ADAPTATION OF CUTANEOUS TACTILE RECEPTORS

V. The Release of Potassium from Frog Skin by Mechanical Stimulation*

By HUDSON HOAGLAND and MORTON A. RUBIN

(From the Physiological Laboratory, Clark University, Worcester)

(Accepted for publication, November 1, 1935)

The hypothesis that the liberation of potassium from the epithelial cells of frog's skin is responsible for the adaptation of the tactile endings (Hoagland, 1934, 1935 *a*, *b*) raises the question whether potassium is actually liberated from the skin on stimulation. The following experiments were devised to test this.

A piece of skin 1 x 2 cm. was cut from the back of a frog and placed in Ringer's solution for 5 minutes. The Ringer's was continually agitated in the region of the skin so as to insure complete washing away of electrolytes released from the cut epithelial cells. The skin was then mounted, inner side down, on a glass microscope slide by means of two rubber bands and again washed with Ringer's to eliminate electrolytes that might have been squeezed out of the skin as a result of the manipulation involved in mounting it. Several pieces of fine glass rod were cemented onto the microscope slide lengthwise, giving a corrugated effect, to allow washing the under side of the skin with a stream of Ringer's delivered by a medicine dropper, the tip of which was pulled out to a small diameter. The slide with the skin was placed vertically in a small glass container and the air jet used in the stimulating experiments previously described (Hoagland, 1933) was directed on the skin. 10 cc. of Ringer's solution was placed in the vessel and the air jet was turned on, producing repetitive stimulation of a spot at a frequency of 140 per second with a nozzle pressure of 400 mm. Hg. During stimulation lasting for 3 minutes the 10 cc. of Ringer's was repeatedly streamed down the under side of the skin by means of the 2 cc. medicine dropper. Some twenty such washings were delivered during the 3 minutes that the stimulation lasted. The "wash" was then collected in a clean vial and tightly stoppered to be analyzed later.

Control experiments were carried out in the following manner. A sample of skin was washed and mounted on the slide in the same way as previously described.

* This investigation has been aided by a grant to one of us (H. H.) from the National Research Council.

The skin was then given twenty 2 cc. washes with 10 cc. of Ringer's but without being stimulated by the air jet. The Ringer's from these control experiments was collected and then analyzed along with the other washes.

To prevent personal bias from entering the analyses, only one of the authors knew which of the samples was from a control or a "beaten" experiment. The vials were numbered and given to the other investigator (M.A.R.) for analysis. Only after the analyses were completed did the analyst learn the nature of the contents of the vial. Occasionally samples of plain Ringer's solution were placed among the vials without the knowledge of the analyst.

The wash was analyzed only for potassium, calcium, phosphate, and chloride ions. The analytical procedures were those indicated in a previous paper (Rubin, 1936), with the exception of calcium. For calcium the method described by Cameron and White (1930) was used. Table I shows the results of the analyses. The term "beaten" in the table refers to samples of wash from skin stimulated by the interrupted air jet.

TABLE I

Ion	No. beaten analyses	Excess in wash (average)	Extremes	No. control analyses	Excess in wash (average)	Extremes
		mg. per cent	*mg. per cent*		*mg. per cent*	*mg. per cent*
K	11	0.67	0.33–1.16	13	0.06	0.03–0.11
Ca	6	None	None	5	None	None
PO_4	4	None	None	4	None	None
Cl	4	None	None	4	None	None

The mean excess of 0.67 mg. per cent of K is equivalent to an increase of K in the Ringer's of 9 per cent. Experimental error ± 2 per cent.

From Table I it is evident that, of the four ions investigated, potassium is the only one passing out of the skin on stimulation with the air jet. It is possible that some other ions may be "squeezed" from the epithelial cells and do reach the nerve endings in the region of stimulation, but they must be almost negligible in quantity as compared with potassium, since the latter passes out into the wash. No attempt was made to analyze the wash for other ions because the four ions investigated are the most abundant in the skin, and the other ions would not be expected to have as an appreciable inhibitory effect on the excitability of the tactile nerve endings as does potassium.

Potassium in frog's skin is extremely diffusible. This can be demonstrated by soaking a piece of skin in about ten times its weight of distilled water. In six such experiments (Table II) the amounts

(128 ± mg. per cent of skin) found in the water at the end of 2 hours are nearly equal to that found in the skin itself. The skin on the back and sides of *Rana pipiens* contains about 133 mg. per cent potassium (Rubin, 1936).

That potassium is released from the epithelial cells of frog's skin when it is stimulated by the air jet may be regarded as established by these experiments. Calcium, phosphate, and chloride do not appear to be released. There still remain the possibilities that (1) other inorganic ions may be freed from the epithelial cells and exert some influence in very small concentrations, although it is improbable as Hoagland (1936) has demonstrated that only calcium has an effect on the tactile endings similar to that of potassium, but the effect is

TABLE II

Potassium Dialyzed from Frog's Skin by Distilled Water

Mg. K/100 gm. wet skin
135.3
126.7
131.9
120.5
128.8
124.8
Mean.....128.0

essentially irreversible, while the potassium effect is readily reversible; or (2) some organic humor (acetylcholine (?), histamine (?)) may be involved to complicate the picture. Preliminary evidence has been obtained, to be presented in a subsequent paper, which indicates that something, probably of an organic nature, may be liberated from the frog skin when it is beaten by the air jet and to a lesser degree when it is washed without the beating. It is, however, quite possible that this substance may have very little, or nothing, to do with adaptation.[1]

[1] Since this paper went to press, one of us (M. A. R., unpublished) has precipitated potassium in frog skin by a modified form of Macallum's method. The potassium, in the form of orange-yellow crystals of potassium-sodium-cobaltinitrite,

SUMMARY

Potassium is released from the epithelial cells of frog's skin on stimulation by an interrupted air jet. This evidence is consistent with the hypothesis that potassium is involved in the adaptation of the tactile nerve endings in frog's skin.

CITATIONS

Cameron, A. T., and White, F. D., 1930, A course in practical biochemistry, New York, The Macmillan Company, 172.

Hoagland, H., *J. Gen. Physiol.*, 1933, **16,** 911.

Hoagland, H., *J. Gen. Physiol.*, 1934, **18,** 255.

Hoagland, H., 1935*a*, Pacemakers in relation to aspects of behavior, New York, The Macmillan Company.

Hoagland, H., *J. Gen. Physiol.*, 1935*b*, **19,** 221.

Hoagland, H., *J. Gen. Physiol.*, 1936, **19,** 943.

Hogg, B. M., *J. Physiol.*, 1935, **84,** 250.

Rubin, M. A., *J. Gen. Physiol.*, 1936, **19,** 935.

---

is concentrated in the epidermis, a little is found around the gland cells, and very little, if any, in the corium. The free, tactile endings, which adapt rapidly, terminate between the epithelial cells and are, therefore, in close proximity to cells most rich in potassium; the slowly adapting pressure receptor endings (Hogg, 1935) are found in the corium (Syrocki, unpublished).