

TREMOR INDUCED BY TREMORINE (1, 4-DIPYRROLIDINO BUT-2-YNE) AND ITS ANTAGONISM BY CERTAIN ANTIHISTAMINICS

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Everett (2) made revolutionary advances in the field of Neuropharmacology by introducing tremorine-a 'Parkinsonimimetic' agent. It exerts unique action in several species of laboratory animals producing centrally mediated fine fast tremor and peripheral parasympathetic stimulation causing salivation, lacrimation, diarrhoea, urination and hypothermia. These effects can be antagonised by antiparkinson's drugs (4, 6 and 1). Tremorine test has become a useful tool for the evaluation of potential antiparkinson's agent (7). In the present study an attempt has been made to evaluate the possible antitremorine action of certain antihistaminic compounds.

MATERIALS AND METHODS

The anti-tremor property of antihistaminics has been carried out in albino rats of both sexes. Some of these drugs possess both antihistaminic and antiserotonin property e.g. cypheptadine and promethazine hydrochloride. On the other hand diphenylpyraline hydrochloride is purely an antihistaminic agent (Personal communication from Dr. S.D. Patel, 1963).

CONTROL EXPERIMENT

Control experiments were carried out in order to find out the challenging dose of tremorine in several groups of rats by rotating rod method of Stone (11). Doses of tremorine ranging from 2.5 to 15.0 mg/Kg were administered intraperitoneally in eight groups of animals. Within 10 minutes of administration, severe tremors, profuse salivation and mucus secretion spreading up to neck and jaw and diarrhoea were noted. The resulting tremor was sufficient enough to impair the motor capacity. The animals were not able to remain on the slowly rotating rod (5 rotations per minute) for a period of one minute. It was seen that untreated rats maintained their balance and kept their hold whereas tremorine treated rats invariably fell down within a minute at 15 mg/Kg dose level. In some of the animals, the weakness and akinesia were so marked that it was impossible for them even to sit and maintain their balance. From this experiment it was decided to use 15 mg/Kg of tremorine intraperitoneally for producing tremor and other effects in all our subsequent investigations.

Groups of animals were pretreated with test drugs subcutaneously 30 minutes prior to the rotating rod test. Tremorine was given 20 minutes before the test. Antitremor effect of

various compounds was measured by determining their ability to restore the capacity of tremorine injected rats to remain on slowly rotating rod during one minute period. The median effective dose (ED_{50}) was determined for each drug by the method of Miller and Tainter (10). The incidence of salivary secretion and diarrhoea was also noted.

STANDARD DRUG

Atropine sulphate was chosen as standard drug. Atropine has got powerful antitremorine and anti-cholinergic properties (3). Atropine sulphate afforded only 20% protection against tremorine induced tremor at 0.4 mg/Kg. At a dose level of 8 mg/Kg it completely antagonised the tremor, salivation and diarrhoea and 100% protection was recorded. The (ED_{50}) obtained from such experiments was 1.30 ± 0.291 mg/Kg. This dose was taken as unity and subsequently the potency ratio of other drugs have been calculated in respect of atropine.

RESULTS

Cyproheptadine :

The data obtained in the experiments demonstrate that cyproheptadine possesses antitremor activity. Seven groups of animals were employed for this study. The dose of 0.5 mg/Kg of cyproheptadine did not show any protective action. Ninety percent protection was obtained with a dose of 10 mg/Kg. It was observed that the tremors persisted even at the highest dose employed (10 mg/Kg). The peripheral effects were not influenced with this drug.

Diphenylpyraline Hydrochloride :

Diphenylpyraline hydrochloride antagonised completely the tremorine-induced effects. At a dose level of 12 and 14 mg/Kg the tremor was abolished completely which was even visually apparent. Both diarrhoea and salivation were not observed at these dose levels signifying thereby that it has got marked anticholinergic effects. Eight groups of animals were treated with varying dose (1 to 14 mg/Kg) of this compound. With the 2 mg/Kg dose only 10% protection was observed whereas 90% protection was obtained with 14 mg/Kg. The calculated (ED_{50}) found for the drug was 5.1 ± 0.712 mg/Kg.

Promethazine Hydrochloride :

Promethazine hydrochloride has marked antitremor property. Nine groups of animals were subjected for study. The dose varied from 1—20 mg/Kg. At 1 mg/Kg the compound did not have any influence either on tremor or cholinergic effects. Ten percent protection was noted at 2 mg/Kg. At 16 and 20 mg/Kg dose level, both tremor and cholinergic effects of tremorine were completely controlled. The (ED_{50}) for promethazine obtained was 7.8 ± 1.344 mg/Kg. The results are represented in the following graphs (Fig. 1, 2, 3, & 4).

TABLE I

Showing comparison of anti-tremorine potency of drugs with atropine

Sl. No.	Name of drug	ED ₅₀ in mg/Kg with S.E.	Potency ratio
1.	Atropine Sulphate	1.30 ± 0.291	1.0
2.	Cyproheptadine	3.2 ± 0.603	0.406
3.	Diphenylpyraline Hydrochloride	5.1 ± 0.712	0.255
4.	Promethazine Hydrochloride	7.8 ± 1.344	0.167

Although cyproheptadine is more active than diphenylpyraline and promethazine hydrochloride in antagonising the tremors yet the compound did not have any influence on the peripheral cholinergic effects. The potency ratio for cyproheptadine, diphenylpyraline and promethazine in decreasing order was 0.406, 0.255 and 0.167. Both Diphenylpyraline hydrochloride and Promethazine Hydrochloride almost completely antagonised the tremors as well as the peripheral effects of tremorine at higher dose levels.

DISCUSSION

Since these antihistaminics drugs successfully antagonised the both effects of tremorine (tremors and cholinergic effects) one may consider a possible role of histamine and acetylcholine in the tremorine-induced tremors. The biochemical and other studies indicate that histamine and acetylcholine metabolism plays a definite role in tremorine induced effects (14,8). Ungar and Witten (14) have shown the relation between histamine concentration and tremorine induced tremors. The challenging concept of Mc Geer *et al* (9) that there is equilibrium within the brain between two groups of neurochemical substances (i) Serotonin and Catecholamine (ii) Acetylcholine and histamine and any disequilibrium between these will result in Parkinson's disorder; also supports the involvement of histamine in the causation of Parkinsonism. Friedman and Everett (7) have also speculated the efficacy of antihistaminics in Parkinson's disease on the basis of their antihistaminic action. Perhaps these agents combat the tremorine-induced effects by antagonising the raised histamine level caused by tremorine. Amongst the compound studied, cyproheptadine and promethazine also possess antiserotonin property. Stern and Gasparovic (12) have shown that lack of serotonin is responsible for the genesis of tremorine-induced tremor. Friedman *et al* (5) have also demonstrated a progressive rise in 5HT level after tremorigenic dose of tremorine in the rats brain stem. If 5HT plays a role as a mediator in tremorine-induced tremor, it is just probable to ascribe the antitremor effect of compounds having antiserotonin property to their antagonism towards 5HT. The antagonism of

cholinergic effects of tremorine by these compounds can be explained on the basis that these compounds possess anticholinergic effects (13).

SUMMARY

Both promethazine and diphenylpyraline hydrochloride antagonised the tremors and peripheral cholinergic effects of tremorine. On other hand, cyproheptadine has anti-tremor action only. The present study may provide a basis for the use of these agents in clinical cases of Parkinson's disorder. Perhaps it may be worth while to conduct a controlled clinical trial with these agents in Parkinsonism.

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