DIFFERENTIATION BETWEEN NERVOUS AND HUMORAL INFLUENCES BY SIMULTANEOUS RECORDING OF THE MOTOR ACTIVITY OF INNERVATED AND DENERVATED LOOPS OF SMALL INTESTINE

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The most accurate information concerning humoral changes may be obtained by the use of biochemical methods of determination of biologically active substances in the blood. However, because of the complexity of these methods, the necessity of taking quantities of blood, and also, in some cases, the inadequate sensitivity of the methods, they cannot always be used. Moreover, it is not always possible to determine accurately the time of taking the blood, so that the humoral phase of the reaction may easily be lost sight of.

A widely used method of studying humoral influences is that of recording the reactions of denervated structures. However, this method also is unable to differentiate between the nervous and humoral components of the reaction in the same experiment. Such an analysis can be undertaken only by the simultaneous recording of the responses of innervated and denervated structures to the same stimulus.

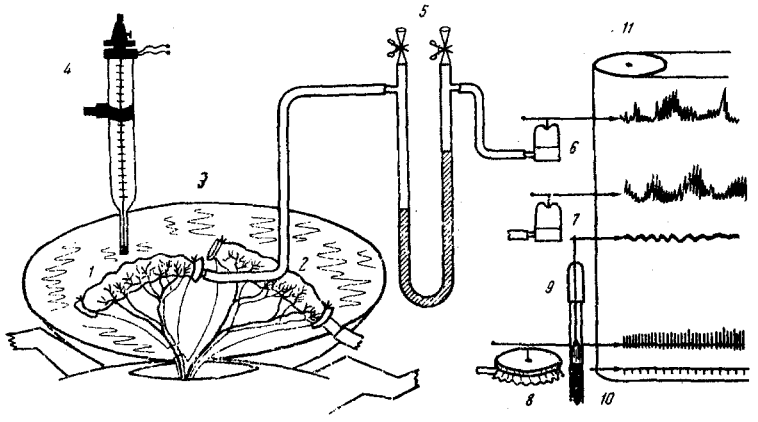

Fig. 1. Scheme of apparatus. 1) Denervated loop of small intestine; 2) innervated loop of small intestine; 3) heating chamber; 4) contact thermometer; 5) water manometer of the system for recording the motor activity of the small intestine (the second manometer is not shown); 6,7) recording tambours; 8) Marey's tambour for recording respiration; 9) mercury manometer for recording arterial pressure; 10) time marker; 11) drum of kymograph.

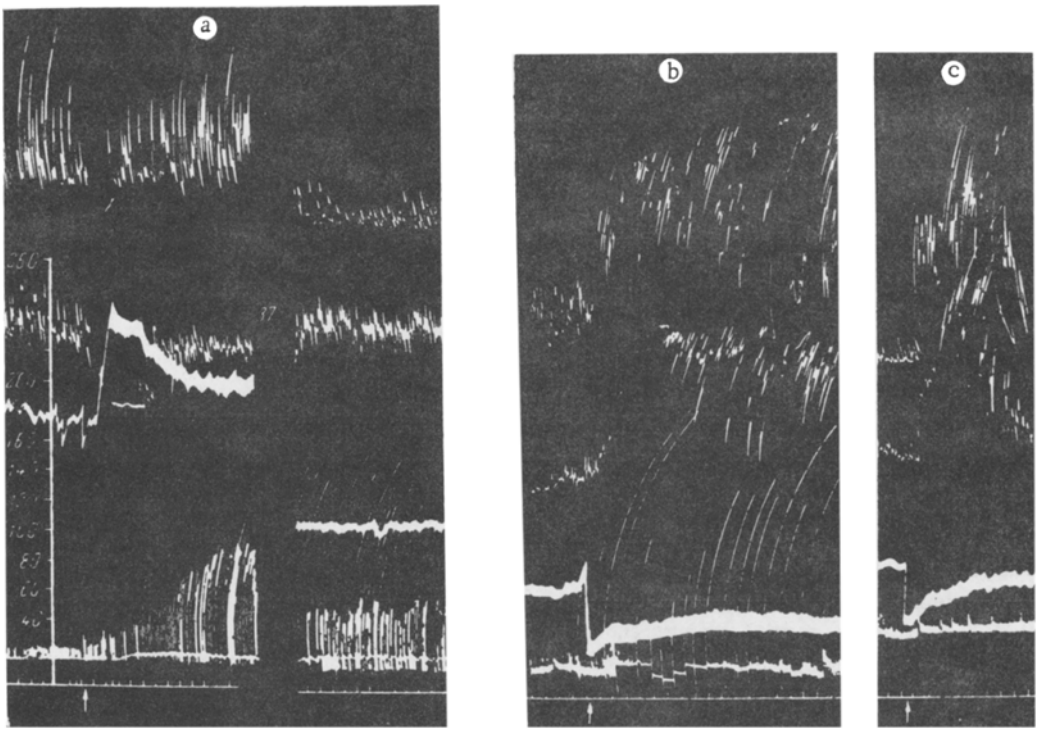

Fig. 2. Autonomic reactions during injection of carbachol. A) Effect of suboccipital injection of carbachol in a dose of 5 μg/kg; B) effect of intra-arterial injection of the same dose of carbachol; C) effect of intravenous injection of the same dose of carbachol. The arrow indicates the moment of injection. Significance of the curves (from top to bottom): motor activity of innervated loop of small intestine, motor activity of denervated loop of small intestine, tracing of arterial pressure, tracing of respiration, zero line of arterial pressure, time marker (30 sec).

In such a method it is important to note that the innervated and denervated organs must be physiologically equivalent, and that the denervation must be simple and absolutely reliable; the organs must be highly sensitive to nervous and humoral agents; the response reactions must develop quickly and must be capable of quantitative and qualitative evaluation. The method which we suggest for simultaneous recording of the reactions of innervated and denervated loops of small intestine satisfies all these demands.

The action of the humoral factor causes the same type of reaction in both loops of intestine, but in the denervated loop, which is more sensitive to humoral influences (Cannon), the reaction is more marked. In the case of a purely reflex mechanism, only the innervated loop reacts. Finally, in phased neuro-humoral reactions, each phase is characterized by definite variations in the innervated and denervated structures.

In an acute experiment laparotomy is performed through a midline incision, the small intestine extracted, and two neighboring loops identified in its middle third. After ligation of the vessels of the arcade, the loops are resected. All the tissues of the mesentrey around the blood vessels of one of the loops are carefully dissected and divided. The vessels are treated with 30% phenol solution, after which all visible nerves are divided, and the adventitia is tripped from the artery.

The proximal ends of the loops formed in this manner are ligated, and into the distal ends are introduced glass cannulas (diameter about 8 mm), through which the loops may be filled with physiological saline. The loops of intestine are then connected by cannulas to water manometers and the recording system (Fig. 1). To stabilize the experimental conditions the loops are placed in a dish containing physiological saline, maintained at a constant temperature (36°) by an ultrathermostat.

The suggested method may be used for various experimental purposes, such as the study of the fine mechanisms of the reactions of the intesting arising in response to stimulation of the interoceptors of various parts of the vascular

system (the vessels of the intestine, carotid sinus, etc.), of the mechanico- and chemoreceptors of the gastro-intestinal tract, and of the sensory endings of the cerebrospinal and autonomic nerves, and also the reactions evoked by stimulation of various brain structures (the hypothalamus, the limbic cortex, etc.).

We have used this method to study the mechanisms of the autonomic changes during injection of cholinergic drugs into the cerebrospinal fluid. It is known that the autonomic changes resulting from the central administration of cholinergic substances (acetylcholine, carbachol) are usually opposite to the effects observed when the same substances are injected into the general circulation [1-7]. However, the neurohumoral mechanism of the autonomic changes has not yet been elucidated.

In our experiments the arterial pressure and respiration were recorded at the same time as the reactions of the innervated and denervated loops of intestine. The experiments were conducted on cats anesthetized with ether and thiopental. Carbachol was injected suboccipitally, intra-arterially, and intravenously in doses of 5-10 μg/kg body weight.

A transient but clear inhibition of motor activity of both the innervated and the denervated loops of intestine was observed 5-15 sec after suboccipital injection. At the same time the arterial pressure was raised by a varied amount (by 10-80 mm), and respiration was momentarily arrested, after which the amplitude of the respiratory movements was increased sharply and their frequency slightly (Fig. 2, 4).

The fact that the reactions of both the innervated and the denervated loops of intestine was inhibitory demonstrated the presence of a humoral sympathetic change. The sympathetic character of the reaction was also confirmed by the changes in the arterial pressure and respiration. The subsequent development of the reaction revealed complete normalization of the motor activity of the denervated loop and a more or less marked inhibition of the motor activity of the innervated loop, which could be interpreted as reflex.

After intra-arterial and intravenous injection of the same dose of carbachol both loops showed the same type of reaction of an increase in tone and an increase in the amplitude of the contractions of the two loops. These reactions, like the changes observed in the arterial pressure and respiration (Fig. 2B, C), indicated a marked humoral parasympathetic influence.

The suggested method can also be used to differentiate between the direct action of injected drugs on the smooth muscle of the intestine and their action when mediated through the nervous system and the glands of endocrine secretion (in the last case the character of the reaction is assessed from its rate of onset).

The method of simultaneous recording of the activity of the innervated and denervated loops of small intestine may also be used in chronic experiments on dogs following the preliminary exteriorization of two loops by the Thiry-Vella technique and the subsequent denervation of one of these loops.

SUMMARY

A method is suggested which makes it possible to record simultaneously (in an acute experiment) the reaction of the innervated and denervated small intestine loops for differentiation of the nervous and humoral factors acting upon an intact organism.

The results obtained by using this method for ascertaining neuro-humoral mechanisms of the autonomic changes following central administration of cholinergic agents are decanted.

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