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THE EFFECT OF RESERPINE ON THE ACETYLCHOLINE CONCENTRATION IN HYPOTHALAMUS, ILEUM, AND HEART OF THE DOG AFTER BILATERAL VAGOTOMY AND GANGLIONIC BLOCKADE

BY

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Malhotra & Pundlik (1959) observed that after intravenous administration of reserpine there was a significant increase of acetylcholine content in various areas of dog brain but not in the hippocampus, which showed a significant decrease. Malhotra & Das (1962) showed that reserpine given intravenously not only increased the acetylcholine content of the hypothalamus (central area) but also that of the sino-atrial node, the right auricle and the ileum (peripheral tissues). At the same time there was sedation, bradycardia and purgation. It was suggested that these effects might be due to an increase in the acetylcholine content of the tissues studied. Malhotra & Prasad (1963) found that 0.75 mg of reserpine administered into the cerebral ventricle caused a significant increase in the acetylcholine content of the sino-atrial node, the ileum and the hypothalamus of the dog. There was also bradycardia, purgation, miosis and sedation. The same dose of reserpine given intravenously did not increase the acetylcholine content of the ileum or the hypothalamus though there was some increase in the acetylcholine content of the sino-atrial node. There were, however, no significant general effects. These studies indicated that the peripheral parasympathetic effects as well as the increase in the acetylcholine content of the peripheral tissues, after reserpine, were probably due to a central action of reserpine.

In order further to elucidate the central mediation of the peripheral effects, we studied the general effects of intravenous reserpine as well as the acetylcholine content of certain tissues after bilateral vagotomy and during ganglion blockade.

METHODS

The experiments were performed on thirty-five healthy mongrel dogs of either sex weighing between 7.5 and 14 kg. The dogs were divided into five groups of seven each. The acetylcholine content of the following areas was estimated 30 min after intravenous administration of 0.5 mg/kg of reserpine: the hypothalamus, the ileum, the right ventricle, the right atrial appendix and the sino-atrial node. General effects, such as sedation, salivation, emesis, miosis and bradycardia, were noted.

Group 1 served as a control and in this group an equivalent volume of saline was given intravenously. In group 2, pentolinium (2 mg/kg) was administered intravenously and after 50 min the acetylcholine was estimated. In group 3, pentolinium (2 mg/kg) was given intravenously followed 20 min later by reserpine

(0.5 mg/kg) intravenously. The acetylcholine content of tissues was estimated 30 min after reserpine (50 min after pentolinium). In group 4 bilateral vagotomy was performed aseptically by removing 4 to 5 cm of the vagus in the neck and the tissues were removed after 48 hr for estimation of acetylcholine content. In group 5, bilateral vagotomy was performed and after 47.5 hr reserpine (0.5 mg/kg) was given intravenously and the acetylcholine content of tissues was estimated 30 min later.

Acetylcholine was extracted from the tissues with acidified frog-Ringer solution containing physostigmine at 95 to 100° C and assayed on frog rectus abdominis muscle by the method of Nachmansohn, as modified by Anand (1952). With this method, acetylcholine standards are prepared in previously treated tissue extracts, in order to compensate for the presence of sensitizing substances. The control experiments were interspersed between the experiments with reserpine. The experiments were done during ether anaesthesia and the dogs were bled before taking out the tissues.

RESULTS

The acetylcholine concentration of the five different groups of dogs are given in Table 1 and Fig. 1. The general effects before and after reserpine in these groups are given in Table 2

TABLE 1

ACETYLCHOLINE CONCENTRATIONS OF DIFFERENT TISSUES OF THE DOG

Acetylcholine concentrations are expressed in μg/g of fresh tissue. There were seven dogs in each group, and values are means and standard deviations. Drugs were injected intravenously. Probabilities of no significance were calculated for Groups 2 and 4 compared with Group 1 (controls) and for Group 3 compared with Group 2 and Group 5 compared with Group 4 (calculated by the *t*-test); for all comparisons $P>0.1$, except the pairs marked *($P<0.01$) and †($P<0.05$)

Group	Treatment	Acetylcholine concentration (μg/g) in Hypothalamus	Ileum	Right ventricle	Appendix of right atrium	Sino-atrial node
1	Control	4·8±0·5	3·4±0·7	0·7±0·2	3·0±0·7	6·7±1·1
2	Pentolinium (2 mg/kg)	4·3±1·8	3·7±1·3	1·1±0·3	4·7±1·9	8·3±2·8
3	Pentolinium and reserpine (0·5 mg/kg)	*8·4±2·2	4·9±1·3	1·2±0·3	5·7±1·6	9·5±2·0
4	Bilateral vagotomy	†5·3±1·7	4·6±1·4	0·8±0·2	4·5±1·7	7·2±2·0
5	Bilateral vagotomy and reserpine (0·5 mg/kg)	†6·7±0·3	5·1±1·8	0·9±0·1	6·0±1·8	8·6±2·3

TABLE 2

SOME GENERAL EFFECTS OF INTRAVENOUS ADMINISTRATION OF RESERPINE (0·5 MG/KG) IN PENTOLINIUM- AND RESERPINE-TREATED AND VAGOTOMIZED AND RESERPINE-TREATED DOGS

Heart rates are means for seven dogs. Numbers in parentheses give numbers of dogs showing the effect in a group of seven

Group	Treatment	Heart rate (beats/min)	Purgation	Salivation	Pupil size	Behaviour
2	Pentolinium	127	No	No	Slight dilatation (5)	Normal
3	Pentolinium and reserpine	134	No	No	Normal	Sedated (6)
4	Bilateral vagotomy	156	No	No	Normal	Normal
5	Bilateral vagotomy and reserpine	150	No	Marked	Miosis (7)	Sedated (6)

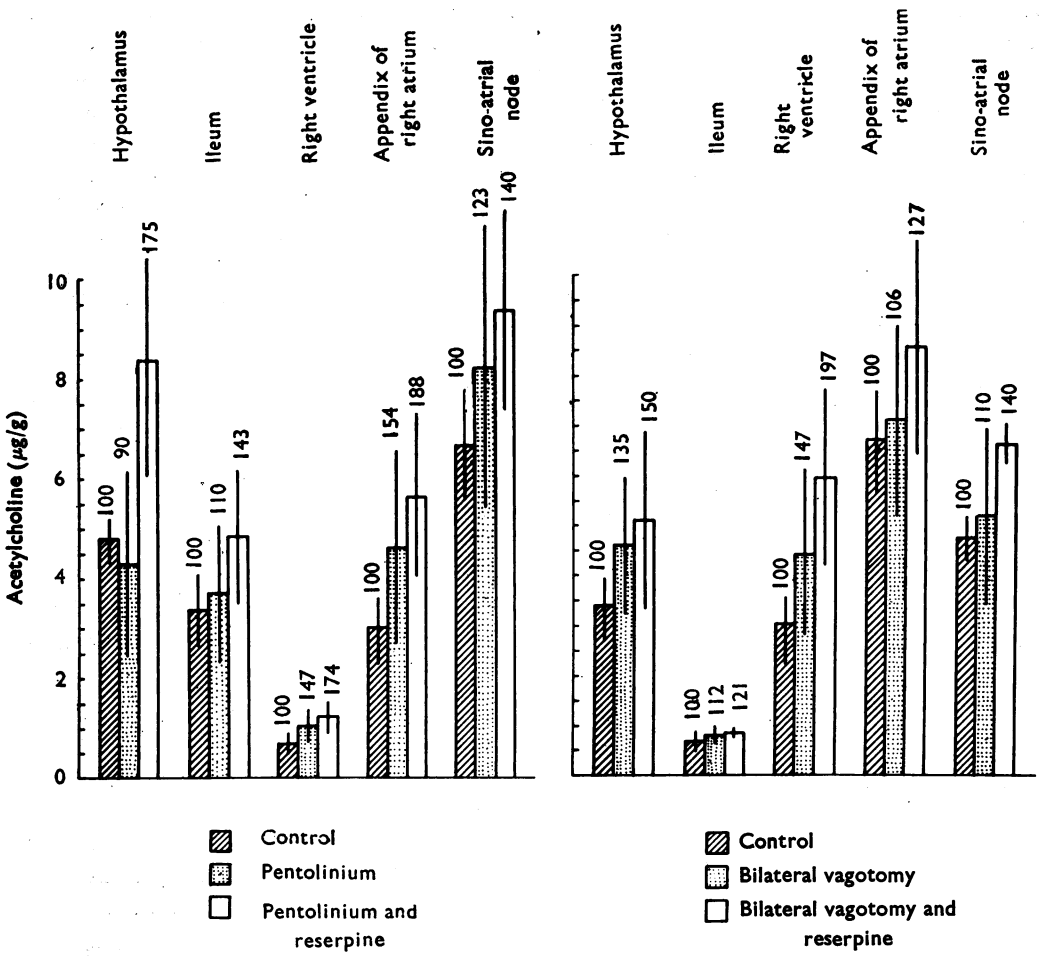

Fig. 1. Histogram showing the distribution of acetylcholine in different tissues of dogs. On the left, controls (hatched columns) after pentolinium (2 mg/kg) (dotted columns), and after pentolinium and reserpine (0.5 mg/kg) (empty columns). On the right, controls (hatched) after bilateral vagotomy (dotted) and after bilateral vagotomy and reserpine (empty). All drugs were injected intravenously. Acetylcholine concentrations are in μg/g of fresh tissue. Numbers over the columns show the percentages of acetylcholine in relation to the control value (100%) for the tissue. Vertical lines give standard deviations.

DISCUSSION

The results indicate that in normal dogs the sino-atrial node contains the highest amount of acetylcholine while next in descending order are the hypothalamus, the ileum, the appendix of the right atrium and the right ventricle. After pentolinium there is no significant change in the acetylcholine level of these tissues compared with the normal group. There is, however, a statistically significant ($P<0.01$) increase in the acetylcholine content of the hypothalamus in the group treated with pentolinium and reserpine compared with the

pentolinium-treated group. The change in the peripheral tissues is not significant. Similarly, after bilateral vagotomy there is no significant change in the acetylcholine content of the tissues compared with the normal group, while in the vagotomized and reserpine-treated group there is a significant ($P<0.05$) increase in the acetylcholine content of the hypothalamus alone compared with the bilateral vagotomized group. As far as general effects are concerned, it is interesting to find that in the vagotomized and reserpine-treated group there was sedation, miosis and salivation but no bradycardia, emesis or purgation. On the other hand, in the pentolinium- and reserpine-treated group there was only sedation and no miosis, salivation, bradycardia or purgation. It is well known that reserpine given alone produces peripheral parasympathetic symptoms, such as bradycardia, increased intestinal motility, salivation and miosis. Bogdanski, Sulser & Brodie (1961) have also shown that previous administration of ganglion-blocking agents abolish the miosis and salivation seen after reserpine. Ludány, Gáti & Hideg (1958) have, however, reported that the increased motility of intestinal villi after giving reserpine is not affected by vagotomy but is blocked by atropine, which suggests a peripheral action of reserpine.

Our findings suggest that the increase in the acetylcholine concentration of the peripheral tissues, as well as the peripheral parasympathetic effects like miosis, salivation, bradycardia and purgation, observed after intravenous reserpine, are central in origin.

Sedation, which is undoubtedly due to the central action of reserpine, was present both in the vagotomized and reserpine-treated group as well as in the pentolinium- and reserpine-treated group.

SUMMARY

1. The effect of intravenous administration of reserpine on the acetylcholine concentration of the hypothalamus, ileum, right ventricle, appendix of the right atrium and the sino-atrial node was studied in thirty-five dogs divided into five equal groups. General effects (sedation, bradycardia, miosis, salivation, emesis and purgation) were noted.

2. Group 1 served as a control. Group 2, which received pentolinium, showed no significant change from the controls in the acetylcholine level of the tissues.

3. Group 3, which received pentolinium and reserpine, showed a statistically significant increase in the acetylcholine level of the hypothalamus alone; there was also sedation but no other general effects.

4. Group 4, which had bilateral vagotomy, did not show any significant change from the controls in the acetylcholine levels, or any general effects.

5. Group 5, which had bilateral vagotomy and reserpine, showed a significant increase in the acetylcholine content of the hypothalamus alone; there was sedation, miosis and salivation but no other general effects.

6. These findings suggest that the peripheral parasympathetic effects of reserpine are due to a central nervous action.

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