Sodium-Lithium Exchange in Sarcolemmal Vesicles From Canine Superior Mesenteric Artery

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Exchange of intracellular sodium for extracellular lithium readily occurs in vascular smooth muscle, but the mechanism of this exchange is not known. These studies examined whether a sodium-lithium countertransport system was present in the cell membrane of vascular smooth muscle. A sarcolemmal-enriched vesicle preparation was obtained from canine superior mesenteric artery via a magnesium aggregation and differential centrifugation technique. An outwardly directed gradient for lithium stimulated $^{22}$Na uptake by the vesicles, and an inwardly directed gradient for lithium stimulated $^{22}$Na efflux. These effects were not due to an alteration in membrane potential, and sodium uptake was not stimulated by lithium in the absence of a gradient for lithium. The lithium gradient-stimulated component of sodium uptake was not affected by a change in membrane potential and was insensitive to ouabain. Both sodium-lithium exchange and sodium-proton exchange in sarcolemmal-enriched vesicles were inhibited by two compounds that inhibit the sodium-lithium countertransport system in red cells, phloretin and quinidine. Ethylisopropylamiloride also inhibited both sodium-lithium exchange and sodium-proton exchange in the vesicles. In support of the possibility that sarcolemmal sodium-lithium exchange and sodium-proton exchange are mediated by a single cation exchange mechanism with affinity for sodium, lithium, and protons, we found that an inwardly directed sodium or lithium gradient stimulated proton efflux, and that the stimulation of sodium efflux by external lithium or protons was not additive. It is concluded from these studies that sarcolemmal vesicles from canine superior mesenteric artery contain an electroneutral, phloretin, quinidine, and ethylisopropylamiloride inhibitable sodium-lithium exchange transport system. This system may be analogous to the sodium-lithium countertransport system in red cells that has been linked to the hypertensive process in man. Sodium-lithium exchange and sodium-proton exchange in the vascular smooth muscle cell membrane may be mediated by a single transport system. (Circulation Research 1988;62:478–485)

It has been recognized for over a decade that incubation of vascular smooth muscle tissue in a lithium-containing solution results in replacement of intracellular sodium ions with lithium.1,2 The mechanism of this exchange, however, is not understood. Although a Na-Li countertransport system is present in the red cell membrane,3,4 it has not yet been determined whether a specific transport system is present in the sarcolemma, which has affinity for both sodium and lithium, and mediates the exchange of one cation for the other. The possible presence of such a transport system in vascular smooth muscle could have important implications because several laboratories have reported that the activity of Na-Li countertransport is elevated in red cells from patients with essential hypertension.5-7

We have developed a method for preparing a membrane vesicle fraction enriched in sarcolemma from the superior mesenteric artery of the dog.8 In the present study, we show that a Na-Li exchange transport system is present in these membranes and that this system shares several properties with the red cell Na-Li countertransport system. Finally, we present data that indicate that sarcolemmal Na-Li exchange and sarcolemmal Na-H exchange may be mediated by the same transport system.

Materials and Methods

Vesicle Preparation

Mongrel dogs of either sex were killed with intravenous pentobarbital, and the superior mesenteric arteries were dissected free. A sarcolemmal-enriched vesicle preparation was prepared by a magnesium aggregation and differential centrifugation technique, as previously described and outlined as follows.8 Adhering veins, fat, and nervous tissue were removed in the cold, and the arteries were thoroughly minced with scissors. The tissue was suspended in 10 ml/g wet wt in (mM) 200 mannitol, 10 tris(hydroxymethyl)-aminomethane (Tris), and 16 N-2-hydroxyethylpiperazine-N'-2-ethanesulfonic acid (HEPES), pH 7.5, at 4°C and homogenized with a Polytron homogenizer. MgSO$_4$ was added to a final concentration of 10 mM, and the homogenate was incubated on ice for 30 minutes. The suspension was centrifuged at 1,035g for 4 minutes, and the resultant supernatant was filtered through four layers of gauze and centrifuged at 48,000g for 30 minutes. The resultant pellet was resuspended...
in 0.5 ml of homogenizing medium with 10 mM MgSO₄, vortexed in the presence of a glass rod, and incubated on ice for an additional 30 minutes. This suspension was brought up to 20 ml by the addition of homogenizing medium with 10 mM MgSO₄ and centrifuged at 1,035g for 4 minutes. The resultant supernatant was centrifuged at 48,000g for 30 minutes to yield the final pellet, which was enriched fourfold in sarcosomal membranes relative to a microsomal fraction. This pellet was resuspended by vortexing in the presence of a glass rod, recentrifuged twice in the original homogenizing medium without MgSO₄, and resuspended to a protein concentration of about 3–5 mg/ml. Protein was measured by the method of Lowry using bovine serum albumin standards.

**Sodium Transport**

The transport of sodium by the vesicle preparation was studied using ²²Na and a rapid Millipore filtration technique (Millipore, Bedford, Massachusetts). In general, the transport of ²²Na was measured under conditions in which the external and intravesicular ion concentrations had been preset to certain values according to the goals of each particular experiment. Aliquots of membranes (30–50 μg protein) that had been preincubated in the desired solutions at 22° C for 90 minutes were incubated with 1 mM ²²Na at 22° C, as described in the figure legends. Uptake was terminated at the desired time points by rapidly diluting the incubating membranes with 3.5 ml of cold (0–4° C) “stop solution” that contained 112 mM MgSO₄, 1 mM Tris, and 1.6 mM HEPES, pH 7.5. The membranes were separated from external media by Millipore filtration (0.65 μm, DAWP filter) and washed three times with 3.5-ml aliquots of cold stop solution. The filters were immersed in scintillation cocktail and counted. Sodium uptake by the membranes was determined by subtracting a filter blank, which was obtained in the absence of membranes. Sodium efflux studies were performed by preincubating vesicles with 1 mM ²²Na for 90 minutes and diluting them 50-fold in sodium-free media at 22° C. At 0- and 30-second time points, the sodium content of the vesicles was determined by the same cold stop, Millipore filtration, and washing technique just described. Sodium efflux was calculated as the difference in the sodium content between the 0- and 30-second time points. Valinomycin was added to vesicles from an ethanolic stock to achieve a valinomycin concentration of 50 μg/ml and an ethanol concentration of 1% (vol/vol). All presented data represent the mean ± SEM of the mean of triplicate determinations.

**Proton Transport**

The transport of protons by the final membrane fraction was calculated as the difference in the sodium content and is trapped in its protonated form if the intravesicular pH is lower than external pH. This results in immediate quenching of acridine orange fluorescence. As the inside acidic pH gradient dissipates, trapped acridine orange leaves the vesicles, thereby reducing fluorescence quenching. The fluorescence signal increases with time until a steady-state level is achieved, at which point protons are in electrochemical equilibrium across the membrane. Vesicles were preincubated with 181 mM mannitol, 40 mM MES, 2 mM Tris, and 3.2 mM HEPES, pH 5.0. Vesicles (15–25 μg protein) were rapidly mixed with 1 ml of external buffer containing 6 μM acridine orange plus 100 mM LiCl, NaCl, or N-methyl-D-glucamine (NMG) CI plus 16 mM HEPES-Tris, pH 7.5. Fluorescence was recorded over time by activating at 493 nm and recording the emission at 530 nm with a Perkin-Elmer (Norwalk, Connecticut) 650-10S fluorescent spectrophotometer attached to a chart recorder, as previously described. The fluorescence data were fitted by computer analysis to the general equation ΔF = c x e⁻ᵏᵗ, where ΔF is the difference between final fluorescence (at 20 minutes) and fluorescence at time, t, after mixing (3–20 seconds), and c and k are constants. In each experiment, the data fitted the above equation with a regression coefficient of 0.99 or greater. The value for k was taken to represent the first-order rate constant for proton gradient dissipation. Statistical analysis of these data was performed using Student’s t test.

**Voltage Measurements**

The effect of an outwardly directed potassium gradient in the presence of the potassium ionophore, valinomycin, was assessed in the sarcolemmal vesicle preparation by monitoring the fluorescence of the voltage-sensitive probe 3,3'-dipropylthiadicarboxyanine (diS-C₃(5)), as previously described. Two milliliters of buffer, at 22° C, containing 55 or 11 mM KCl, plus 2 μM diS-C₃(5), were added to a magnetically stirred polystyrene cuvette in a spectrofluorometer that had previously been zeroed with deionized water. The excitation and emission wavelengths were 620 and 669 nm, respectively. The fluorescence signal was set to 90 units with the sensitivity control, and 10 μl of vesicles (about 50 μg protein), which had been preloaded with 55 mM KCl, was rapidly added to the cuvette. Fluorescence was continuously recorded with a strip chart recorder.

²²Na (200 μCi/μg Na) was obtained from Amer sham, Arlington Heights, Illinois; diS-C₃(5) iodide from Molecular Probes, Junction City, Oregon; and quinidine, phloretin, valinomycin, and acridine orange from Sigma Chemical, St. Louis, Missouri. 5-(N-Ethyl-N-isopropyl) amiloride was synthesized as previously described.

**Results**

**Na-Li Exchange**

The uptake of 1 mM ²²Na was measured in vesicles in the presence of an outwardly directed gradient for either lithium or choline (25 mM Li⁺, 5 mM Na⁺). As shown in Figure 1, the presence of intravesicular lithium stimulated sodium uptake by 89% at a 30-second time point. In this experiment, sodium uptake was measured...
in the presence of equal internal and external potassium concentration and in the presence of the potassium ionophore valinomycin to clamp membrane potential to zero. Thus, the stimulation of sodium uptake by internal lithium could not be explained by an inside positive lithium diffusion potential.

A representative time course of sodium uptake into lithium and choline preloaded vesicles is shown in Figure 2. Sodium uptake by lithium preloaded vesicles was higher at 10-, 20-, and 30-second time points, but uptake at the 90-minute equilibrium time point was the same for both sets of vesicles. Thus, the stimulation of sodium uptake by internal lithium at early time points cannot be explained by an increase in the intravesicular volume or binding of sodium to the membranes.

Figure 3 demonstrates the dependence of sodium uptake on internal lithium concentration. As shown, the rate of sodium uptake progressively increased as intravesicular lithium concentration was raised from 0 to 5 mM. The stimulation of sodium uptake was fully saturated at 10 mM intravesicular lithium. The concentration of internal lithium that half-maximally stimulated sodium uptake was about 3 mM.

To rule out the possibility that the sarcolemmal Na-K pump was responsible for lithium gradient-stimulated sodium uptake, the 30-second uptake of 1 mM 22Na by lithium and choline preloaded vesicles was measured as in Figure 1, in the presence and absence of 1 mM ouabain, an inhibitor of the Na-K pump. Ouabain did not affect sodium uptake by lithium or choline preloaded vesicles. The lithium gradient-stimulated component of sodium uptake in the presence of 1 mM ouabain was 105 ± 7% of uptake in the absence of ouabain (n = 3, p = NS). Thus, the lithium gradient-stimulated uptake of sodium in these studies was not due to operation of the Na-K pump.

To assess the effect of an inwardly directed lithium gradient on sodium efflux, vesicles were preloaded with 1 mM 22Na and diluted into external media containing 25 mM Li or choline. As shown in Figure 4, the 30-second efflux of 22Na was stimulated 80% by external lithium. Thus, an inwardly directed lithium gradient stimulated the transport of sodium in the opposite direction. The stimulation of sodium efflux by external lithium could not be explained by an inside positive lithium diffusion potential since this experiment was performed in the presence of valinomycin and equal internal and external potassium concentration.

To rule out the possibility that lithium merely increased the permeability of the membrane to sodium, the uptake of 1 mM 22Na was measured in the presence of 25 mM internal and external lithium or choline. As shown in Figure 5, the 30-second uptake of sodium was not affected by ambient lithium versus choline. Thus, an oppositely directed gradient for lithium, but not lithium per se, stimulated sodium transport. Taken together, the preceding data indicate that the vesicles used in these studies contain a Na-Li exchange transport system.

**Effect of Membrane Voltage**

To determine the effect of membrane voltage on Na-Li exchange, the uptake of 1 mM 22Na was measured in vesicles preloaded with 25 mM Li or choline, in the presence of valinomycin and an outwardly directed potassium gradient, or equal internal and external potassium concentration. The outwardly directed potassium gradient in the presence of valinomycin should hyperpolarize the vesicles, whereas voltage should be clamped to zero by equal internal and external potassium concentration. The relative membrane voltage of the vesicles in the presence and absence of the outwardly directed potassium gradient was assessed by monitoring the fluorescence of the voltage-sensitive dye diS-C3(5), as shown in Figure 6. When vesicles preloaded with 55 mM KCl were mixed with external solution containing 55 mM KCl and 2 μM diS-C3(5) (Kex = Kint), the increased turbidity of the solution caused the fluorescence to fall to a value that

![Figure 1](http://circres.ahajournals.org/)

**Figure 1.** Effect of an outwardly directed lithium gradient on sodium uptake in sarcolemmal-enriched vesicles. Vesicles were preincubated with (in mM) 25 choline Cl or LiCl, 55 KCl, 40 mannitol, 16 HEPES-Tris, pH 7.5, plus 50 μg/ml valinomycin for 90 minutes at 22°C. The 30-second uptake of 1 mM 22Na was assayed at 22°C in the presence of (in mM) 5 Li, 5 choline, 55 K, 66 Cl, 68 mannitol, and 16 HEPES-Tris, pH 7.5. Values are means ± SEM from five experiments.

![Figure 2](http://circres.ahajournals.org/)

**Figure 2.** Time course of sodium uptake by sarcolemmal-enriched vesicles preloaded with or without lithium. Vesicles were preincubated and the uptake of 22Na at the indicated times was assayed, as described in Figure 1 legend. Values are means of triplicate determinations from a representative experiment.
Dependence of sodium uptake on internal lithium concentration in sarcolemmal-enriched vesicles. Vesicles were preincubated for 90 minutes at 22°C with (in mM) 150 mannitol and 16 HEPES-Tris, pH 7.5, plus 0, 2.5, 5, 10, or 25 LiCl, and enough choline Cl such that total salt concentration equaled 25 mM. The uptake of 1 mM $^{23}$Na was assayed for 30 seconds at 22°C in the presence of (in mM) 5 UCl, 5 choline Cl, 178 mannitol, and 16 HEPES-Tris, pH 7.5. Values are means of triplicate determinations from a representative experiment.

Remained constant over time. When an identical aliquot of the vesicle suspension was mixed with external solution containing 11 mM KCl and 2 μM diS-C$_3$-(5) (K$_{in}$ > K$_{out}$), fluorescence of the solution fell to a lower value than when K$_{in}$ = K$_{out}$, and then slowly rose with time. These findings indicate that the outwardly directed potassium gradient induced an inside negative membrane potential that caused the cationic dye, diS-C$_3$-(5), to enter the vesicles, resulting in fluorescence quenching. As the potassium gradient dissipated with time, the vesicles became less negative inside, diS-C$_3$-(5) effluxed from the vesicles, and fluorescence gradually increased. It would appear, then, that the vesicles were indeed rendered more electronegative inside in the presence of an outwardly directed potassium gradient. Under each voltage condition, the uptake of sodium by lithium preloaded vesicles minus uptake by choline preloaded vesicles was taken as the activity of Na-Li exchange. Although total sodium uptake by both lithium and choline preloaded vesicles was stimulated by the inside negative membrane potential, Na-Li exchange activity was not affected by the change in voltage (Figure 7). These data suggest that Na-Li exchange in vascular smooth muscle is electroneutral, as is the case for Na-Li exchange in the red cell membrane.4,12

Inhibitor Studies

To explore further the possibility that the sarcolemmal and the red cell Na-Li exchangers are homologous systems, we tested whether phloretin and quinidine, two known inhibitors of red cell Na-Li exchange,4,13 would inhibit sarcolemmal Na-Li exchange. In addition, we tested whether these agents would inhibit sarcolemmal Na-H exchange, which has been proposed to be homologous with red cell Na-Li exchange.13,14 Na-Li exchange activity was measured by determining the internal lithium-stimulated component of sodium uptake. Na-H exchange activity was measured by determining the internal proton-stimulated component of sodium uptake, as previously described.4 These measurements were made in the presence and absence of 1 mM phloretin or quinidine. As shown in Table 8, 1 mM phloretin inhibited both Na-Li exchange and Na-H exchange by about 50%. As also shown in Figure 8, 1 mM quinidine inhibited Na-Li exchange and Na-H exchange by 60–70%.

The sensitivity of Na-Li exchange and Na-H exchange in the sarcolemmal vesicles to ethyliosopropylamiloride was also tested. This drug inhibits Na-H exchange in cultured vascular smooth muscle cells.15 As shown in Figure 9, both Na-Li exchange and Na-H exchange were inhibited about 90% by 1 mM ethyliosopropylamiloride and were inhibited about 50% by 0.1 mM ethyliosopropylamiloride.

Proton Transport

The similarities between red cell Na-Li exchange and renal brush border Na-H exchange have prompted several investigators to propose that these transport events are mediated by a single monovalent cation exchanger with affinities for sodium, lithium, and protons.13,14,18 If such a relation exists between Na-Li exchange and Na-H exchange in the sarcolemma of vascular smooth muscle, then Li-H exchange should be demonstrable in sarcolemmal vesicles. This was tested in the following experiment. Vesicles were preloaded with buffer at pH 5.0, and incubated in external solutions at pH 7.5, containing 100 mM Li, Na, or NMG, an ionic strength control. The rate of collapse of the inside acidic pH gradient was monitored by measuring the fluorescence quenching of the pH-sensitive probe, acridine orange. As shown by a representative experiment in Figure 10, external lighth-
Uptake (% Choline Cl)

FIGURE 5. Effect of ambient lithium on sodium uptake in sarcolemmal-enriched vesicles. Vesicles were preincubated with (in mM) 25 LiCl or choline Cl, 55 KCl, 40 mannitol, and 16 HEPES-Tris, pH 7.5, plus 50 μg/ml valinomycin for 90 minutes at 22° C. The 30-second uptake of 1 mM 22Na was assayed at 22° C from an identical external solution. The uptake of sodium by lithium preloaded vesicles was expressed as a percent of uptake by choline preloaded vesicles that averaged 0.17 nmol/mg protein. Values are means ± SEM from four experiments.

Concurrent Na-Li and Na-H Exchange

To test further the possibility that sarcolemmal Na-Li exchange and Na-H exchange are mediated by the same transport system, we determined whether the stimulation of sodium efflux by external lithium and external protons was additive. Vesicles were preloaded with 1 mM 22Na at pH 7.5 and diluted into external solutions containing 25 mM Li or NMG at pH 7.5 or 6.5. As shown in Figure 11, at pH 7.5, external lithium stimulated sodium efflux by 80%. In the absence of lithium, lowering external pH from 7.5 to 6.5 stimulated sodium efflux by 73%. This latter effect is consistent with the operation of the Na-H exchanger. However, when both lithium and increased acidity (pH 6.5) were present in the external solution, the stimulation of sodium efflux was not additive. In fact, sodium efflux under these conditions was not higher than when external lithium or external acidity alone was present in the outside solution. These data are consistent with the possibility that Na-Li exchange and Na-H exchange are mediated by the same system. If such were the case, saturation of the external cation binding site of the exchanger by 25 mM Li, or separately by protons (pH 6.5), would preclude the further stimulation of sodium efflux by the simultaneous presence of 25 mM Li and protons (pH 6.5).

Discussion

Incubation of intact vascular smooth muscle tissue in a lithium-containing solution leads to lithium uptake accompanied by sodium efflux. Exchange of lithium for sodium occurs, at least in part, across the vascular
smooth muscle cell membrane.\(^1\)\(^2\) This exchange process in intact tissue has several features characteristic of a mediated membrane transport system. For example, it has been demonstrated that ouabain-resistant sodium efflux from the cellular compartment of rabbit portal anterior mesenteric vein into an external lithium solution was markedly inhibited by lowering temperature from 37\(^\circ\) C to 2\(^\circ\) C. In addition, removal of calcium from the external media greatly inhibited ouabain-resistant sodium efflux.\(^2\) Nevertheless, the mechanism of Na-Li exchange in intact vascular smooth muscle cell has not been determined. Several different possibilities could potentially underlie this exchange phenomenon. Extracellular lithium could enter the vascular smooth muscle cell via a conductive pathway, such as diffusion or a channel, and electrically induce the efflux of sodium through a conductive pathway for that ion. It is also possible that lithium, once inside the cell, could displace sodium from intracellular binding sites, resulting in higher intracellular free sodium concentration and greater sodium flux out of the cell. A third possibility is that, as in the red cell, a discrete membrane transport system with affinity for sodium and lithium is present in the sarcolemma, which exchanges one cation with the other.

In the present studies, we have demonstrated that an outwardly directed lithium gradient stimulated the uptake of sodium in a sarcolemmal-enriched vesicle preparation from canine superior mesenteric artery. In addition, an inwardly directed lithium gradient stim-

![Figure 8. Effect of phloretin and quinidine on Na-Li exchange and Na-H exchange in sarcolemmal-enriched vesicles. Na-Li exchange was measured by determining the 30-second uptake of 1 mM \(^{22}\)Na by lithium preloaded vesicles minus the uptake by choline preloaded vesicles, as described in the legend of Figure 1, in presence or absence of 1 mM phloretin or quinidine. Na-H exchange was measured by preincubating vesicles for 90 minutes at 22\(^\circ\) C with (in mM) 181 mannitol, 17 Tris, and 28 HEPES, pH 7.5, or 181 mannitol, 40 MES, 2 Tris, and 3.2 HEPES, pH 5.0. The 30-second uptake of 1 mM \(^{22}\)Na was assayed in the presence of (in mM) 1 Cl, 104 mannitol, 8 MES, 47 Tris, and 65 HEPES, pH 7.5, in the presence or absence of 1 mM phloretin or quinidine. The uptake of sodium by vesicles preincubated at pH 5.0 minus the uptake by vesicles preincubated at pH 7.5 was taken as the activity of Na-H exchange. Na-Li exchange and Na-H exchange in the presence of phloretin or quinidine are expressed as percent inhibition relative to these transport rates in the absence of inhibitor. Results are means ± SEM from four experiments.

![Figure 9. Effect of ethylisopropylamiloride on Na-Li exchange and Na-H exchange in sarcolemmal-enriched vesicles. The 30-second activities of Na-Li exchange and Na-H exchange were determined as described in the legend of Figure 8, and the percent inhibition by 1.0 and 0.1 mM ethylisopropylamiloride calculated. Results are means ± SEM from four experiments.

![Figure 10. Effect of external sodium or lithium on the dissipation of an inside-acidic pH gradient in sarcolemmal-enriched vesicles. Vesicles were preincubated with (in mM) 181 mannitol, 40 MES, 2 Tris, and 3.2 HEPES, pH 5.0, for 90 minutes at 22\(^\circ\) C. Five microliters of vesicle suspension (30 \(\mu\)g protein) were rapidly mixed with 1 ml of external buffer containing 6 \(\mu\)M acridine orange, plus (in mM) 100 LiCl or NaCl or NMG Cl, plus 16 HEPES-Tris, pH 7.5. Fluorescence was measured starting 3 seconds after mixing, by activating at 493 nm and recording the emission at 530 nm with a chart recorder (right to left). Data shown are from a representative experiment.\]
suggest the presence of a Na-Li exchange transport system in the sarcolemma.

The Na-Li exchange mechanism examined in the present studies has several features in common with the red blood cell Na-Li countertransport system. The present studies have demonstrated that Na-Li exchange in sarcolemmal-enriched vesicles was insensitive to a change in membrane voltage and was inhabitable by quinidine and phloretin. These features are also characteristics of the red cell Na-Li countertransporter.14,15

Several laboratories have demonstrated that the activities of Na-Li countertransport are elevated in red cells from patients with essential hypertension.17 The relation between this transport system and blood pressure has been obscure; however, increased red cell transport activity could be a marker for increased transport activity of a similar system in vascular smooth muscle. Indeed, Friedman has demonstrated increased ouabain-resistant sodium influx into a lithium solution from intact tail arteries of the spontaneously hypertensive rat versus the Wistar-Kyoto control rat.17

Since lithium is not normally present in the body, the physiological role of Na-Li exchange in the red cell or vascular smooth muscle cell is obscure. Aronson18 and Funder et al19 have independently proposed the hypothesis that red cell Na-Li countertransport could be an operative mode of a Na-H exchange. This proposal is based on the many similarities between the red cell Na-Li countertransporter and the renal brush border Na-H exchanger. Both systems are quinidine inhabitable,3,18 electroneutral monovalent cation exchangers,4,12,19 with affinities for sodium and lithium, greater affinity for lithium than sodium,12,18 and no affinity for cesium, rubidium, or potassium.3,19-21 It is possible that vascular smooth muscle Na-Li exchange and Na-H exchange are mediated by the same transport system.

In the present study, we show that Na-Li exchange and Na-H exchange in the sarcolemmal-enriched vesicle preparation were similarly inhibited by two compounds that inhibit Na-Li countertransport in red cells, phloretin and quinidine.14 We show that ethylisopropylamiloride, which inhibits Na-H exchange in cultured vascular smooth muscle cells,15 similarly inhibited Na-Li exchange and Na-H exchange in the vesicle preparation. We also show that an inwardly directed lithium gradient stimulated proton efflux and that sodium efflux rates, which were stimulated by an oppositely directed gradient for lithium or protons, were not additive. These data are consistent with the single transport system hypothesis. It is of interest to note that Wasserman et al2 found that calcium removal inhibited the exchange of intracellular sodium for external lithium in intact vascular smooth muscle. These data are also consistent with a common identity of the sarcolemmal Na-Li and Na-H exchangers since intracellular calcium has been shown to stimulate the Na-H exchanger in a variety of cell types including cultured vascular smooth muscle cells.22-23 It must be pointed out, however, that it is not yet known with certainty whether the Na-Li exchanger and Na-H exchanger in vascular smooth muscle are indeed the same transport system.

The physiological functions of vascular smooth muscle Na-Li exchange, and the range of transport events mediated by this system in vivo, are not currently known and require further study. Additional investigation is needed to determine whether a link exists between this transport system in vascular smooth muscle and the hypertensive process.

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