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Lithium and rubidium: Effects on locomotion of planaria *(Dendrocoelum lacteum)*

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Summary. The gliding locomotion of planaria *(Dendrocoeleum lacteum)* was suppressed similarly by LiCl and RbCl.

The use of lithium and rubidium as drugs to treat mental disorders has stimulated interest in their effects on behavioral processes in laboratory animals[3,4]. Invertebrates have been used occasionally to determine whether monovalent cations influence basic behavioral processes[4,5]. Previously, we studied effects of lithium and rubidium on rhythmic contractile movement of jellyfish[6], which have as simple a nervous system as can be found in the animal kingdom[7]. Now, we report on the influence of lithium and rubidium on locomotion in planaria, an animal with a very primitive CNS[5,8].

Materials and methods. We carried out experiments during August–December. Planaria of the species *Dendrocoelum lacteum* (Turbellaria, Tricladida, Pladudicola) were collected from lakes and streams in Aarhus, Denmark. They were 0.8–1.4 cm long and weighed 20–30 mg. They were housed together with free access to food (boiled egg yolk) in a beaker (18 cm diameter) containing 3 l of water from their natural habitat at room temperature (20 °C) with air bubbled vigorously in the beaker at all times. Fluorescent lights provided about 100 footcandles illumination in the vicinity of the planaria.

For tests, the planaria were placed individually in a Petri dish (10 cm diameter) containing 10 ml of water from the natural habitat (medium). A piece of white graph paper divided into squares (1 mm × 1 mm) by light blue lines was located under each Petri dish. Planaria that crossed 25–60 lines during 1 min after 15 min in the Petri dish (baseline conditions) were selected for experiments. 48 of the about 80 planaria examined met this criterion. After baseline recordings, the planaria were assigned at random to 1 of 4 equal groups. Each planaria was used only once. 3 groups had a solution (195 mmoles) of either NaCl, LiCl or RbCl added automatically to the medium in the center of the Petri dish by an infusion pump at a rate of 6 ml/h, while the 4th group received no treatment and served as a control. The media were not stirred because preliminary observations indicated that mechanical disturbances impaired activity of planaria. The number of lines crossed by each planaria was measured every 5 min for 1 min during 30 min. Analysis of variance was used to determine the statistical significance of the results. After test, 7 planaria in each group were selected at random, rinsed in saline, weighed and frozen. In addition, samples of media were taken at the center and along the edge of Petri dishes to which LiCl had been added. The concentration of sodium and potassium was determined by flame photometry and that of lithium and rubidium was measured by spectrophotometry.

Results. The figure presents the effects of lithium, rubidium and sodium on the rate of locomotion of planaria. Inspection of the data shows that lithium and rubidium reduced the rate of locomotion of planaria. Statistical analysis indicated significant differences for the overall effects of treatments (main treatment effect $p < 0.001$) as well as for pairwise comparisons between lithium and control, lithium and sodium, rubidium and control and rubidium and sodium (p's < 0.001), but not between lithium and rubidium or sodium and control. It is evident that locomotion tended to decrease as the concentration of lithium or rubidium increased, and statistical analysis demonstrated significant differences for the overall effect of the amount of salt added (main concentration effect $p < 0.001$) as well as for the amount of lithium or rubidium added (p's < 0.001). There was also a significant tendency for locomotion to decrease during the experiment in the control group ($p < 0.01$), but the decrease in locomotion in planaria treated with either lithium or rubidium was significantly greater than that observed in the controls (p's < 0.001).

Concentration of sodium, potassium, lithium and rubidium in planaria *(Dendrocoeleum lacteum)* treated with 195 mmoles solutions of NaCl, LiCl or RbCl added to 10 ml medium for 30 min at a rate of 6 ml per h. Values are means ± SEM for 7 planaria per group

	Cation concentration (mmole/kg)[a]			
	Sodium	Potassium	Lithium	Rubidium
Untreated	16.1 ± 2.5	30.7 ± 2.7	0	0
NaCl-treated	39.9 ± 8.8	36.5 ± 8.6	0	0
LiCl-treated	18.7 ± 1.9	36.2 ± 2.6	11.4 ± 2.0	0
RbCl-treated	16.5 ± 1.2	38.2 ± 1.5	0	10.5 ± 0.8

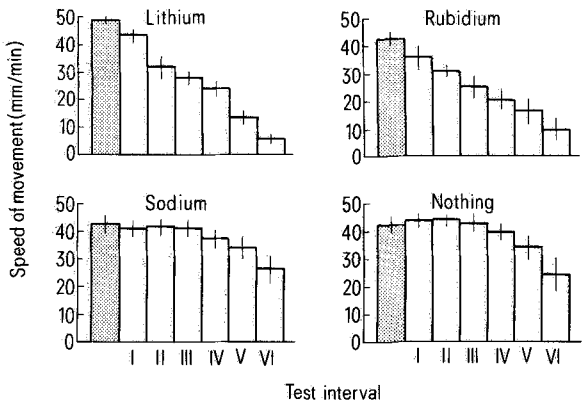
[a]The initial concentrations of sodium, potassium, lithium and rubidium in the medium were 1.5, 0.2, 0 and 0 mmoles, respectively.

The concentration of lithium at the center and the edge of Petri dishes to which LiCl had been added ranged between 60 and 70 mmoles and 40 and 45 mmoles, respectively. Visual inspection of planaria during tests provided no signs of chemotactic locomotion, but the usual elongated shape of planaria changed to an oval form with ruffled edges in those treated with LiCl or RbCl. No changes were evident in the other 2 groups.

The table shows the concentration of sodium, potassium, lithium and rubidium in planaria after the 30 min test.

Discussion. The present findings show that lithium and rubidium influence the movement of planaria. Most previous studies show lithium and rubidium to influence behavior in opposite ways[6,9,10]. In the present study, however, lithium and rubidium both had an activity-suppressant action. We suppose that the gliding locomotion shown by planaria in our test was governed by exploratory, food-seeking and photophobic responses. Thus, lithium and rubidium appear to have similar actions on behavior mediated by these stimuli in planaria.

Previous studies show that the gliding locomotion of planaria is governed by the movement of cilia[5] and the secretion of mucus[11,12], processes that probably require energy supplied by ATP[13]. In addition, biogenic amines seem to be involved in locomotion of planaria[14]. Lithium and rubidium are also known to influence enzymes involved in ATP production[15,16] as well as the metabolism of some monoamines[17,18]. Perhaps effects of lithium and rubidium on ATP-dependent energy systems and monoamines play a role in their activity-suppressant actions in planaria. Further studies are clearly needed to determine whether these speculations are correct. Nevertheless, the present findings support the notion that phylogenetically low animals may be of use in studies on basic effects of monovalent cations on behavioral processes.



Effect of lithium, rubidium, sodium or nothing (control) on the speed of locomotion of planaria *(Dendrocoelum lacteum)*. The speed of movement was first recorded under baseline conditions (stippled bars). Then solutions of LiCl, RbCl or NaCl (195 mmoles) were added to the 10 ml medium for 30 min at a rate of 6 ml per h during which time the locomotion of each planaria was recorded for 1 min every 5 min. Test intervals I, II, III, IV, V and VI correspond to observations made 5, 10, 15, 20, 25 and 30 min, respectively, after baseline measurements. Each bar corresponds to the mean ± SEM for 12 planaria.

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Inhibition of DNA synthesis in erythroleukemic cells by a liver protein fraction

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Summary. The liver inhibitory factor (LIF) extracted inhibits in vitro the proliferation of erythroleukemic cells. This inhibition takes place 24 h after LIF treatment and is promptly reversible on removal of the factor. ^{3}H-thymidine autoradiography shows that the effect is probably due to a blockage of the cells at the end of G_1.

A liver protein fraction has been isolated which inhibits hepatoma cells in vitro[2], and delays hepatic regeneration in vivo[1-3]. This inhibitory substance (LIF) also blocks the proliferation of various kind of tumoral cells within 24 h; it has been shown that this effect is not due to a toxic influence on the cells[2]. The present work was undertaken to