The conformations of cholinergic agonists

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We have been determining the conformations of nicotinic and muscarinic agonists by X-ray diffraction analysis of crystals of the compounds. By correlating the observed conformations of agonists which selectively activate specific receptors, and by considering possible perturbations of the conformations observed in crystals, it is possible to determine a unique conformation for any specific class of compounds. A correlation of the crystal structures of the nicotinic agonists acetylcholine, 1,1-dimethyl-4-phenyl piperazinium, nicotine, lactoylcholine and acetyl-α-methylcholine allows specification of the conformation of such agonists at nicotinic receptors. It appears that the ganglionic receptor can accommodate a longer molecule than the motor end plate receptor. A similar correlation of the crystal structure analyses of the muscarinic agonists muscarine, 2-methyl-4-trimethylammoniummethyl-1,3-dioxolan, acetyl-β-methylcholine, acetyl-α-methylcholine and *trans*-acetoxy cyclopropyl trimethylammonium allows the active conformation at muscarinic receptors to be specified. The absolute configuration of potent muscarinic agonists whose stereoisomers have been separated and tested is entirely consistent. It appears the conformation of acetylcholine relevant to the nicotinic junction and to the muscarinic junction is the same, but the molecule fits on the receptors in different ways.

The conformations of muscarinic antagonists

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We have determined the conformations of hyoscine hydrobromide and hyoscyamine hydrobromide by X-ray diffraction analysis of crystals of the compound. The two observed conformations are almost identical though the crystal packing is completely different. A comparison of these conformations and that of the muscarinic antagonist quinuclidinyl benzilate analysed by Carlstrom and Meyerhöffer (1969) shows that the three muscarinic antagonists whose structures are known are very similar. By using this evidence on the preferred conformation of muscarinic antagonists and one rule about the preferred conformation of an organic functional group, it is possible to specify the required conformation of muscarinic antagonists and the chemical groups necessary for potent antagonistic activity. The conformations of many compounds in this class can be predicted in this way.

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Saturation effects in the uptake of decamethonium in skeletal muscle

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Decamethonium has been found to accumulate in the end-plate region of mice, but no evidence of saturation was found (Waser, 1965). In rats which have been injected with labelled decamethonium the compound enters muscle fibres in the

end-plate region (Creese & Maclagan, 1967) and remains for long periods (Taylor, Dixon, Creese & Case, 1967).

Rat diaphragms were soaked in solutions which contained varying concentrations of decamethonium-(^{3}H-methyl) chloride, and after 1 h the muscles were washed for 10 min in physiological saline, frozen, sliced into strips 1 mm wide and counted by scintillation methods (Taylor, Creese, Nedergaard & Case, 1965). At concentrations of 10–100 μM there was a peak uptake in the strip which contained the band of end-plates, and the radioactivity in the tissue increased linearly with time. The uptake at the end of the fibres was much smaller.

At lower concentrations of decamethonium the peak uptake was progressively reduced, and at a concentration of 0·01 μM the uptake at the end-plate region was no different from that at the end of the fibres. Saturation could be demonstrated by plotting the uptake as a clearance (ml/g) against concentration of decamethonium, and half-saturation occurred at 2·5 μM. Similar curves have been obtained with denervated guinea-pig muscle, and also with rat diaphragms which were depolarized in solution containing potassium methyl sulphate (for example, England, 1969).

At high concentrations the peak uptake (when expressed as ml/g) was again reduced and the kinetics resembled those of a carrier-like system. If the results in rat muscle are interpreted in terms of receptors then at least two sites are necessary, a high-affinity site with half-saturation at 2·5 μM plus a low-affinity transport system with half-saturation at approximately 2 mM.

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Apparent correlations between pA_2 and pD_2' values in a group of drugs with antihistaminic and anticholinergic properties

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A group of seventy-five substances was tested on the guinea-pig isolated ileum for antihistaminic activity and on the rat isolated intestine for anticholinergic activity. The majority of the substances were newly synthetised antihistaminic substances, belonging to various chemical classes; a few were classical antihistaminic or anticholinergic drugs. For sixty-six substances both pA_2 and pD_2' values could be calculated with respect to the histaminergic system and for sixty-nine substances with respect to the cholinergic system. The pA_2 and pD_2' values are measures of the affinity of the drug to the specific and to the non-competitive receptors, respectively (van den Brink; 1969).