

Dependence of Potassium Ion and Sodium Ion Exchange on Light in the Crayfish Retina

H. Stieve *, T. Malinowska, and D. Sonnemann

Institut für Neurobiologie, Kernforschungsanlage Jülich GmbH

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The exchange (release resp. uptake) of K^+ , Rb^+ , and Na^+ ions in the isolated retina of the crayfish *Astacus leptodactylus* with the surrounding medium was measured using flame photometry and radioisotopic techniques.

1. The net release of K^+ into a Na^+ and K^+ free, choline containing medium, as measured by means of flame photometry, is increased by illumination.
2. The release of $^{86}Rb^+$, used as an indicator for K^+ , in retinas preloaded with the radioisotope is also increased during the period of illumination.
3. The net release of Na^+ into a Na^+ and K^+ free medium containing choline, as measured by means of flame photometry, is decreased by illumination.
4. The $^{22}Na^+$ uptake of the retina is increased by illumination. The increase is greater after an illumination period of 20 min than after one of 2 min.
5. The uptake of $^{22}Na^+$ of the retina in the dark is increased when the retina has been previously poisoned by ouabain.
6. The results of these experiments are in agreement with the assumption that: a. Na^+ -ions are the main carrier of the inward current causing the depolarizing receptor potential of *Astacus* photoreceptor cell membranes and b. the repolarization is caused by a corresponding outflow of K^+ -ions.

Introduction

Numerous data have been published which lead to the conclusion that sodium and potassium fluxes following light stimulus are responsible for the receptor potential of the arthropod photoreceptor cell (Stieve¹; Stieve and Wirth²; Stieve *et al.*³; Stieve and Malinowska⁴; Millecchia and Mauro^{5, 6}; Mc Brown *et al.*⁷; Lisman and Brown⁸; Fulpius and Baumann⁹; Wulff^{10, 11}). However, besides the paper of Holt and Brown¹² we know of no publications of direct measurement of ionic fluxes through visual cells of arthropods. The ion movements underlying the receptor potential in other invertebrate photoreceptors were investigated of DePont *et al.*¹³; Duncan *et al.*¹⁴; Hagins and Adams¹⁵.

The interpretation of the ionic mechanism of the excitation of the photoreceptor cell is generally considered analogous to that which is known about the excitation mechanism of the nerve membrane, especially of the squid giant axon. Since the electrical measurements show certain important differences between the ionic dependence of the membrane

potential of the squid nerve and the arthropod photoreceptor (*e.g.* Stieve¹; Stieve and Malinowska⁴) it is necessary to study the ionic fluxes directly in more detail. Furthermore there are indications that the ionic mechanisms of the receptor potential of *Astacus* are somewhat different than those of *Limulus*, for instance there seems to be no Ca^{2+} -influx into *Limulus* visual cell during excitation in contrast to crustacean light sense cells such as *Astacus* and *Balanus*^{1, 7, 16}.

It is the aim of this paper to measure the exchange of K^+ and Na^+ ions in the dark and in the illuminated *Astacus* retina by means of flame photometric and radioisotopic methods. This paper is thought to be the beginning of a number of measurements of the ionic movements in the isolated retina of the crayfish compound eye by means of radioisotopes.

Material and Methods

Isolated retinas from compound eyes of the crayfish *Astacus leptodactylus* Eschscholz were used for the investigations. The retinas were dissected from the eyes by a method described elsewhere (Stieve and Wirth²; Stieve *et al.*¹⁷).

Requests for reprints should be sent to Prof. Dr. H. Stieve, Institut für Neurobiologie der Kernforschungsanlage Jülich GmbH, D-5170 Jülich 1, Postfach 365.



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The preparation consists only of the layer of receptor cells, pigment cells and the cut postretinal fibres behind the basilar membrane. In this preparation some areas of the surface of the visual cells are in direct contact with the extracellular fluid, and are not surrounded by a special layer of glial cells or pigment cells in contrast, for instance, to the visual cells of the *Limulus* eye (Krebs¹⁸).

The measurements of the light stimulated retinas were compared with those of dark adapted retinas. The retinas were illuminated continuously with white light from a tungsten lamp or rhythmically by means of flashes of 0.5 sec duration with a flash interval of 1 sec. The intensity of the light was ca. 5300 lx white light.

The experiments were carried out in van Harreveld's solution¹⁹. In experiments where sodium and potassium free solution were required the sodium and potassium ions were replaced by 212.3 mM choline chloride. The pH-values of all solutions used were close to 7.5.

In all radiometric experiments 0.1 mM/l Na_2HPO_4 and 10 mM/l glucose was added to the van Harreveld solution.

Radioisotope solutions:

1. van Harreveld solution containing 5 mM K^+ /l and ca. $8 \cdot 10^{-6}$ mM $^{86}Rb/l$ (^{86}Rb — von Amersham Buchler GmbH and Co. KG, Braunschweig). Specific radioactivity of the solution was about 2 $\mu Ci/ml$.
2. van Harreveld solution containing approximately $9.1 \cdot 10^{-4}$ mM $^{22}Na/l$ (specific activity about 2 $\mu Ci/ml$), that is about $4.4 \cdot 10^{-4}$ per cent of the sodium ions present in solution.

In a number of experiments ouabain (1 mM/l final concentration) was added to the extracellular solution to inhibit the active sodium and potassium transport.

The experiments were carried out at room temperature or at 15 °C.

Flamephotometric measurements

For the flamephotometric measurements the retina was placed on a millipore filter in a vessel with a volume of approx. 600 mm³ and perfused with a continuous flow of the saline (approx. 0.3 ml/min). The streaming solution contained neither sodium nor potassium ions but equivalent amounts of choline ions.

The outflowing saline after having flown around the retina was collected for 10 min periods and the potassium or sodium concentration in each sample was determined. During the first 60 min (pre-

period) of the experiment the retina was allowed to adapt to the dark and to the temperature of the experiment which was 14.5–15.5 °C. In this time 6 samples were taken. The retina was then illuminated for 5 or 10 min by steady light and then dark adapted during the following 60 min. The control experiments were performed in the dark for the corresponding time.

The determinations of K^+ and Na^+ concentrations were performed with a Zeiss, Spektralphotometer PMQ II, flame photometer. The sensitivity for potassium and sodium measurements was respectively 0.1 $\mu g K^+/ml$ and 0.5–4 $\mu g Na^+/ml$.

Radioisotopic measurements

The excised retinas were placed in the polyethylene test vials of the Packard spectrometer, containing non streaming radioactive solution and transferred by means of a small platinum wire-loop from one test tube with non-active washing solution to another. All washings were done in a dim red light.

^{86}Rb as an indicator for K^+ -fluxes

It was assumed (see below) that the visual cell cannot discriminate very well between rubidium and potassium^{13, 15, 20, 21}.

Loading procedure:

After dissection from the eye the retinas were loaded with $^{86}Rb^+$ by allowing them to incubate in a van Harreveld solution containing a small amount of $^{86}Rb^+$ in addition to the normal amount of 5 mM K^+/l (see above). The temperature was maintained at 15 °C during the incubation. During this period the retinas were illuminated for 30 min by steady light and afterwards kept in the dark for a certain period, usually 90 min. Illumination and dark adaptation during the incubation period was done in order to load a sufficient amount of the radioisotope in the retina.

In order to measure the effect of loading time on actual Rb^+ uptake by the retina, the retinas were allowed to incubate for differing lengths of time after illumination. They were then put in non-active van Harreveld solutions three times for 20 min intervals.

The main period of the experiment — the rubidium efflux as light-response:

In order to measure the ^{86}Rb efflux the retina was transferred at 2 min intervals into a new test tube which contained 2 ml of non-active van Harreveld solution. After 3 intervals in the dark, some retinas were illuminated for 30 sec by steady light during three more sampling periods. Afterwards

the retinas were again out in the dark and were transferred at 2 min intervals into fresh van Harreveld solutions.

These release of ^{86}Rb of the illuminated retinas was compared with that of the retinas which stayed in the dark for the whole corresponding period.

The measured rubidium release was normalized for a statistical evaluation of the experiments. The average of the first three samples was taken as the reference value (100 per cent). The uptake of ^{86}Rb by the retinas was referred to the amount of tissue water of the retina which was determined by a wet weight- and dry weight-determination. The content of ^{86}Rb in the washing solutions and in the retinas was determined by means of a Packard liquid scintillation-spectrometer model 3320.

^{22}Na as indicator for Na^+ fluxes

An expected increased uptake (or eventually a release) of sodium of the retinal cells caused by illumination was measured by different procedures. The excised retinas were incubated in 2 ml non-streaming, non-active van Harreveld solution for 60 min in the dark. Afterwards they were put into test tubes containing 2 ml of van Harreveld solution containing ^{22}Na for 20 min in the dark at 15 °C. After the loading procedure, some of the retinas were illuminated for various lengths of time in active solutions and transferred seven times into non-active van Harreveld solutions. Each test tube contained 5 ml of van Harreveld solution and the retinas remained for about 10–15 sec in each one. The radioactivity of the retina in the seventh test tube was determined together with 5 ml of the washing solution.

The control experiments were performed with the second retina of the same animal treated identically as the other one but without illumination. The obtained radioactivity was standardized in the comparison with the radioactivity of the retina of the other eye of the same animal. ^{22}Na was determined in each of the seven test tubes by means of a Packard γ scintillation counter model 3003/578.

The illuminations were applied as light flashes in order to produce a sodium influx as high as possible since we believe that the sodium influx is especially great during the transient phase of the receptor potential.

Standardization of results

The obtained ^{22}Na content of the retina in the first group of experiments was referred to the water content of the retina. Later it was compared to the protein content of the retina determined by the Lowry method²². Since this determination was cali-

brated using cattle albumin the protein content of the retina was expressed in mg albumin equivalent.

Since during our experiments it turned out that the right and the left eye of a crayfish might differ quantitatively from one another in the sodium uptake, care was taken in the later experiments always to alternate the eyes which were illuminated; in one experiment the left retina was illuminated and in the other, the right eye. The reference retina of the same animal was always treated in the same manner as the illuminated retina to insure that the only difference between the two retinas was the illumination.

Results

1. Flame photometer

1. Potassium

In one set of experiments ($n=8$) the retinas were kept in streaming sodium- and potassium-free, choline-containing van Harreveld solution and illuminated for 10 min by steady light in the main-period, following the 60 min preperiod; in corresponding experiments the retinas stayed in the dark for the whole duration of the main period.

Fig. 1 shows the evaluation of these experiments. As the result of the illumination there occurs a transient increase of the potassium concentration in the perfusing solution through the experimental chamber (~ 190 per cent) which is explained by an increased potassium efflux (Fig. 1). This increase is not to be seen in the samples in which retinas were kept in the dark.

2. Sodium

In two series of 7 experiments the retinas were kept in sodium- and potassium-free, choline containing solution, illuminated for 5 or 10 min and tested to see if the sodium efflux of the retina changed upon illumination. Illumination occurred after the 60 min preperiod.

In both series a decrease of the sodium efflux occurring transiently during and after the illumination was found (Table I).

This decreased sodium efflux caused by light is in contrast to the observed light-induced (10 min illumination) increase of ^{22}Na uptake in experiments with normal external sodium and potassium concentrations in a choline-free medium (see below).

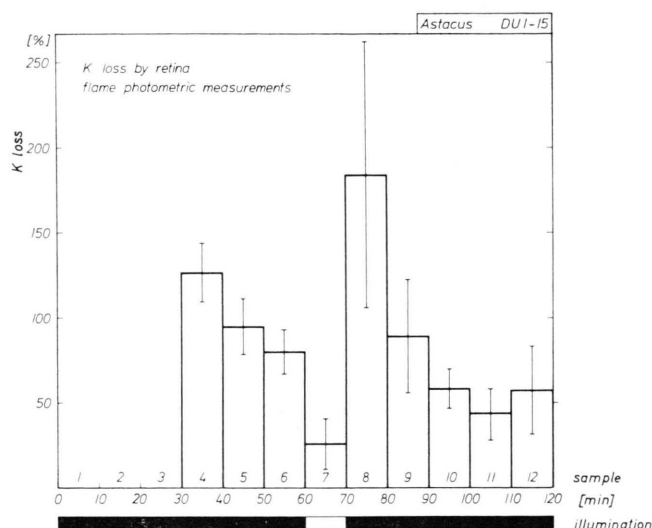


Fig. 1. Potassium efflux from the *Astacus* retina ($n=8$) incubated in sodium- and potassium-free, choline containing van Harreveld solution after 10 min steady illumination (5300 lx) (mean \pm S.E. of mean). Flame photometric measurements.

Table I. Sodium efflux from the *Astacus* retina incubated in sodium- and potassium-free, choline containing van Harreveld solution for varying conditions of illumination. Series I: 10 min illumination, 5300 lx; Series II: 5 min illumination, 1000 lx. 15 °C. Flame photometric measurements (means \pm S.E. of mean).

Sample period [min]	Series I ($n=7$) % of reference value * (4–6 period)	Series II ($n=7$) % of reference value **
Preperiod		
30–40	103 \pm 26	104 \pm 20
40–50	100 \pm 26	119 \pm 23
50–60	95 \pm 19	77 \pm 16
Main period		
light \rightarrow 60–70	36 \pm 12	30 \pm 16
70–80	40 \pm 12	45 \pm 21
80–90	44 \pm 15	61 \pm 21
90–100	62 \pm 32	46 \pm 11
100–110	60 \pm 30	35 \pm 7
110–120	184 \pm 117	39 \pm 16

* absol. reference value 12 \pm 4 mM Na⁺/min.

** 21 \pm 10 mM Na⁺/min.

II. Radioisotopes

1. ⁸⁶Rb⁺ as indicator for potassium

1.1. The loading of the retina with ⁸⁶Rb⁺

In 6 groups each of seven experiments single retinas kept in the van Harreveld solution containing ⁸⁶Rb⁺ were illuminated for 30 min with steady light and remained in the radioactive solution in the darkness for 0, 15, 30, 60, 90 and 180 min at 22 °C temperature. Following this period the retinas

were washed quickly three times with inactive saline and after that the radioactivity remaining in the retina was measured.

The radioactivity was referred to the amount of tissue water. The average wet weight of the retina was about 1.1 \pm 0.2 mg; the dry weight about 0.7 \pm 0.1 mg (tissue water 64.1 \pm 2.4%).

Fig. 2 shows the results of these experiments. The uptake of radioactivity in preparations increases with increasing incubation time until

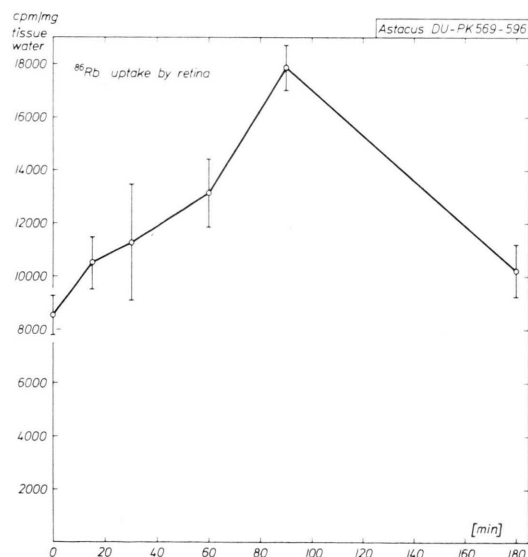


Fig. 2. Uptake of ⁸⁶Rb⁺ into *Astacus* retina in the darkness. Samples were previously illuminated for 30 min (5300 lx, steady light) in radioactive van Harreveld solution (22 °C) (mean \pm S.E. of mean). Radiometric measurements.

90 min. Longer incubation periods yield a smaller uptake of radioactivity. This decrease in uptake of $^{86}Rb^+$, is not easy to understand and will be discussed later.

1.2. $^{86}Rb^+$ -efflux from the loaded retina

In seven experiments the single retinas were loaded in van Harreveld solution containing $^{86}Rb^+$, during an illumination period of 30 min followed by a subsequent dark period lasting 90 min.

After that the retinas were washed in an inactivate solution ($3 \times$ for 20 min). In the following main period the non-active solution surrounding the retina was changed every 2 min (9 times). After three changes in the dark the retinas were illuminated with steady light during three periods in the first 30 sec of that every period. During the next three 2 min-periods the retinas were kept in the dark again.

Fig. 3 shows a significant increase up to about 150 per cent of the Rb^+ -efflux due to the illumination. This increased efflux is reversible. After illumination the efflux of ^{86}Rb returns to its former level in the dark. In the group of experiments, in which the retinas were kept in the dark (during the main period) after the loading period, no corresponding increase of potassium efflux was indicated.

2. $^{22}Na^+$ uptake by the retina

In 8 groups of at least seven pairs of eyes the light induced uptake of sodium by the retina was studied. The retinas were kept in the dark in an inactive

solution for 60 min. Following this they were put into a van Harreveld solution containing $^{22}Na^+$ for 20 min. After this dark preperiod the retinas were illuminated for different periods by repetitive light flashes (2–20 min) in the various series (see Table II) and washed seven times. The radioactivity of the washing solutions was counted, each one separately, and the radioactivity remaining in the retina and in the last washing solution was determined together as the last sample.

The results were normalized in comparison to the Na^+ -uptake of the non-illuminated retina from the other eye (reference retina).

2.1. Comparison of left and right retina

In one series of experiments 11 pairs of retina from eleven crayfishes were investigated. Both retinas were handled separately and were kept in the dark for the entire period to compare the $^{22}Na^+$ -uptake of the left retina with the radioisotope-uptake of the right one. In all other respects the preparations were treated as in the other sets of experiments.

Surprisingly enough we found a difference in the activity of retinas from left eyes as compared with retinas from right eyes of the same animal. Left retinas gave a value of 948 ± 81 cpm/mg alb. equivalent while the retinas from the right eyes showed a $^{22}Na^+$ uptake of 1166 ± 65 cpm/mg alb. eq. which is 131 ± 12 per cent of the activity in the left retina ($p=0.035$). However, the albumin equivalent of both retinas did not differ significantly. The left retinas had on the average 2.2 ± 0.2 mg albumin equivalent/retina. The albumin equivalent content

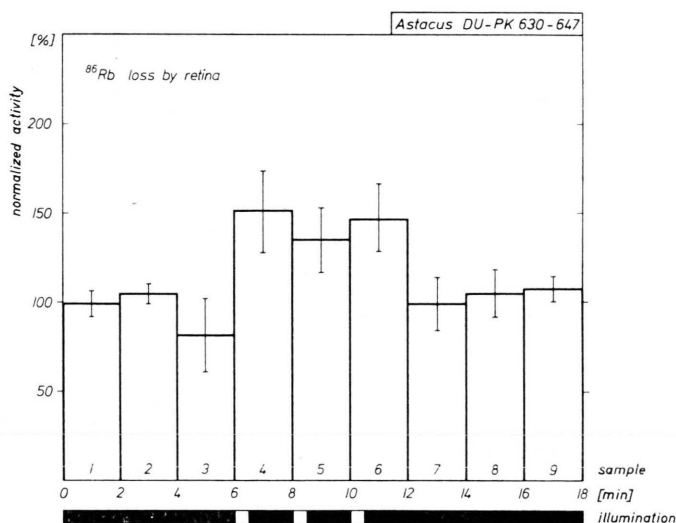


Fig. 3. $^{86}Rb^+$ efflux from the preloaded *Astacus* retina into inactive van Harreveld solution before and after illumination 3×0.5 min. This graph shows the main period of experiment. In the preperiod (180 min) retinas were illuminated 30 min in radioactive solution, 90 min dark adapted and washed out three times (3×20 min). Radiometric measurements.

of the right retinas were 98 ± 7 per cent of the left retinas.

These control experiments were the last experiments of all the series of experiments described here. Therefore for the other series we had to re-evaluate our other experiments and could only use those experiments in which we could compare an equal number of right and left illuminated retinas with corresponding dark retinas of the same animals.

2.2. $^{22}\text{Na}^+$ -influx as light-response for various times of illumination

2.2.1. Flash-illumination period — 20 min

In these experiments the retinas were illuminated by repetitive light flashes of 0.5 sec duration with a repetition rate of one flash per one second. The total light exposing time in this group of experiments was 20 min of which the effective light exposure-time was 10 min. The results are compiled in Table II. The illuminated retinas contained significantly more $^{22}\text{Na}^+$ (123 ± 6 per cent) than the dark retinas ($p = 0.005$).

The measurements of the washing solutions showed changes in radioactivity corresponding to the measurements of the retinas. In the illuminated retinas there is a significant increase of sodium released into the surrounding medium as compared to the dark retinas.

2.2.2. Flash-illumination period — 2 min

In a second series the light-exposing time for seven retinas was only 2 min flash light, the results show that the sodium uptake of the illuminated retinas is not significantly increased as compared to the dark retinas.

Other experiments with 15 min light-exposing time show intermediate values which cannot be compared with the experiments here for reasons stated above (left and right retina-differences).

2.3. Ouabain application

2.3.1. 40 min before illumination

In this series both the illuminated (20 min) and the dark retinas were treated as in the first series, but 1 mM/l ouabain was added 40 min before illumination or at the corresponding time in the control "dark" experiment. The results show a highly increased uptake of radioactivity in both the dark and the illuminated retinas. The increase of $^{22}\text{Na}^+$ -uptake of the illuminated retina compared to the dark retina, however, was not significant. The illuminated retinas had 107 ± 7 per cent of the radioactivity of the dark retinas.

2.3.2. At the beginning of illumination

In this series the retinas were also poisoned by ouabain, but 1 mM/l ouabain was added immediately at the beginning of the light period (20 min) or at the corresponding time to the control experiment in the dark. In this case also, both the dark and the light retinas showed a somewhat higher sodium uptake as compared to the non-poisoned retinas, but there is still significantly higher uptake of sodium in the illuminated retina.

Discussion

It is reasonable to assume that the ionic exchange of potassium and sodium is done mainly by the visual cell and that the pigment cells etc. can be neglected for their effects on a first approximation.

Table II. Uptake of $^{22}\text{Na}^+$ into *Astacus* retina during 20 min incubation in radioactive van Harreveld solution expressed in cpm pro mg albumine equivalent (mean \pm S.E. of mean). Conditions of illumination:

- a. 20 min light period:
20 min flashing period
b. 2 min light period:
2 min flash-light
- } 0.5 sec flash duration, frequency $1 \cdot \text{sec}^{-1}$.

Applied ouabain concentration 1 mM OU/l.

Treatment	Dark	[cpm/mg alb. eq.] Light	Light in % of dark	n	P
20 min light	731 \pm 43	896 \pm 65	123 \pm 6	12	0.005
2 min light	803 \pm 66	856 \pm 55	109 \pm 9	7	0.350
20 min light OU added 40 min before light	1385 \pm 103	1453 \pm 99	107 \pm 7	8	0.350
20 min light OU added at light begin	957 \pm 82	1124 \pm 99	120 \pm 7	12	0.015

In our experiments with crayfish retinas we have to account for two types of ionic movements, a. passive exchange movements down the ionic gradients and b. active transport of potassium ions into the cell, and sodium ions out of the cells by means of metabolically driven transport mechanisms (sodium-potassium pump).

The flamephotometric measurements are measuring somewhat different parameters than the radiometric measurements. With the flamephotometer we measure the net efflux of potassium or sodium whereas the results of radioisotope experiments in our conditions are a measure of the exchange of the labeled ions primarily in one direction. This means for instance, that the measured influx of $^{22}Na^+$ can be accompanied by a more or less remarkable efflux of non-labeled sodium ions which escapes our measurements.

The differences in the sodium uptake between retinas from the left and the right eye are very surprising to us, especially since the protein content measured as albumin equivalent by means of the Lowry method did not show differences. We do not know any remarkable difference in the treatment of the two retinas which could account for this phenomenon.

Potassium

The measurements reported here show an increased potassium efflux from the retinal cells caused by light. The increased $^{86}Rb^+$ efflux determined radiometrically is in close agreement with the increased potassium efflux determined by flame photometric measurements. This increased potassium efflux is also in close agreement with the results of Holt and Brown¹² and fits our conception of the nature of the membrane potential and its change caused by illumination. We assume an influx of positively charged ions (presumably mainly sodium) caused by illumination and a compensative efflux of potassium ions.

The question whether the potassium permeability is increased in the course of the receptor potential (see: Holt and Brown¹²) cannot be answered by our experiments reported here.

The experiments show that using ^{86}Rb as a radioactive marker for potassium current yields qualitatively the same results as the flame photometric measurements of potassium. This makes it

probable that the visual cell membrane cannot distinguish very well between potassium and rubidium as was assumed from literature data for other excitable-including photoreceptor cell-membranes^{5, 6, 14, 20, 23-26}. The observed significant decrease in uptake of ^{86}Rb in the last period of the experiment may be due to a deterioration of the retina during the long sojourn in the unstirred saline at room temperature.

The results of the flame photometric measurements in which choline was used as the most prominent extracellular ion (which influences certain properties of the cell membrane^{1, 2}) are qualitatively the same as the experiments where $^{86}Rb^+$ was used in otherwise unchanged van Harreveld solution. This means that the influence of choline on the cell membrane does not strongly effect the potassium fluxes.

Sodium

The experiments show an uptake of radioactive sodium from the retina in the dark and an increased uptake caused by illumination. One has to expect that the increased sodium uptake caused by illumination depends upon the number of absorbed light quanta. Our experimental results agree with this assumption. The 2 min period of illumination causes an insignificant increase whereas 20 min illumination shows a strong effect.

Under conditions of ouabain poisoning there is a considerably higher Na^+ -uptake in the dark a passive inflow of sodium — probably in exchange for potassium — which is so large, that no differences between the illuminated and the non-illuminated retinas can be noted if the duration of poisoning is long before the onset of the illumination period.

When the blocking of the active transport is done immediately before the illumination there is still an increased sodium uptake in the dark as compared to non-poisoned retinas and still a significant increase of sodium uptake caused by light. The measurements in series No. 2.3.2. are in a way intermediate between the non-poisoned series No. 2.2.1. and the series No. 2.3.1. where ouabain is supplied long before exposure of the retina to light.

These experiments are in agreement with our assumption (Stieve¹⁷) that under our experimental conditions the sodium pump activity is normally considerably high in the photoreceptor cell under

consideration. The retinal compartments between which the ion exchange occurs are most likely relatively small. The intracellular volume of the reticular cells is relatively small as compared with the large amount of ionic currents caused by light. Some of the extracellular spaces surrounding the reticular cells are very small as compared to other excitable cells as for instance in the giant axon of the squid (see Krebs^{18, 27}). Therefore a high pump activity seems to be necessary sustain ionic gradients during and after stimulation. It should be expected that after a longer poisoning period or after several illuminations the increase in the Na⁺-influx should be smaller than in the unpoisoned retina.

The long lasting decrease in sodium efflux into the Na⁺-free, choline containing saline caused by light, as measured in the flame photometer, is in opposition to the radiometric measurements with the ²²Na where the sodium uptake is increased by light. Originally we expected a light induced increase of Na⁺ efflux in the flame photometer measurements due to a light induced increase of Na⁺-permeability. The main difference in the experimental conditions of the two types of experiments consists of the fact that in the flame photometer measurements the extracellular solution contained almost no sodium and potassium, but a very high amount of choline whereas in the radioisotope measurements the extracellular constitution of the solution was normal (*i. e.* normal sodium and potassium and no choline). It could be possible that the differences in the extracellular solution are responsible for the observed differences; the extracellular composition seems to influence only the sodium exchange and not the potassium exchange as seen above.

Concluding remarks

The results are in close agreement with the assumption that sodium ions carry a main component of the current which depolarizes the reticular cell

membrane in response to a light flash and that this current is opposed by a potassium outward current which repolarizes the cell membrane after the end of the light stimulus. So the mechanism of the photoreceptor potential of the crayfish eye can be summarized as follows: The light stimulus causes an increase of the permeability to sodium ions and other ions such as Ca²⁺ and Mg²⁺ (Stieve^{1, 3}; Brown and Blinks²⁸) which causes a depolarization of the cell membrane. This depolarization causes an outflow of potassium ions.

After the stimulus has ended the sodium permeability decreases and the cell repolarises, due to the potassium outflow. Experiments of Holt and Brown¹² indicate that the K⁺-permeability of the visual cell membrane of the ventral eye of *Limulus* is increased during illumination. Our own experiments (Stieve, Malinowska⁴) with the *Astacus* retina treated with TEA favour the assumption, that there is no additional increase of K⁺-permeability responsible for the repolarization after a short light flash — assumed TEA acts in the reticular cell membrane in the same way as on the K⁺-permeability in the squid and frog axon membrane.

We now think it more probable that the K-permeability increases during illumination. This could mean that either TEA acts somewhat differently on the crayfish photoreceptor cell membrane than on the squid nerve membrane or that the light induced permeability change opens "light" channels which are permeable both for Na⁺ (and Ca²⁺ ions) as well as to some degree for K⁺ ions.

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