THE EFFECT OF HISTAMINE, ADRENALINE AND DESTRUCTION OF THE SPINAL CORD ON THE OSMOTIC PRESSURE OF THE BLOOD IN THE SKATE.

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(Received April 30, 1934).

ABSTRACT.

Preliminary experiments on the effect of histamine and adrenaline on the osmotic pressure of the blood of *Raja erinacea* showed a fall with histamine and no effect with adrenaline. Indications are that the histamine effect takes place through the loss of urea. This may occur either through an alteration in the permeability of the gills for urea or through an impairment of the reabsorption mechanism of the kidney. Both possibilities stand in need of experimental investigation. The effect of histamine on the blood plasma chlorides is uncertain. After ten hours the Cl values were below normal but after fifty-seven hours they were considerably above normal.

INTRODUCTION.

It is a well established fact that the blood concentration of the elasmobranchs is greater than the concentration of the sea water in which the animals are living. In a series of 14 *Raja erinacea* Chaisson[1] found the freezing point depression ($\triangle$) of the blood to be $1.80 \pm .01$°C and the sea water $\triangle$ to average 1.72°C. The greater freezing point depression of blood is due to the high urea content known to be present in elasmobranch body fluids which accounts for about half of the $\triangle$ value (Smith[2]).

Chaisson[1] found that destruction of the greater part of the spinal cord of *Raja erinacea* results in an impairing of the mechanism which normally maintains the concentration of the blood above that of the sea water. Inasmuch as Scott and Denis[3] have shown that in the dogfish the only surface where interchange between external and internal media is possible is the gill membrane, it was presumed that the permeable properties of the gill membrane were altered either di-

[1] Chaisson, A. F. *Proc. N. S. Instit. Sci.* **18**, 23-25, (1931).
[2] Smith, H. W. *J. Biol.. Chem.* **81**, 407-419, (1929).
[3] Scott and Denis. *Amer. J. Physiol.* **32**, 1-7, (1913).

rectly through the destruction of a nervous regulatory mechanism or, what was believed more probable, through shock and the liberation of a histamine-like substance causing increased permeability of the capillary walls.

During the summer of 1933 the opportunity was presented to continue this investigation at the Atlantic Biological Station, St. Andrew's, N. B. Besides the effect of destruction of the spinal cord, the effects of histamine and of adrenaline on the blood concentration were studied.

METHODS.

The animals, *Raja erinacea*, were used after at least three days had elapsed from the time trawled, as MacKay[4] and our own observations show that recently caught skates do not respond at all well to drugs. Throughout the experiment they were kept in tanks of running sea water, the Cl content and $\triangle$ of which were determined at intervals. Some animals were injected with either adrenaline or histamine, others had their spinal cord destroyed. Adrenaline (1 cc. of 1:10,000 to 10 cc. of 1:1,000) and histamine (0.5 to 20 mg.) were injected subcutaneously. (It is worthy of note that MacKay[4] found that histamine had no effect on the circulation of the skate when given intravenously and no local effect when given subcutaneously. However, our determinations were made at least seven hours after injection and so there can be no true comparison with her relatively much shorter time intervals). Destruction of the cord was carried out by passing a long wire needle down the neural canal from just below the medulla to the region of the anus. The skin at the point of insertion of the needle was then sewn watertight. The brain and medulla were not injured; this was noted from the fact that in these operated animals the respiratory and heart rates were good even at the end of 16 days, whereas injury to the medulla leads to almost immediate respiratory cessation.[5]

Gastric juice samples were obtained by aspiration. Blood for chloride and $\triangle$ determinations was obtained as described

[4] MacKay, M. E. *Contr. Can. Biol.* Fish. **7**, 17-29, (1931).

[5] Babkin and M'Gonigle. *Contr. Can. Biol.* Fish. **6**, 315-339, (1931).

by Chaisson[1]; a small glass cannula was ligated into the conus arteriosus and the blood was propelled into a test tube by the force of the heart beat.

RESULTS.

Cord Destroyed. After the first or second day of the operation large amounts of gastric juice of a high acidity (pH 2.0 to 3.8) could be obtained: for example, animal No. 37 yielded a total of 68 cc. in 5 days. That the fluid aspirated from the stomach was gastric juice and not sea water is evident from the low pH and the difference in the $\triangle$s of the sea water and stomach fluid.

The blood $\triangle$ fell progressively from the normal $\triangle$ 1.800°C. to an average $\triangle$ of 1.665°C.; in animal No. 18 it was found to be as low as 1.515°C. nine days after the destruction of the cord. The blood plasma chloride values remained practically unaltered.

Adrenaline. As with animals whose spinal cords had been destroyed, skates that had been given adrenaline were found to contain large volumes of fluid in the stomach. This fluid, however, was probably not the result of gastric secretion but merely sea water that had entered the stomach due to the paralytic dilatation of the cardiac sphincter by the adrenaline. Both pH and $\triangle$ values of the fluid are those of sea water.

The blood plasma chloride and blood $\triangle$ remained unchanged even after prolonged and repeated administration of tremendous doses of adrenaline.

Histamine. The influence of histamine on gastric secretion in the skate is not noticeable. Its influence on the blood concentration, however, is marked; it causes a fall in the blood $\triangle$ to an average of $\triangle$ 1.660°C. with $\triangle$ 1.423°C. as the lowest value recorded. The effect on the blood plasma chlorides is uncertain: at the end of 10 hours the Cl values were found to be all below the normal, and averaged 0.83%, whereas at the end of fifty-seven hours the Cl values were very much above normal, and averaged 0.99%.

Table 1 summarizes the above results. It should be added that though in the normal skate there is a continuous secretion of acid gastric juice, in the starving animal, the amount secreted is very small and does not permit of analyses.[6]

Table 1

Experiment	Gastric Contents		Blood Plasma	Whole Blood	Sea Water		
	pH	△ °C.	Cl%	△ °C.	pH	Cl%	△ °C.
Normal Animal	—	—	0.91	1.800	8.2	1.80	1.720
Cord Destroyed	3.1	1.497	0.93	1.665	—	—	—
Adrenaline	7.6	1.719	0.92	1.798	—	—	—
Histamine	—	—	0.83 to 0.99	1.660	—	—	—

DISCUSSION.

Our investigation confirms previous evidence that the osmotic pressure of the skate's blood is diminished when the spinal cord is destroyed. We found also that it falls when histamine is injected but is not affected by adrenaline. It is conceivable that the fall in the osmotic pressure of the blood is due to loss of the inorganic constituents, chiefly of the chlorides which make up the greatest part of the mineral compounds of the blood. However, after destruction of the cord the blood plasma chlorides remain unaffected and histamine diminishes them only in the first hours following its administration but later on they return to, or even exceed, their initial value. No definite explanation of this observation can be given but it is possible that the variations in the Cl values were partly due to a shifting of the Cl from plasma to corpuscles. Therefore it seems more probable that the lowering of the osmotic pressure of the blood in these cases is due to a loss of urea, a substance which normally is responsible for about half of the blood △ value.

[6] Babkin and Friedman. *Amer. J. Physiol.* **109**, 3, (1934).

The question arises, by what path is the urea eliminated from the body? Nothing is known about the excretory capacity of the skin and skin glands in the skate. According to Smith[7], "the gills of elasmobrånch fishes are relatively impermeable to urea, and in addition, the kidneys of these animals conserve this substance by reabsorbing it from the glomerular filtrate." But Babkin and Komarov[8] have shown that the gastric mucosa excretes urea in concentrations up to 1%. It is not known if the intestinal mucosa has the same property. Since histamine does not activate a gastric secretion it is improbable that the urea is lost through the gastric mucosa. Again, the animals could not be regarded as starving. One of two hypotheses would explain the histamine effect: either the permeability of the gills is altered by histamine and urea can be passed through them or else the reabsorption mechanism of the kidney for urea is impaired. Both of these suppositions require experimental investigation. In the case of the destruction of the spinal cord, the lowering of the blood Δ may also be explained by a functional impairment of either the gills or the kidney. However, since the destruction of the cord results in a "paralytic" gastric secretion which often is very profuse[6], a part of the blood urea may be excreted with this secretion.

Judging from the fact that adrenaline does not influence the osmotic pressure of the skates' blood, it is probable that it does not affect the reabsorption powers of the kidney nor, under the conditions of our experiments, does it alter the permeability of the gill membrane. The results do not support the supposition made by Keys and Bateman[9] that adrenaline increases the permeability of the elasmobranch gill.

We wish to express our deep appreciation to Prof. B. P. Babkin for his interest in and criticism of the problem, and our thanks to the Biological Board of Canada for the facilities afforded.

[7] Smith, H. W. *Sigma Xi Quart.* **21**, 141-151, (1933).
[8] Babkin and Komarov. *Contr. Can. Biol.* Fish. **7**, 13-15, (1931).
[9] Keys and Bateman. *Biol. Bull.* **63**, 327-336, (1932).