Brit. J. Pharmacol. (1954), **9**, 218.

A COMPARISON OF THE PERIPHERAL PARASYMPATHOLYTIC AND AUTONOMIC GANGLION BLOCKING ACTIVITIES OF METHANTHELINE ("BANTHINE") AND PROPANTHELINE ("PRO-BANTHINE") WITH ATROPINE AND HEXAMETHONIUM

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(RECEIVED MARCH 6, 1954)

Originally prepared by Burtner and Cusic (1943), methantheline (β-diethylaminoethyl xanthene-9-carboxylate methobromide) has been studied pharmacologically by several workers, notably Hambourger, Cook, Winbury, and Freese (1950), Lehman and Knoeffel (1944), Chittum, Longino, Metcalf, and Grimson (1949), Longino, Grimson, Chittum, and Metcalf (1950), and Lyons, Reeves, and Grimson (1950). Their work shows that with progressively increasing doses and concentrations of methantheline there appears first an atropine-like action, followed, at a higher dosage range, by autonomic ganglion blockade and finally, in relatively high doses, by a curare-like action.

Propantheline (2′-di*iso*propylaminoethyl xanthene-9-carboxylate methobromide) is the isopropyl analogue of methantheline and differs from it only in degree of activity (Hambourger, 1952), its actions being qualitatively similar.

Although methantheline and propantheline seem to have considerable atropine-like activity, there have been few comparative studies with atropine in animals. It is the purpose of the present work to provide such a study. At the same time a comparison of the autonomic ganglion blocking activities, and of the curare-like activities, of the two drugs has been made. The local anaesthetic activities have also been investigated.

METHODS

The substances used were methantheline bromide, propantheline bromide, hexamethonium bromide, atropine sulphate and acetylcholine chloride. Doses and concentrations, and comparisons of activity, are given throughout this paper in terms of these salts.

Comparisons of Peripheral Parasympatholytic Activity

Isolated Guinea-pig Ileum.—Steady sub-maximal contractions to ACh were obtained at two-minute intervals. The dose of antagonist was given one minute before the next dose of ACh. The antagonistic effects of three separate doses of atropine, followed by three of methantheline, were measured by the percentage reduction in height of the subsequent contraction due to ACh.

Five such comparative assays of methantheline and atropine and five of propantheline and atropine were performed on pieces of gut from 10 guinea-pigs.

The percentage reduction in height of contraction was plotted against the logarithm of the dose of antagonist; the potency of methantheline or propantheline, compared with atropine, was obtained as the mean of the ratios of doses required to produce 50% inhibition of the height of contraction.

Blood Pressure in the Cat.—In each experiment a suitable dose of ACh was discovered (0.1 to 1.0 μg.) which would give an adequate fall in blood pressure. Comparisons of activity were made by relating the dose of each antagonist needed to inhibit to an equal degree the response to ACh.

Mydriatic Activity in the Mouse.—The method of Ing, Dawes and Wajda (1945) was used.

Intraperitoneal injections of atropine (2, 4, and 8 μg. in 0.2 ml. 0.9% NaCl) were made into three groups of five mice and the pupil diameter measured 15 min. later by viewing the eye under a strong light with the aid of a binocular microscope provided with a scale in the eyepiece. This procedure was repeated twice on the same day in six further groups of five mice, so that 45 animals in all were used. Three doses of methantheline (10, 20, and 40 μg., or 5, 10, and 20 μg.) were investigated in another 45 mice on the same day; four such complete experiments were performed. Similarly three complete experiments with

4, 8, and 16 μg. doses of propantheline, and 2, 4, and 8 μg. doses of atropine, were performed.

The results from the four experiments on methantheline and the three on propantheline were pooled separately and subjected to a suitable form of statistical analysis for calculating the regression line of the dose of mydriatic on mean pupil diameter (Emmens, 1948). From the two regression lines for methantheline and atropine, and the two for propantheline and atropine, the comparisons of potency were made.

Information about the duration of mydriatic activity was obtained by plotting curves relating mean pupil size to the time after intraperitoneal injection of the equiactive doses of 40 μg., 16 μg., and 8 μg. of methantheline, propantheline, and atropine respectively. Groups of 15 mice received atropine and methantheline in one experiment, and atropine and propantheline in another.

Salivary Secretion in the Cat.—The method used was that of Bülbring and Dawes (1945). A steady flow of saliva was produced by an intravenous infusion at a uniform rate (0.5 ml./min.) of 0.004% (w/v) of carbachol in Ringer–Locke solution. Adrenaline hydrochloride (0.002%) was included to offset to some extent the fall in blood pressure produced by the carbachol. The rate of secretion was recorded by a drop timer.

The relative activity of methantheline or propantheline and atropine was found by recording two effects of a dose of one substance between two effects of a dose of the other. This procedure was repeated until an exact match was obtained, or until two doses of one drug were found more and less effective respectively than a fixed dose of the other. The activity was judged by the maximum effect of each injection. Five cats were used for each assay. In each of the ten cats a comparison was made at several dose levels. In eight of the cats the activities of methantheline and atropine or propantheline and atropine in inhibiting the carbachol-induced fall in blood pressure were recorded.

Comparisons of Ganglion Blocking Activity

Nictitating Membrane of the Cat.—The autonomic ganglion blocking property of methantheline bromide was compared with that of hexamethonium in four cats anaesthetized with pentobarbitone sodium, and that of propantheline with hexamethonium in a further four cats, by relating the ability of the drugs to reduce the recorded height of contraction of the nictitating membrane in response to pre-ganglionic nerve stimulation. Stimulation was by square wave shocks, of 800 μsec. duration and 45 c/sec., automatically applied, every 2 min. for 3 to 10 sec., to the central end of the cut vago-sympathetic trunk. Steady contractions were obtained and the drug was given by vein at a fixed time before the subsequent contraction. The reduction in height of the contraction following the drug was expressed as a percentage of the previous contraction height and was plotted against the logarithm of the dose/kg. for each drug. Because the lines were not parallel, the relative potency was obtained by comparing the two log. dose-response lines for each pair of drugs at various response levels.

Blood Pressure in the Cat.—The relative ability of methantheline and hexamethonium to lower the blood pressure was observed in four cats, and that of propantheline and hexamethonium in three. All the cats were anaesthetized with pentobarbitone sodium.

Acetylcholine Hypertension in the Atropinized Cat.—In four cats anaesthetized with pentobarbitone sodium a comparison was made of the relative powers of hexamethonium, methantheline, and propantheline to inhibit the rise in blood pressure caused by the ganglion stimulation produced by large doses of ACh (0.1 to 0.2 mg./kg.) after large doses of atropine. Atropine (1 mg./kg.) was given initially by vein. Several doses of methantheline or propantheline and of hexamethonium were tried in order to find one of each which would almost prevent the rise of blood pressure following ACh.

Comparisons of Curare-like Activity

Cat Sciatic Nerve-Gastrocnemius Preparation.—The cats were anaesthetized with pentobarbitone sodium. Drug injections were given into the proximal side of the ligatured ipsilateral external iliac artery. The sciatic nerve was stimulated by single supra-maximal thyratron shocks at 10 sec. intervals. In two cats the relative potency of propantheline and methantheline was compared in producing inhibition of the contraction of the gastrocnemius muscle.

Comparisons of Local Anaesthetic Activity

An attempt was made to compare any local anaesthetic activity of methantheline with that of cocaine hydrochloride by the guinea-pig method of Chance and Lobstein (1944). The local anaesthetic was applied to both eyes and the corneal reflex tested 45 sec. later and at every minute for 5 min. The number of times the reflex was present out of a possible maximum of 10 for both eyes was recorded for several concentrations of methantheline (5, 2.5 and 1%) and cocaine hydrochloride (0.2%). The substances were dissolved in 0.9% NaCl.

Information regarding the duration, intensity, and any evidence of irritant properties of methantheline and propantheline as local anaesthetics was obtained by limited experimentation in two human subjects. Injections of 0.2 ml. of solutions of methantheline, propantheline or procaine hydrochloride were made into the skin of the flexor surface of the forearm. The solutions were made up in 0.9% NaCl; control injections were of saline alone.

RESULTS

Peripheral Parasympatholytic Activity

Guinea-pig Ileum.—The results of assays on guinea-pig ileum show propantheline to have a mean potency (relative to atropine=1) of 1.95±

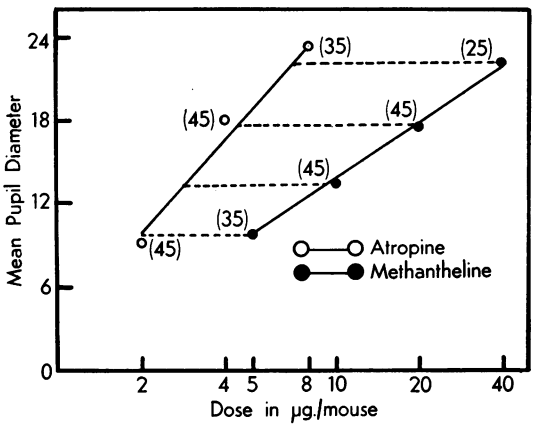

FIG. 1.—The log dose-response lines for methantheline bromide and atropine sulphate in producing mydriasis in the mouse. Ordinate: mean pupil diameter in arbitrary units of the microscope eyepiece scale. Abscissa: µg. per mouse intraperitoneally. The number of animals used for each point is indicated on the graph. The points shown on the graph are the observed mean values. The dose-response lines are calculated. The horizontal interrupted lines indicate the response levels at which the potency comparisons were made.

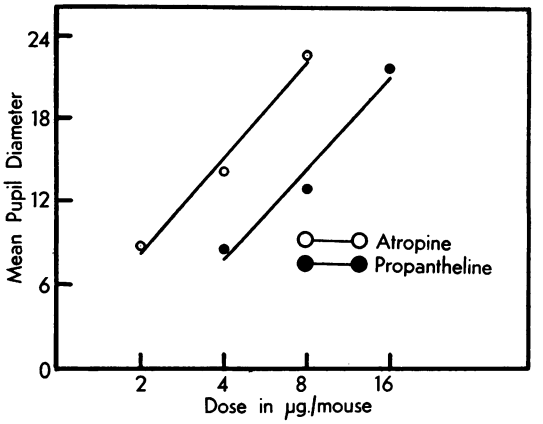

FIG. 2.—The log dose-response lines for propantheline bromide and atropine sulphate in producing mydriasis in the mouse (45 mice for each dose). Ordinate: mean pupil diameter in arbitrary units of the microscope eyepiece scale. Abscissa: µg. per mouse intraperitoneally. Points on graph are observed mean values. Dose-response lines are calculated.

0.08 S.E. (5 expts.) compared with 1.73±0.19 S.E. (5 expts.) for methantheline.

Intravenous Acetylcholine in the Cat.—In four cats the ratio of activity of methantheline to atropine lay between 0.7 and 1.0; in a further four cats the drugs were about equiactive.

Mydriasis in the Mouse.—The two log dose-response lines for methantheline and atropine are shown in Fig. 1 and those of propantheline and atropine in Fig. 2. The points shown on the graph are the observed mean values, whereas the dose-response lines are calculated.

As can be seen, the log dose-response lines for methantheline and atropine are far from parallel; it is therefore impossible to make an accurate estimate of the relative potency of the two drugs. An average has been taken of the relative potencies calculated at four response levels, and these levels are represented by the horizontal dotted lines between the regression lines. The regression lines for propantheline and atropine, however, are parallel. The mean relative potency of methantheline and atropine (=1) is 0.28 and the relative potency of propantheline and atropine (=1) is 0.47.

The durations of action of methantheline and atropine are compared in Fig. 3 and of methantheline and propantheline in Fig. 4. Doses of drugs used were those known to be equiactive at 15 min. The time for half recovery to the pre-injection diameter for methantheline is much less than for atropine (45 min. and 120 min. for 40 µg. and 8 µg. respectively of methantheline and atropine). Propantheline, however, approached atropine in time for half recovery (67.5 min. and 77.5 min. for 16 µg. and 8 µg. respectively of propantheline and atropine).

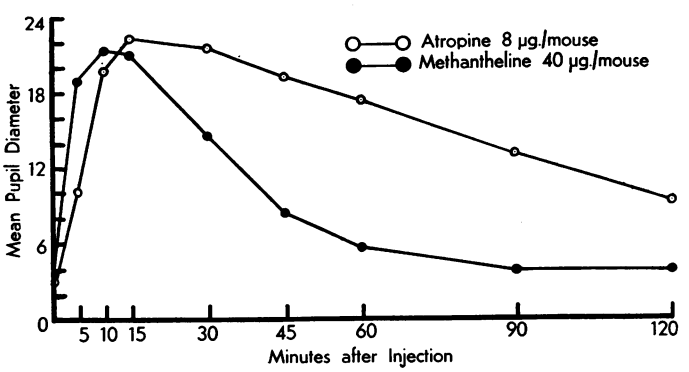

FIG. 3.—Duration of mydriasis in the mouse produced by 40 µg. of methantheline bromide is much less than that following 8 µg. of atropine sulphate intraperitoneally. These doses were equiactive at 15 min. Ordinate: mean pupil diameter from 15 mice in arbitrary units of the microscope eyepiece scale. Abscissa: time after injection in min.

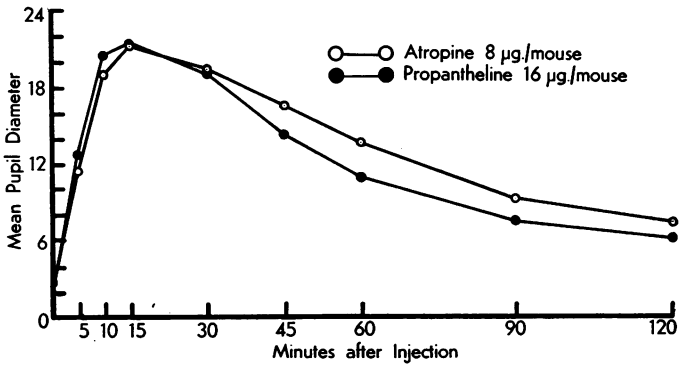

FIG. 4.—Duration of mydriasis in the mouse produced by 16 µg. of propantheline bromide is similar to that following 8 µg. of atropine sulphate intraperitoneally. These doses were equiactive at 15 min. Ordinate: mean pupil diameter from 15 mice in arbitrary units of the microscope eyepiece scale. Abscissa: time after injection in min.

Salivary Secretion.—The results of several comparative assays of methantheline and atropine from each of five cats and those of several assays of propantheline and atropine in each of a further five cats were averaged. The mean relative potency of methantheline (relative to atropine=1) calculated from all the results is 2.23±0.37 S.E. (5 expts.) and that for propantheline 2.95±0.41 S.E. (5 expts.).

In inhibiting the hypotension produced by the infusion of carbachol, methantheline and atropine were approximately equipotent, whereas propantheline had a mean potency of 2.7 (relative to atropine=1).

Ganglion Blocking Activity

Nictitating Membrane of the Cat.—The extent of the fall in blood pressure produced by hexamethonium was such that a dose which would cause over 50% inhibition of contraction could not be given in some cases without fear of killing the animal. The log dose/kg.-response lines for the action of methantheline and propantheline and hexamethonium on the nictitating membrane are not parallel; estimates of relative potency were therefore made at several response levels and the mean calculated for each experiment. Methantheline has a mean potency (relative to hexamethonium=1) in these experiments of 0.40±0.06 S.E. (4 expts.) and propantheline (relative to hexamethonium=1) of 0.63±0.08 S.E. (4 expts.) (Fig. 5).

In two cats it was found that 5 mg./kg. of methantheline or of propantheline had no effect on the contraction of the nictitating membrane produced by intravenous adrenaline.

Reduction in Blood Pressure.—In three cats, 0.5 mg. hexamethonium produced a fall in blood pressure equal to that due to 5 mg. of methantheline and in three other cats 5 mg. of propantheline produced a fall in blood pressure equivalent to from 2.5 to 4 mg. of hexamethonium.

ACh Hypertension in the Atropinized Cat.—Hexamethonium and methantheline were equiactive: propantheline was twice as active as hexamethonium.

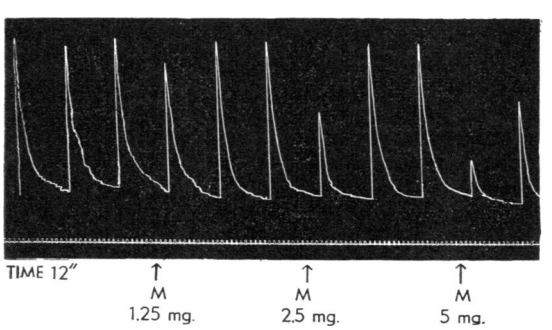

FIG. 5.—A typical record showing the effect of methantheline bromide (M) in inhibiting the contraction of the nictitating membrane of the cat in response to pre-ganglionic sympathetic stimulation. Cat anaesthetized with pentobarbitone sodium. Time intervals, 12 sec.

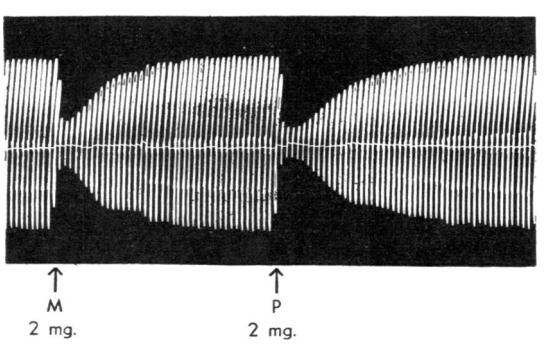

FIG. 6.—Record of the contractions of gastrocnemius muscle of the cat in response to stimulation of the sciatic nerve by single supramaximal shocks from a thyratron stimulator at a rate of 5/min. The inhibitory effects of 2 mg. of propantheline bromide (P) and 2 mg. of methantheline bromide (M) given intra-arterially are seen to be equal. Cat anaesthetized with pentobarbitone sodium.

Curare-like Activity

Propantheline and methantheline showed curare-like activity only in very large doses. They were approximately equiactive. The effects of 2 mg. of propantheline and of methantheline are shown in Fig. 6.

Local Anaesthesia

Using three concentrations of methantheline (5%, 2.5%, 1% in saline) instilled into the guinea-pig eye, no local anaesthetic activity could be demonstrated in any of nine guinea-pigs. Compared with 0.2% cocaine hydrochloride, it produced marked signs of irritation, especially with the 5% and 2.5% concentrations. Similar results were obtained with propantheline in six guinea-pigs, except that there was some evidence of anaesthesia with a 5% concentration. As with methantheline, marked signs of irritation were produced by the 2.5% and 5% solutions.

Experiments in man confirmed the irritant action shown in the guinea-pigs. Intracutaneous injections were followed by the development of a spreading weal and surrounding flare of vasodilatation. Methantheline 1% in two subjects produced anaesthesia of slow onset but prolonged duration in the skin overlying the weal produced by the injection. Several minutes were needed for the full development of anaesthesia, but it lasted for 6 to 7 hr. This is in contrast to the immediate anaesthesia produced by 0.25% procaine, and which continues for only 25 to 30 min. A higher concentration of methantheline, 2%, produced anaesthesia of slightly more rapid onset and of much longer duration. There was still some degree of anaesthesia 24 hr. later. The low concentration of 0.5% methantheline had a similar effect to the 1% concentration. After 1 and 2% methantheline, scarring and hypersensitivity of the skin were present for at least two weeks. Exactly similar results were obtained with propantheline, using 5%, 2.5%, and 1% solutions. It is interesting to note that 0.1 c.c. of each of these solutions intradermally (equal to approximately 8 mg.) in each subject produced complete arrest of salivary secretion and an increased pulse rate. The effect on salivary secretion lasted for approximately five hours. Inhibition of gastro-intestinal activity was suggested by a feeling of fullness in the stomach, and loss of appetite.

Discussion

The present results with methantheline differ considerably from those of other workers—notably Lehman and Knoeffel (1944). As far as can be ascertained no systematic pharmacological examination of the properties of propantheline has yet been reported, apart from that of Hambourger (1952).

Lehman and Knoeffel (1944) found methantheline to be 100 times weaker than atropine in inhibiting salivary secretion. Although their assay was done on cats, their method was totally different from that used here. They used pilocarpine as the sialogogue, and both antagonist and sialogogue were injected subcutaneously, in contrast to the intravenous route used by Bülbring *et al.*, and by which we have found methantheline to be twice as effective as atropine.

Lehman and Knoeffel's results for the relative potencies of methantheline and atropine in preventing the vasodepressor action of ACh also differ from ours. They found methantheline less potent than atropine in this respect, whereas we found the two drugs to be almost equipotent. This difference is difficult to explain, but may depend on the fact that dogs were used in their experiments and cats in ours.

From the present work one can say that propantheline is slightly more active than methantheline, but this increase in activity is roughly equally distributed between its peripheral and autonomic ganglion blocking activity. Hambourger (1952) found propantheline approximately five times more powerful than methantheline against ACh-induced spasm of guinea-pig gut, and approximately twice as active as methantheline in producing paralysis of the superior cervical ganglion. We found no very significant difference between the antimuscarinic activity of propantheline and methantheline, though our results in the nictitating membrane experiments do correlate well with Hambourger's. No satisfactory explanation can be offered for the former anomaly.

The local anaesthetic property of methantheline on the rabbit eye, shown by Lehman and Knoeffel (1944), could not be demonstrated on the guinea-pig eye, though human experiments indicate that both propantheline and methantheline have some local anaesthetic activity when given intracutaneously. How specific this is is not very certain. It may be that methantheline and propantheline damage the nerve endings in the skin, for the injected area remains scarred for several weeks. The skin does not completely regain its normal sensitivity for some time, anaesthesia being followed, for several days after the injection, by a period of hypersensitivity of the protopathic type.

The curare-like activity of both compounds is equal, but very weak (Fig. 6), 2 mg. of each drug

intra-arterially being needed to produce a moderate effect.

One main difference between the two drugs appears to be their duration of action. In the mouse mydriasis experiments the duration of action of propantheline closely approximated that of atropine, whereas the action of methantheline was much shorter.

Although propantheline and, to a lesser extent, methantheline, approaches hexamethonium in ganglion blocking activity, this activity is only present in a dose range, or concentration range, greatly in excess of that necessary for parasympathetic blockade. It seems, therefore, that ganglion blockade is not likely to be responsible for any therapeutic effects when these substances are used clinically—unless a dose range is used which would produce pronounced signs of peripheral parasympathetic blockade.

Clinical reports have stressed the advantages of methantheline and propantheline as atropine substitutes—in respect of relative absence of side-effects—in doses adequate for treating certain gastro-intestinal disorders. Propantheline is claimed to be better than methantheline from this point of view. It may be argued that the relative infrequency of serious side-effects with these drugs is further evidence that their ganglionic blocking activity contributes little if anything to their effectiveness in clinical use.

Summary

1. Propantheline and methantheline, two atropine-like agents claimed to have in addition considerable ganglion blocking activity, have been compared with atropine and hexamethonium in their power to produce peripheral parasympathetic blockade and autonomic ganglion blockade respectively.

2. Both are more potent antimuscarinic agents than atropine, propantheline having 1½ to 2 times the activity of methantheline.

3. Their ganglion blocking potency, which is exhibited in a much higher dosage range, varies from 1/10 to twice that of hexamethonium, depending on the test preparation used. Propantheline is, again, the more active.

4. Both drugs have some weak curare-like action. They possess little, if any, local anaesthetic activity.

5. The significance of these findings, in relation to previous work and to the clinical effects of the drugs, is discussed.

The authors wish to thank Dr. W. E. Hambourger and Messrs. G. D. Searle Ltd. for kindly supplying the methantheline bromide and propantheline bromide used in these experiments. One of the authors (E.A.J.) carried out this work during the tenure of a Medical Research Council grant for training in research methods.

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