocardial receptors (Mannaioni, 1960), or to an indirect action based on the release of catecholamines (Went, Szucs, & Feher, 1954), it is of value to establish the mechanism of histamine relaxation of cat tracheal chains.

Tracheal rings of cats, untreated or pretreated with reserpine (0·5 mg/kg, injected intraperitoneally 48 and 24 hr before the experiment), were prepared according to the modification by Akcasu (1959) of the method described by McDougal & West (1953), and studied in Tyrode solution aerated with oxygen 95% and carbon dioxide 5% at 37 ± 0·5°. A dose of carbamylcholine chloride (carbachol), chosen from dose-response curves to produce a 60 to 85% of maximum contraction in a 2-ring chain, was left in contact with the tissue for 6·5 min. Histamine dihydrochloride was added for the last 1·5 min of this time. When antagonists were used they were added with the carbachol. All doses are expressed as μg of salt per 30 ml of bath volume. Responses, magnified 16 times, were recorded on a smoked drum kymograph with an auxotonic pendulum lever (Paton, 1957). Statistical calculations were made according to Snedecor (1957).

Only 7 of the 19 preparations taken from untreated cats showed a relaxation to 50 μg of histamine while all relaxed to 100 μg (Table 1). The relaxation was dose-dependent and reached a maximum with 200 μg of histamine. The relaxing action of histamine was only partially prevented by mepyramine maleate (40 μg). Similarly pronethalol (40 μg), a dose which fully blocked the relaxing effect of 1-(−)-noradrenaline bitartrate (2·5 to 10 μg) was only partially effective against histamine. Neither inhibitor, in the concentrations stated, altered

TABLE 1. DEPRESSION IN MM BY HISTAMINE (H) OF CARBACHOL[1]-CONTRACTIONS[2] OF CAT TRACHEAL RINGS.

| H-Dose: μg/30 ml | 50 | 100 | 200 | 400 | 800 | 1600 | Slope ± s.e. |
|---|---|---|---|---|---|---|---|
| | | | Untreated | | | | |
| Mean ± s.e. (n) | −19·3 ± 4·16 (7) | −28·2 ± 4·97 (19) | −43·5 ± 6·48 (19) | −44·2 ± 6·79 (13) | | | 30·35 ±10·81 |
| | | | Reserpine-pretreated | | | | |
| Mean ± s.e. (n) | | | | −9·7 ± 2·06 (6) | −18·3 ± 2·97 (7) | −33·0 ± 5·24 (7) | 39·35 ± 8·96 |

(1) Mean dose: 1·41 ± 0·081 μg/30 ml (2) Mean contraction height: 92·7 ± 3·45 mm