Table 1. Response of ATP-ADP exchange activity to NaCl after treatment with *N*-ethyl maleimide (NEM). After the preincubation described in the text, the samples were again incubated for 30 minutes at 26°C with 0.003*M* $MgCl_2$; 0.005*M* tris ATP; 0.0012*M* tris ADP-C^{14}; 0.04*M* tris-HCl, *p*H 7.5; 1.5 μg of microsomal protein in experiment 1 and 2.5 μg in experiment 2; additional salt was added as indicated; the total volume was 25 μl. Control refers to enzyme incubated with H_2O in place of *N*-ethyl maleimide. Under these conditions of the maleimide treatment, adenosine triphosphatase activity is inhibited by about 80 percent.

Additions	Exchange rate (% of control)		
	Expt. 1	Expt. 2	Untreated with NEM
None	23	17	100
$5 \times 10^{-5}M$ NaCl	26		
$1 \times 10^{-4}M$ NaCl	28		
$5 \times 10^{-4}M$ NaCl	60		
$1 \times 10^{-3}M$ NaCl	126		
$2 \times 10^{-3}M$ NaCl	250	312	110
$1 \times 10^{-2}M$ NaCl		800	88
$5 \times 10^{-2}M$ NaCl		500	98
$1 \times 10^{-1}M$ NaCl		450	
$5 \times 10^{-2}M$ LiCl		20	
$5 \times 10^{-2}M$ KCl		18	

percent β-mercaptoethanol in 0.05*M* tris at *p*H 7.5. From this solution, portions were transferred to tubes containing substrates for the exchange reaction. The separation of the radioactive nucleotides was accomplished by a thin-layer chromatographic method.

The calculation of exchange rate (Table 1) is based on the percent of total radioactivity in ATP; a series of control enzyme dilutions was run with each experiment to obtain a standard curve, upon which the exchange rate was plotted. In the control experiments, equilibration occurred to an extent of 18 to 22 percent of the total radioactivity in ATP, corresponding to an exchange rate of approximately 228 mμmole of phosphorus per minute per milligram of protein (*8*).

Treating the particles with *N*-ethyl maleimide inhibits the Mg^{++}-dependent exchange activity about 80 percent (Table 1). The original rate of exchange, however, can be restored by less than $10^{-3}M$ NaCl; in contrast, the reaction catalyzed by untreated particles is insensitive to Na^+. With increasing concentrations of NaCl, the rate of the exchange reaction becomes several fold greater than that of the original Na^+-insensitive exchange. This suggests that Na^+ is not merely reactivating the original Mg^{++}-dependent exchange. Since the marked response to Na^+ is specific (Li^+ and K^+ are ineffective, Table 1), it is reasonable to consider that this Na^+-sensitive exchange may be a component of the *Electrophorus* Na^+-K^+-ATPase. The ATP-ADP transphosphorylation implies the occurrence of a high-energy phosphorylated intermediate in the adenosine triphosphatase reaction (presumably enzyme-bound), and the present experiments indicate that the role of sodium ions in both reactions may involve a transfer of high-energy phosphate. Since the Na^+-sensitive exchange is not evident until after the enzyme reacts with *N*-ethyl maleimide, some change in enzyme configuration may be induced by this reagent.

Longer treatment of the microsomes with the maleimide inactivates the adenosine triphosphatase more completely, but it also begins to inhibit the Na^+-activated exchange rate. There is no significant incorporation of radioactivity from ADP into AMP with control enzyme or maleimide-treated enzyme. The enzyme preparation had previously been found to be virtually free of adenylate kinase activity (*6*).

It should be emphasized that the evidence for the identification of this exchange reaction as a component of the hydrolytic reaction remains purely circumstantial.

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4 May 1964

Root Pressure in Conifers

Abstract. *Exudation of sap was never observed to occur from stumps of detopped seedlings of loblolly pine or white spruce, but measurable exudation occurred from apical root segments, 4 to 8 centimeters long, which were removed from the root systems and observed individually. Fully suberized root segments of loblolly pine exuded as much or more sap than unsuberized roots. Exudation also occurred from detached sugar maple root segments, but not from stumps attached to entire root systems.*

Exudation rarely is observed from stumps of coniferous seedlings or large trees, and it generally is assumed that conifers do not develop root pressure. However, there are enough reports of exudation from conifer roots to indicate that root pressure sometimes is developed. Eaton (*1*) raised *Thuja orientalis* and *Cedrus deodara* in solutions containing high concentrations of salt and, by replacing the salt solution with tap water at the time the tops were removed, he was able to produce exudation. Daniel (*2*) obtained barely measurable volumes of exudate from a single seedling of *Pinus ponderosa* and from two seedlings of *Pinus radiata*. Pitra (*3*) observed exudation from stumps of plants of *Thuja occidentalis, Cupressus horizontalis, Cupressus funebris*, and *Pinus insignis*, but he detected no root pressure exudation in *Juniperus ericoides, Taxus baccata*, or *Picea alba*. Under field conditions exudate has been collected from detached roots of pine (*4*, *5*), spruce (*5*, *6*), and larch (*6*).

Our observations on seedlings of loblolly pine (*Pinus taeda L.*) and white spruce [*Picea glauca* (Moench) Voss] provide new information. The seedlings were grown for several months in a greenhouse in aerated, nutrient solution (*7*) before the experiments were started. Observations were made in the spring, with a root temperature of approximately 24°C. No exudation ever occurred from stumps of detopped seedlings. Likewise, no exudation was ever observed from stumps of detopped seedlings which had been grown in sand or in soil.

The results were very different, however, when individual root tips of loblolly pine and white spruce were removed from the seedlings growing in solution. Apical segments 4 to 8 cm long were removed from root systems, and their bases were sealed into pipettes so the volume of exudate could be observed. The root segments were immersed in nutrient solution during the experiments. All of the 75 root segments studied showed exudation, but the rate was very low, and the total volume exuded over a period of 72 hours was small compared with roots of angiosperms studied at the same time.

Since the roots used were of various sizes, some common basis was needed for comparison of exudation from various species, and the area of the absorbing surface was chosen as the most logical. The average volumes (in microliters per square centimeter of root surface) of exudate obtained in 72 hours were as follows: *Pinus taeda*, 1.0; *Picea glauca*, 2.0; *Betula populifolia*, 5.0; *Liriodendron tulipifera*, 17.0; *Acer saccharum*, 17.5; *Acer rubrum*, 27.5. It was interesting to note that *Acer saccharum* roots behaved like those of the two conifers. Entire root systems showed no exudation, but apical segments of roots showed relatively high exudation.

It usually is assumed that only growing roots exhibit root pressure, but it was found that fully suberized, nonelongating, apical segments of loblolly pine roots also show root pressure. It was slow in developing, and often no exudation occurred during the first 24 hours; but after 80 hours the total exudation from suberized root segments was about 2.5 times that from the unsuberized roots of similar size maintained under similar conditions.

The exudation observed by Dimbleby (*4*) and by White *et al.* (*5*) was from detached roots. The exudate collected from *Picea abies* and *Larix decidua* by Reuter and Wolffgang (*6*) apparently also came from detached roots. All three investigators apparently used roots which had been cut loose from the central axis of the plant. White *et al.* (*5*) used roots which were 0.5 to 1.0 cm in diameter and bore many smaller branches and root tips. The roots used in our study were approximately 1 to 2 mm in diameter and 4 to 8 cm long and were unbranched.

These results pose the interesting problem of why excised apical root segments show exudation although the entire root systems from which they are detached show no exudation. Failure of root systems to exhibit exudation might occur either because of failure to accumulate enough salt in the xylem to produce appreciable pressure or because of a high resistance to water flow through the root system.

The concentration of salt in the xylem solution of loblolly pine was very low, suggesting that it either has a limited capacity to accumulate salt or is unable to retain it in the xylem. However, the salt concentration in the xylem solution from white spruce was considerably higher than that of the solution in which the roots were immersed, indicating that there is no failure to accumulate salt in this species. White *et al.* (*5*) reported pressures of about 80 cm of water in roots of conifers. The resistance to flow in loblolly pine was not high, much larger volumes of water being moved through pine root systems under a pressure of 1 bar than through sugar maple root systems of similar size. The volume of solution pushed through white spruce root systems under 1 bar of pressure was approximately the same amount as that pushed through the sugar maple root systems. There seems to be no obvious explanation for the failure of the intact conifer root systems to show exudation when excised individual roots do show exudation.

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2 May 1964

Turgor Pressures in Phloem: Measurements on Hevea Latex

Abstract. *Hydrostatic pressures in laticiferous phloem tissues of* Hevea brasiliensis *trees exhibit a diurnal fluctuation, with minimum values occurring during the day. Pressure decreases with increasing height up the trunk, the gradient becoming greater under conditions of rapid transpiration. Sieve-tube turgor must be controlled independently if pressure-flow is to occur in the expected direction.*

A number of attempts have been made to measure hydrostatic pressures occurring within plants. For example, measurements were obtained by Marvin on maple sap (*1*), Scholander *et al.* on lianas (*2*), and Bourdeau and Schopmeyer (*3*) and Vité (*4*) on oleoresin exudation in *Pinus* species. No direct measurement on the phloem of actively growing plants appears to have been published, although various estimates based on determinations of osmotic pressure have been made (*5*).

In the investigation described here, hydrostatic pressures in the phloem tissue of trees of *Hevea brasiliensis* were measured with simple capillary manometers similar to those developed by Bourdeau and Schopmeyer (*3*). It is assumed that this hydrostatic pressure is largely the result of pressure within the latex vessels. A small hole is bored in the bark down to the wood and a hypodermic needle fitted to a sealed glass capillary (10 to 12 cm long) is inserted into the hole. Latex flows in rapidly and the pressure is estimated from the difference between the initial and final lengths of the air column in the capillary. Maximum pressure is usually attained within 10 minutes.

Figure 1 illustrates the pressures observed when measurements were made at 3-hour intervals for 1½ days at two heights (50 and 950 cm above ground) on the trunk of a large untapped seedling tree. Five fresh manometers were used for each reading; the maximum pressure observed (rather than the mean) has been taken as the best estimate of the true pressure. During the 1st day, which was fine and sunny, pressure fell to a minimum by 1500 (3 p.m.) and then rose again to become more or less constant during the night. On the 2nd day, the