INTESTINAL REACTION TO DRUGS IN DIFFERENT FISHES.—By N. B. DREYER, Department of Pharmacology, McGill University, Montreal, (and at the Marine Biological Laboratory, Woods Hole, Mass., U.S.A.).

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ABSTRACT.

In elasmorbranchs the vagus seems to exert very little effect on the gut movements, since pilocarpine in small or big doses produces no change.

Urea is not essential in perfusing fluids for elasmobranch organs.

Bony fish gut receives a motor supply from the vagus and an inhibitory supply from the sympathetic.

The response of mammalian gut to different drugs has been repeatedly studied and the reactions of the two types of nerve endings—sympathetic and parasympathetic—can be easily brought out in isolated strips of intestines or of the gut *in situ*. Very little evidence has been brought forward as to the innervation of the alimentary canal of fish. It has been found that electrical stimulation of the sympathetic gives rise to a motor response in the dogfish, and that the vagus has little or no effect on the gut movements.

The results described below were obtained while working at The Marine Biological Laboratory at Woods Hole during the summer of 1928. It was originally intended to record the movements of the intestine *in situ*, but, as the spinal dogfish is quite a lively animal, this method was discarded, and perfused strips of intestine used instead.

The nutritive fluid was made up according to the formula of Mines[1] in which urea in concentration of 2% seems to be essential, but was modified by adding sodium bicarbonate, 0.5 gram per liter. The parts selected for study were the ascending limb of the stomach and the spiral intestine. The figures reproduced below were obtained by registering the movements of the longitudinal coat only.

It has been claimed that urea is essential in solutions for perfusing the isolated elasmobranch heart[2]. With this in view the urea content was cut down to 1%, sodium chloride being added to maintain the osmotic pressure of the fluid

constant. On replacing all the urea by sodium chloride, the intestine was found to beat quite well (Fig. I), the contractions were as good as when urea was used. Fig. II shows the movements of the same piece of gut when urea in 2% concentration was restored to the perfusing medium. This change of rhythm was often noticeable in a strip of muscle where urea was kept constant. In all cases, whether urea was cut down to half its original concentration or left out completely, the response to adrenaline was unaltered. A fact not to be overlooked is the absence of sodium bicarbonate from Mines' solution. The addition of this substance no doubt makes it possible to leave out the urea. It is possible that a small amount of ammonia liberated from the urea acts as a buffer. The presence of ammonia in a solution of urea standing at room temperature is easily detected by its odour, and is well marked after twenty-four hours. In some cases urea was not replaced by sodium chloride, yet in spite of this strips of intestine went on contracting with hardly any change in tone or rhythm. The urea would seem to play a part in the intact animal as regards keeping the osmotic pressure of its tissue fluids constant, but does not seem to be essential for isolated organs.

For routine perfusion of intestinal or stomach strips from skates or dogfish, a solution of the following composition was used:— NaCl 1.8%, KCl 0.05%, $CaCl_2$ 0.04%, $MgCl_2$ 0.05%, Urea 1.0%, $NaHCO_3$ 0.05%.

The stomach and the intestines are innervated from two sources—the vagus and sympathetic nerves. By analogy with mammalian tissues one would expect the vagus to be motor and the sympathetic to be inhibitory in action. Electrical stimulation, e.g. of the vagus, in the mammal does not give uniform results, and in view of the results obtained previously, electrical stimulation of nerves was not carried out. It was thought that drugs which affect the nerve-endings might prove suitable in determining the nature of response given by the vagus and the sympathetic.

Pilocarpine was selected for stimulating, and atropine for paralysing, the vagal endings while adrenaline was used for

stimulating the sympathetic. Fig. III shows the results obtained with pilocarpine. The movements were unaffected by this drug, the gut continuing its normal rhythm. The capacity of the container was 75 c.c. so that the dilution of the drug was not excessive. The doses used are sufficient to cause mammalian intestine to contract strongly, but even when the amount of pilocarpine was raised to 10 mg. no effect could be produced. In one or two instances a slight rise in tone was seen but was too small to be of any significance. The inhibitory action of atropine was also absent. In some cases, however, atropine seemed to exert a slight stimulating action. Fig. IV shows the effect of atropine.

On testing adrenaline no inhibition of movements could be demonstrated, but invariably stimulation resulted (Fig. IV.) A rise in tone and increased force of contractions always resulted with adequate doses of this drug. On the other hand, if the intestinal strip has become hypodynamic and lost all traces of rhythmic movements, minute quantities of adrenaline (1:60 million) will call forth slight movements. On increasing the concentration of adrenaline to 1:40 million, the movements become stronger, accompanied by a rise in tone. Raising the concentration of adrenaline still further produces spasm of the muscle, the rhythmic movements being almost entirely absent. Fig. V shows the response of a fatigued piece of stomach to different concentrations of adrenaline. The action of adrenaline on a segment of stomach contracting rhythmically is shown in Fig. IV.

The spiral or large intestine gives results identical with those obtained on the ascending limb of the stomach. Fig. VI shows, at the left, contractions obtained from a strip of large intestine and on the right, the response of the same preparation to adrenaline. Pilocarpine and atropine had no effect on the movements. The spiral valve was also tested, but no response to the drugs used above was ever obtained. Gradual changes in tone were seen in the few preparations tested. A difficulty in connection with the spiral valve is the great thickness of the mucosa which covers it on both sides and through which

drugs can only penetrate with difficulty. Whether the spiral valve carries out rhythmic movements or not cannot be stated at present, but it is conceivable that it undergoes changes in tone, depending on the amount of food in the large intestine. The contractions of muscle of the large intestine surrounding the spiral valve would suffice to move the intestinal contents toward the rectum.

Having found that the sympathetic was the motor nerve in the dogfish and skate, the reaction of the intestine of some bony fish was investigated. In these the vagus turned out to be the motor nerve, judging by the reaction to pilocarpine and atropine and the sympathetic inhibitory, since adrenaline always gave rise to inhibition. Mackerel, herring, minnows (Fundulus) and pipe-fish all gave similar effects with pilocarpine, namely motor. The contractions produced by pilocarpine were inhibited by atropine. Adrenaline in all these types gave pure inhibitory effects. Fig. VII shows the action of adrenaline on the intestine of Fundulus and Figs. VIIIa and VIIIb the reaction of pipe-fish intestine to pilocarpine, atropine and adrenaline. Henderson[3] has found similar results on the stomach of these fishes.

The "Ringer" fluid used for the intestine of bony fishes was of the following composition:—NaCl 1.1%, KCl 0.055%, $CaCl_2$ 0.04%, $MgCl_2$ 0.024%, $NaHCO_3$ 0.05%.

Since there are data that the first nervous elements descending into the intestine are of parasympathetic origin (vagus), these experiments give certain indications of the different course of innervation of the alimentary canal in different animals. Gaskell[4], Abel[5] and Kunz[6] describe the vagal origin of Auerbach's plexus in the intestine of warm blooded animals, while E. Müller[7] finds a similar condition in *Squalus acanthius*.

DESCRIPTION OF FIGURES.

Fig. I. Dogfish. Ringer without Urea.
Fig. II. Dogfish. 2% Urea in Ringer.
Fig. III. Dogfish. 1% Urea in Perfusing Fluid.
A. Normal; B. Pilocarpine 1/4 mg.; C. Pilocarpine 1/4 mg.; D. Pilocarpine 1 mg.; E. Pilocarpine washed out.

Fig. IV. Dogfish. Capacity of Perfusing Bath 75 c.c. A. Normal; B. Pilocarpine 1 mg.; C. Atropine 1 mg.; D. Adrenaline.

Fig. V. Dogfish.
A. Adrenaline 1:15M.; B. Adrenaline 1:30M; C. Adrenaline 1:60 M.; D. Adrenaline 1:40 M.

Fig. VI. Dogfish Male. Ringer containing 1% Urea. A. Large Intestine; B. Adrenaline 1:10 M.; C. Adrenaline 1:10 M.

Fig. VII. Fundulus Small Intestine.
A. Adrenaline: B. Adrenaline.

Fig. VIII. (a) Pipefish Small Intestine.
A. Pilocarpine 1:½M.; B. Adrenaline 1:10M.; C. Pilocarpine 1:500,000; D. Atropine 1:500,000.

Fig. VIII. (b) Pipefish Small Intestine.
E. Pilocarpine 1:100,000; F. Atropine 1:100,000.

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Fig. I

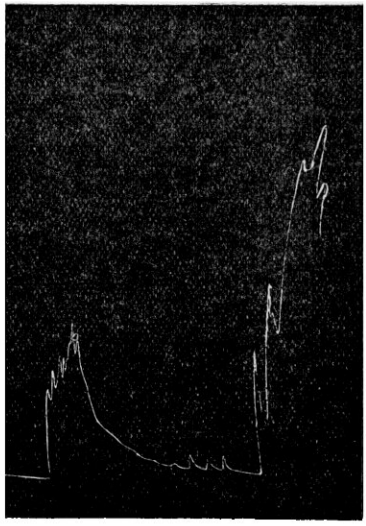


Fig. II

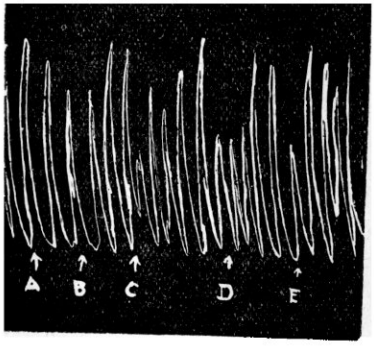
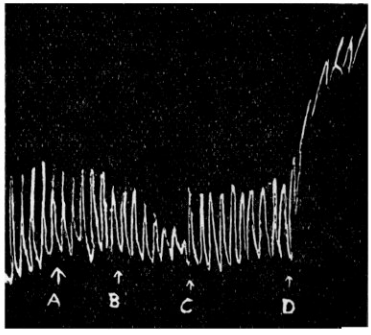


Fig. III



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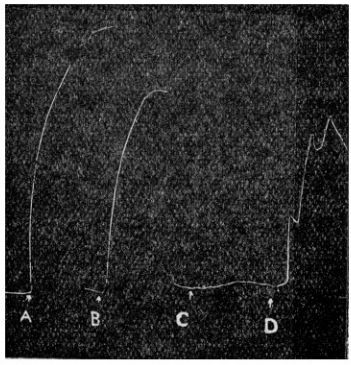

Fig. V

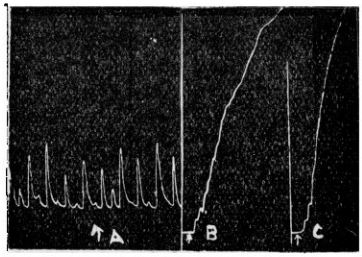

Fig. VI

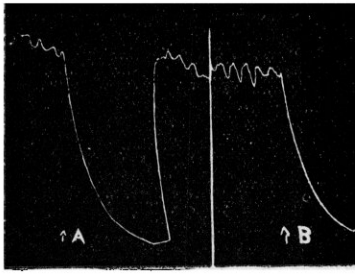

Fig. VII

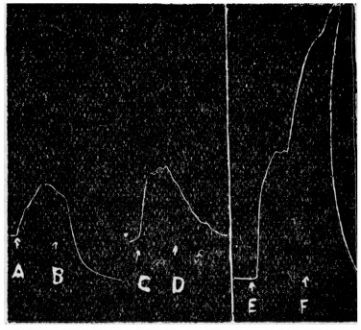

Fig. VIII