Movements of 86Rb+ during Trapping Behaviors in an Aquatic Insectivorous Plant Aldrovanda vesiculosa

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An aquatic insectivor Aldrovanda vesiculosa has many traps each of which is a leaf-blade adapted to entrapping small aquatic animals. A trap consists of a pair of semicircular lobes, which are convex outward and connected together by a midrib. When one of the sensory hairs on the inner surface of the lobes is stimulated mechanically, an action potential spreads over the lobes and abruptly the trap is shut; the lobes bend upward, come into contact with each other at their borders, and make a bladder so that prey is enclosed in it. After the shutting the trap begins to reopen very slowly, unbending of the lobes lasting a several hours. The cause of the shutting is believed to be a sudden loss of turgor in the motor cells which are inner epidermis and localized in a certain area (motor zone) on each lobe of the traps.[1] Ion leakage in a detectable amount from the motor cells with the turgor loss can be expected from the experimental results of other seismonastic plants.[2, 3]

In this study, we intend to determine how much amount of ions in the motor cells are released during the shutting and re-entered during the reopening by exploring the movements of $^{86}Rb^{+}$ used as a tracer of K^{+}. The activity of $^{86}Rb^{+}$ in traps or pond water was measured with a liquid scintillation counter (Aloka, LSC 671). Moreover, to locate the motor zones the areas re-enterd with $^{86}Rb^{+}$ during the reopning will be found by autoradiography.

$^{86}Rb^{+}$ release during the shutting. A plant was immersed into artificial pond water (APW) containing $^{86}Rb^{+}$ (1.5 μCi/ml) for 3 days to feed the radioisotopes into the cells. The traps were carefully cut off from such a plant without giving stimulus and washed 4 times with cold APW. When a sensory hair was bent to stimulate with tip of a micropipette (50 μl) the trap was shut immediately so that the pipette tip was enclosed in the bladder formed by shutting. The APW remained in the bladder was then taken up with the pipette and submitted to counting of the radioactivity. The cold APW in a dish in which the trap has been shut and the trap after removing content in the bladder were also submitted to counting. Table 1 shows these results.

Since the radioactivity leaked with shutting is 0.25 % of that in the trap (Table 1) and the total volume of the motor cells is 3.2 % of that of trap lobes, 7.8 % of the total activity in the motor cells flowed out during the shutting. However, this must be underestimated because we could measure only the activity leaked through the inner surface of the trap, and could not detect the activity flowing out in intercellular spaces.

$^{86}Rb^{+}$ uptake during the reopening. Thirty two open traps were cut off from a plant and immersed into the hot (5 μCi/ml) APW. A half of them were

stimulated and shut at the same time of the immersion. The other half remained unstimulated so that the traps were left open. At 1 hr or more intervals, each one trap stimulated and unstimulated was washed 5 times and submitted to counting of the activity.

The $^{86}Rb^{+}$ contents in the tissues increased linearly with time (Fig. 1). In the stimulated traps the rate of the uptake was higher than that in the unstimulated ones within 3 to 4 hr after the stimulation. This seems that the trap reopening requires to regain an amount of ions which has released during the shutting. Actually the end of the higher-rate uptake (A-B in Fig. 1) coincides with the end of the reopening. Since dinitrophenol (DNP) which prevents the formation of ATP almost inhibited the uptakes (Fig. 1), K^{+}-influx in the trap must be an active ion transport. DNP inhibits the trap reopening also.

Localized uptake of $^{86}Rb^{+}$ in the reopening traps. Fig. 2 shows autoradiographs of stimulated and unstimulated traps, which have been immersed in the hot APW for 2 hr. In the unstimulated trap exposed silver grains were scattered evenly over the trap except for the midrib, a vertical white line in the picture. In the stimulated trap the grains were localized on an area near the midrib in either side of it. This area seems to be motor zone because the cells in this zone accumulate a more amount of ions than in the remaining area during the reopening.

References

1) Ashida J., Mem. Coll. Sci. Kyoto Imp. Univ. Ser. B, 9 (1934) 141.
2) Sibaoka T., Annu. Rev. Plant Physiol. 20 (1969) 165.
3) Samejima M., Sibaoka T., Plant and Cell Physiol. 21 (1980) 467.

Table 1. $^{86}Rb^{+}$ activities in the traps and APW surrounding them.

Exp. No.	APW in bladder (cpm)	Volume of bladder (μl)	APW* used for last washing (cpm)	Shut trap after removing bladder content ($\times 10^3$ cpm)
1	686	10.1	11	273
2	375	31.7	13	320
3	312	9.3	10	234
4	1350	12.1	11	287
5	462	17.8	14	265
6	721	11.1	17	187
Mean ±SE	651 ±380	15.4 ±8.6	12.7 ±2.6	260 ±46

* Measured an equal volume of the bladder content.

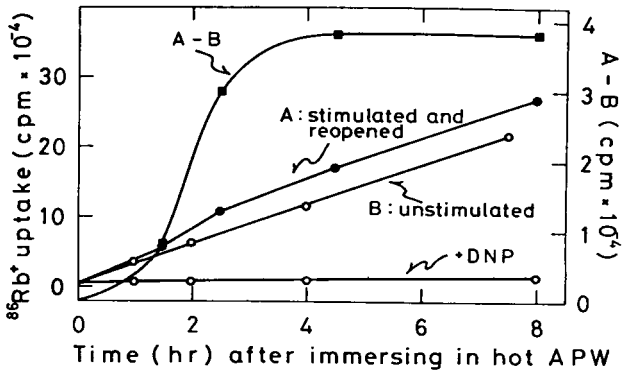

Fig. 1. Time courses of $^{86}Rb^+$ uptake into the stimulated and unstimulated traps. Traps were put in the hot APW at 0 hr.

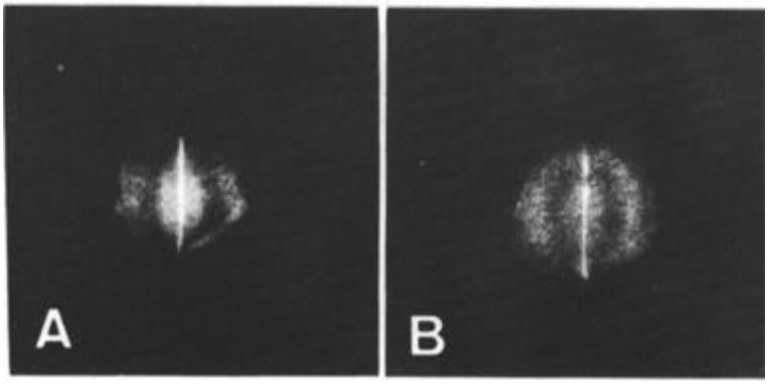

Fig. 2. Autoradiographs made after immersing the traps in the hot APW for 2 hr. A, a trap shut by a stimulus then reopend. B, another trap remaining open without stimulus.