ON THE MECHANISM OF THE IMPAIRMENT OF THE PARASYMPATHETIC INNERVATION OF THE HEART DURING EXPERIMENTAL INTOXICATION WITH BOTULINUS TYPE A TOXIN

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The literature on the mechanism of the impairment of the neural regulation of the cardiovascular system during botulism does not include a description of the nature of the impairment of the effect of the parasympathetic nervous system on the heart, and very little data is available regarding the mechanism by which the functions of the vagus nerve are impaired. Only Guyton and MacDonald [8], during their study of the depressing effect of the vagus nerve on the blood pressure, expressed the hypothesis that botulinus toxin may impair the function of the nerve endings of this nerve. Burgen, Dickens and Zatman [6] and Davis, Morgan, and Wright [7] took the same point of view in their explanation of the mechanism of the impairment of the somatic innervation by botulinus toxin.

It is believed that botulinus toxin attacks only the endings of the somatic nerves in the myoneural areas and that it does not act on the functioning of the nerve cells and nerve processes. On the other hand, the principle symptoms in the clinical picture of botulism, as well as data obtained from histological examination of the organs of those who died from botulism, indicate that the central nervous system is attacked during this toxinfection [1, 2, 5]. K. I. Matveev [4] states that botulinus toxin can attack not only the nervous system, but other tissues as well.

We made our goal the clarification of the impairment of the parasympathetic action on the heart during botulism and the determination of the sections of the parasympathetic innervation of the heart which are the points of attack by the botulinus toxin.

EXPERIMENTAL METHOD

The experiments were carried out on rabbits which were given doses of from 12,000 to 44,000 mouse MLD (from 0.6 to 2.2 mg per kg of body weight) of botulinus type A toxin (1 mouse MLD equivalent to 0.00005 mg dry toxin) dissolved in physiological salt solution, pH 6.8 *, intravenously, intramuscularly, and intra-arterially. With these doses, acute intoxication developed in 2-3 hours. At the peak of the intoxication, when the animal began to suffer sever dyspnea, artifical respiration was initiated. Then a vital test was set up using urethane or hexenal anesthesia.

In order to register the contractions of the heart, the animal's chest was opened and the heart was suspended by a thread from the writing arm of a direct myograph. The vagus nerve was dissected out at the neck and was stimulated by electrodes charged from an induction apparatus (electromotive force of the supply batteries was 2.5 v). The discharge voltage was slightly higher than the threshold.

70 experiments were carried out in all.

* According to E. D. Kushnir [3], botulinus toxin is most active at pH 6.8.

EXPERIMENTAL RESULTS

First, we conducted control experiments with healthy animals. The effects on the heart of stimulating the intact vagus nerves, as well as the proximal and distal ends of the cut nerves, were studied. In all cases of stimulation of the vagus nerves, a weakening and slowing of the cardiac action was registered; noticeable effects were observed when the distance between the induction coils was 160 to 250 mm.

In the next series of experiments, the effect of stimulation of the vagus nerve on the heart of rabbits which had been injected intravenously with the toxin dosage indicated above several hours before the test was studied. Marked impairment of the vagal action was found in the poisoned animals. Thus, the first occurrence as intoxication developed was the interruption of the action of the vagus nerve, which weakens the action of the heart. The slowing action, however, was more persistent and only disappeared in the later stages of intoxication (Figure 1). At the same time, we observed a considerable decrease in the excitability of the vagus nerves: in order to obtain even the slightest effect, the vagus nerve had to be stimulated with a strong current (the distance between the induction coils was 40 to 140 mm).

Thus, as botulism develops in rabbits, first the weakening and then the frequency-reducing actions of the vagus are inhibited.

Later we tried to discover whether the observed inhibition of the vagus nerve is the result of the toxin's action on the peripheral portion of the parasympathetic system or whether it is connected with the action of the toxin on the parasympathetic centers also. In order to determine this, the vagus nerve was cut on one side of the animal's neck, dissevering the central and peripheral sections of the parasympathetic innervation of the heart. The other nerve was left intact. Immediately after the nerve was severed, botulinus toxin was administered to the animal (in the same way as described above). Several hours later, after severe intoxication had developed, the vital test was carried out.

As shown in Figure 2, the destructive effect of the botulinus toxin was noticeable only when the intact vagus nerve was stimulated. Stimulation of the peripheral end of the severed nerve, in spite of the considerable length of time elapsing after the poisoning, resulted in the usual weakening and slowing action on the heart in all of the experiments. Meanwhile, the irritability of the vagus was not lowered perceptibly.

Consequently, experiments in which the vagus was cut prior to the administration of the toxin point to the conclusion that the connection between the peripheral and central sections of the parasympathetic innervation of the heart must be intact if the toxin is to have a destructive effect on the vagus action. When this connection is broken, i.e., when the vagus is severed, the botulinus toxin apparently is unable to disturb the functioning of the peripheral end of the cut nerve. These data contradict the conclusion of Guyton and MacDonald, and others about the exclusively peripheral action of botulinus toxin on the myoneural connections. On the contrary, this toxin apparently has only a central action, disturbing the central, not the peripheral, sections of the parasympathetic innervation of the heart. This point of view is supported also by experiments on the administration of botulinus toxin in a dose of 2000 mouse MLD (0.1 ml at a concentration of 1:1000) directly into the fibers of the vagus. Inspite of the length of time (up to 5 hours) after the administration, in none of these experiments did we detect any effect of the toxin on the vagus. Stimulation of the nerve both at the point where the toxin was administered and above and below it had the usual inhibitory effect on the contraction of the heart.

Having discovered that the destructive action of botulinus toxin is only evidenced when the central and peripheral sections of the parasympathetic innervation of the heart remain joined, we turned to the study of the mechanism whereby the botulinus toxin disturbs the central section of this innervation. We were faced with the problem whether the parasympathetic centers were disturbed by the direct action of the botulinus toxin, or whether they were disturbed as a result of the pathological impulses from the cardiac receptors, stimulated by the poison. In order to solve this problem, we administered the same doses of botulinus toxin used in the preceding experiments into the myocardium in the area of the right or left ventricle. Under these conditions, the huge doses of toxin, irritating the cardiac receptors maximally, should have produced a faster and more severe disturbance of the effectors. However, under these conditions the disturbance of the parasympathetic did not develop sooner than it did in experiments when the toxin was administered intravenously. Apparently, the toxin, when introduced into the myocardium, quickly reaches the general circulatory system due to the rapid circulation in the heart and produces the disturbance of the vagus. At the same time, as was indicated, the harmful effect was observed only on intact vagus nerves. If the vagus was severed before the administration of the botulinus toxin into the myocardium, irritation of its peripheral end produced a noticeable weakening and slowing effect on the cardiac activity.

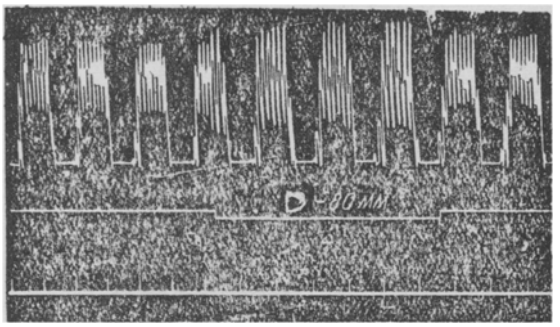
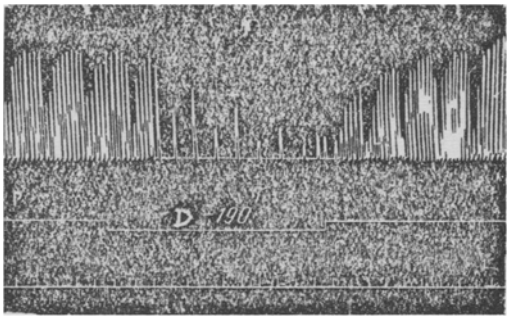


Figure 1. Effect of stimulation of the right (intact) vagus on the heart of a rabbit poisoned 3 hours earlier with botulinus toxin. Disappearance of the inotropic and preservation of the chronotropic action. D) distance between induction coils.

a



b

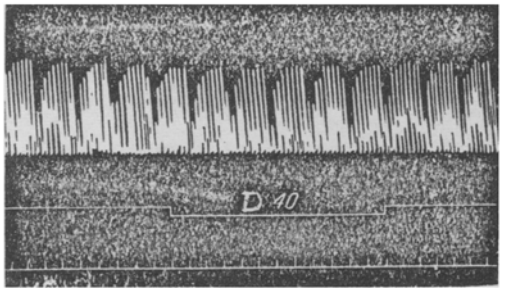


Figure 2. Effect of stimulation of the left (intact) vagus (a) and of the peripheral section (b) on the heart of a rabbit poisoned with botulinus toxin.

The results of the experiments with direct administration of massive doses of botulinus toxin into the myocardium led us to the conclusion that the destructive action of the toxin was directed not at the intramural nervous system of the heart, but at the central section of the parasympathetic system.

In order to confirm this hypothesis, additional experiments were carried out in which we attempted to discover what portion of the reflex arc (the peripheral or central) was first attacked by the botulinus toxin. We produced reflex inhibition of cardiac action by irritating the central end of the severed vagus. The experiments were conducted under the same conditions as before. The results indicated that after the botulinus toxin was introduced, the central portion of the reflex arc was disrupted first, and later the inhibitory function of the peripheral areas of the parasympathetic innervation of the heart was lost. 60-80 minutes after the administration of the indicated dose of botulinus toxin, the irritability of the central ganglia of the vagus began to decrease noticeably: when the central end of the severed vagus was stimulated, its only action on the heart was to slow its contraction, but no weakening effect was observed. 1 hour 45 minutes later, stimulation of the central end of the severed nerve had no action whatsoever on the heart, while stimulation of the nerve still produced a noticeable slowing of the contractions.

Thus, experiments on the reflex inhibition of cardiac action also show that the botulinus toxin attacks the functions of the central parts of the reflex arc first, and then the peripheral.

Our data testify to the fact that botulinus toxin disrupts the activity of the parasympathetic innervation of the heart by acting on its central sections.

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