limitation in the supply of thymus-seeding cells in mice of a similar age.

Donald Metcalf

Cancer Research Laboratory,
Walter and Eliza Hall Institute,
Royal Melbourne Hospital,
Victoria, Australia.

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Role of Sialic Acid in Potassium Transport of *L*1210 Leukaemia Cells

The cell membranes of various tumour strains possess large quantities of sialic acid and a strong, negative surface charge[1-5]. This charge, which is partly due to the ionized carboxyl group of sialic acid[2,4,5], may be important in influencing physiological properties such as transport of ions[6]. Since tumour cells seem to concentrate K^+ ions better than normal cells[7-9], we have begun to investigate the regulation of K^+ fluxes by sialic acid.

*L*1210 mouse leukaemia cells, which had been maintained in the ascitic form, were washed at 3° C with isotonic saline until free of red blood cells. The *L*1210 cells were then incubated for 90 min at 37° C in the incubation media shown in Table 1. Sialic acid was removed from the leukaemia cells by the addition of neuraminidase (*Vibrio cholerae*), which was present at a final concentration of 50 units/ml. Under these conditions the sialic acid released is derived essentially from the cell membrane[4]. Free sialic acid was determined by the method of Warren[10]. Na^+ and K^+ ions were assayed by means of a flame spectrophotometer. The optimum wave-lengths were 589 mμ for Na^+ and 766·5 mμ for K^+. ^{14}C-D-Glucose ($5{\cdot}2 \times 10^3$ c.p.m./μmole) and ^{14}C-L-lysine ($1{\cdot}6 \times 10^5$ c.p.m./μmole) were utilized in measuring glucose and lysine uptakes.

Table 1. Constituents of the Incubation Media

Substance	Conc. (mM)	Incubation medium K-free	K-low$_g$	K-free$_{g,l}$	K-low$_{g,l}$
L-Lysine	0·2	–	–	+	+
D-Glucose	4·9	–	+	+	+
KCl	10·0	–	+	–	+
NaCl	130·2	–	+	–	+
NaCl	140·2	+	–	+	–
Na_2HPO_4	4·8	+	+	+	+
NaH_2PO_4	5·2	+	+	+	+
$CaCl_2$	0·9	+	+	+	+
CH_3COONa	5·0	+	+	+	+

The amounts of sialic acid removed by neuraminidase in the K-free medium were 0·21 μmole/100 mg dry wt of cells after 45 min of incubation and 0·23 μmole/100 mg dry wt at 90 min. The quantities of sialic acid released were nearly the same in any of the incubation media listed in Table 1, and the values obtained approximated those determined for the Ehrlich ascites tumour cell[4].

Different incubation media were utilized in the transport investigations, so that the direction of ion flow could be altered. In the K-free medium, K^+ release from the *L*1210 cells accompanied Na^+ uptake; with the K-low$_g$ medium, K^+ uptake accompanied Na^+ release. Table 2 includes the results of typical experiments, in which the removal of sialic acid inhibited K^+ transport, regardless of direction of flow. When the results from a number of experiments were pooled, the differences between the control and the experimental groups were found to be statistically significant ($P < 0{\cdot}05$). Contrary to the relatively sharp response of K^+ ions to the removal of sialic acid, Na^+ transport was only slightly inhibited when neuraminidase was present, and in this case the results did not yield significant differences.

Table 2. Transport in *L*1210 Cells stripped of Sialic Acid

Medium	Substance measured	Time (min)	μmoles/100 mg dry wt. of cells Control	+ Neuraminidase
K-free	K^+	45	−2·56	−1·97 (23)
		90	−4·60	−3·40 (26)
	Na^+	45	+15·1	+13·4 (11)
		90	+13·8	+12·0 (13)
K-low$_g$	K^+	45	+25·0	+16·0 (36)
		90	+26·0	+17·0 (35)
	Na^+	45	−50·0	−45·0 (10)
K-low$_g$	D-glucose	45	+3·13	+3·09 (1)
		90	+6·31	+6·25 (1)
K-free$_{g,l}$	L-lysine	45	+0·208	+0·204 (2)
K-low$_{g,l}$	L-lysine	45	+0·206	+0·207 (0)

(+) Indicates flow into the cells; (−) indicates flow out of the cells. Percentage decreases from the control values are given in parentheses.

Glucose uptake was determined for both the control cells and the enzyme-treated cells and did not appear to differ between the two groups. This finding suggests that transport in general was not affected by sialic acid, and that K^+ transport in particular was not regulated by the action of membrane-bound sialic acid on glucose utilization. Furthermore, although glucose was absent from the K-free medium, the removal of sialic acid inhibited K^+ outflow.

Finally, sialic acid was found not to affect lysine uptake in the absence or presence of K^+ ions, thereby indicating that sialic acid does not influence transport of positively charged substances in general.

Pardee[11] has postulated that altered surface properties of tumour cells may cause a loss of growth control mechanisms. The accumulation of K^+ ions in malignant cells[7-9] may well be such a mechanism, which is partly responsible for the adaptation of these cells to a state of uncontrolled growth. The presence of K^+ ions is necessary for optimal growth of certain bacteria strains[12,13] and plant tumours[14,15]. Lubin[16] has demonstrated a control of protein synthesis by K^+ accumulation. The work recorded here in turn indicates that sialic acid apparently mediates both the inward and outward diffusion of K^+ ions in a leukaemia cell.

We thank Prof. Arthur B. Pardee for his advice. This work was supported by grants 1–*F*2–*CA*–24245–01 and *AI*–04409 from the U.S. Public Health Service.

J. Leslie Glick
Sherwood Githens III

Department of Biology,
Princeton University,
Princeton, New Jersey.

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Occurrence of Fructose-1-phosphate in Tissues of Higher Plants

There is considerable evidence for a role of fructose-1-phosphate in the metabolism of some animal tissues[1]. Relatively little is known about the occurrence of this compound in higher plants. Cardini[2] has shown that preparations of jack bean seeds can cleave fructose-1-phosphate to dihydroxyacetone phosphate and a compound which is probably glyceraldehyde. On the basis of electrophoretic separation, Schwimmer, Bevenue and Weston[3,4] tentatively identified fructose-1-phosphate in extracts of potatoes. We report here evidence for the

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