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Potassium Fluxes during Potassium Absorption by Intact Barley Plants of Increasing Potassium Content[1]

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ABSTRACT

The presence of previously absorbed K in plants caused a marked reduction in the short term influx of ^{86}Rb-labeled K into roots of barley seedlings. The influx values agreed with net K absorption rates into intact plants, thus suggesting that K efflux was negligible in comparison with influx.

Earlier interpretations of a large K efflux component from excised roots approaching equilibrium K concentrations are considered to be due to an underestimation of net K absorption rates resulting from xylem exudation as the K status of the roots increased.

Net rates of salt absorption have long been recognized to be greater for plant tissues of a low salt status than for those of a high salt status (5, 6, 18). This phenomenon appeared to be explained by observations that when K-starved excised roots were transferred to solutions containing K, they maintained a high influx of K but showed a steadily increasing efflux of K as the net K absorption rate into roots declined (8, 9, 14). The observed efflux was assumed to be an exchange process at the site of K influx (9). This explanation was thought to account for the low rates of K absorption observed in barley plants growing for 20 days at continuously maintained concentrations of K in complete nutrient solutions compared with those reported for K-starved excised roots at similar K concentrations in solution (10).

The experiments reported in the current paper were designed to test the validity of extrapolating from results with excised roots to K absorption by whole plants growing with a continuous supply of K. Influx of labeled K into intact barley roots is compared with net K absorption rates into the whole plant as equilibrium K concentrations within the plant are approached. The term "efflux" will be restricted to the sense used by Jackson and Stief (9) to indicate the loss of K from roots to the external solution at the site of K influx.

MATERIALS AND METHODS

Barley seeds, *Hordeum vulgare* L. cv Beecher, were germinated for 24 hr in flasks of aerated deionized water. Ten batches, each of 300 germinating seeds, were then (day 0) evenly distributed on cheesecloth-covered, epoxy-resin-coated wire screens in plastic buckets containing 4 liters of aerated, K-free basal nutrient solution of initial pH 6.4 and composition (μM): Ca, 250; Mg, 100; N (as NH_4^+), 105; N (as NO_3^-), 600; S, 100; P, 5; Na, 10; Cl, 10; Fe (as sequestrene 138), 10; B, 2; Mn, 0.3; Zn, 0.1; Cu, 0.1; Co, 0.04; Mo, 0.02. On day 5, two pretreatments were imposed by transferring five batches of plants to fresh basal nutrient solutions containing either no K (K_0) or 2000 μM KCl (K_{2000}). The temperature of all nutrient solutions was maintained at 20 $\pm$ 1 C, and the pH was maintained above 5.0 by additions of 0.1 N NaOH and by frequent replacement of nutrient solutions (twice before transfer to pretreatments and three times daily after transfer).

The effect of the plant's K status on K absorption by barley seedlings was followed during 3 days of pretreatment at K_0 and K_{2000} by measuring at intervals (0, 3, 7, 24, 31, 48, 72 hr): (*a*) influx of ^{86}Rb into roots of intact plants, (*b*) influx of ^{86}Rb into excised roots, (*c*) the K contents of the plants.

Influx was determined by measuring ^{86}Rb absorption into attached roots (40 plants) or excised roots (approximately 3.5 g fresh wt excised 30 min prior to each experiment) in 2 liters of complete nutrient solution containing 2000 μM KCl labeled with ^{86} RbCl of high specific activity at pH 6.0 $\pm$ 0.1 and 20 $\pm$ 1 C. The validity of using ^{86}Rb as a tracer for short term K absorption into barley roots has been demonstrated (3, 4, 7). Subsamples of plants and roots were removed periodically (5, 10, 15, and 20 min) from labeled solutions, desorbed in cold (2 C) solutions of 5000 μM KCl + 250 μM $CaSO_4$ (30 sec in 250 ml, 15 min in 400 ml), and then briefly rinsed with water. Roots were cut from tops, blotted, and weighed; and the activity was assayed in an end window G. M. tube. Influx was calculated by regression analysis of the slopes of labeled K content with time.

K contents of the K_{2000} plants were determined on separate samples (40 plants) collected each time influx measurements were made and also 96 hr after transfer to K_{2000}. The plants were briefly rinsed in deionized water; divided into tops, roots, and seed reserve; weighed; oven-dried at 70 C; and digested in a nitric-perchloric acid mixture (11). The digests were analyzed for K by atomic absorption spectrophotometry (1). Average net K absorption rates ($\bar{v}$) over successive days after transfer to K_{2000} were calculated from the equation

$$\bar{v} = \frac{(\log_e W_{R_2} - \log_e W_{R_1})(M_2 - M_1)}{(t_2 - t_1)(W_{R_2} - W_{R_1})} \quad (1)$$

where W_{R_1} and W_{R_2} are the fresh weights of roots and M_1 and M_2 are the total quantities of K in the plant at the start (t_1) and completion (t_2) of the absorption period (12, 22).

RESULTS AND DISCUSSION

Influx into Roots of Intact Plants of Increasing K Status. Influx of ^{86}Rb-labeled K into roots of plants transferred to K_{2000} was

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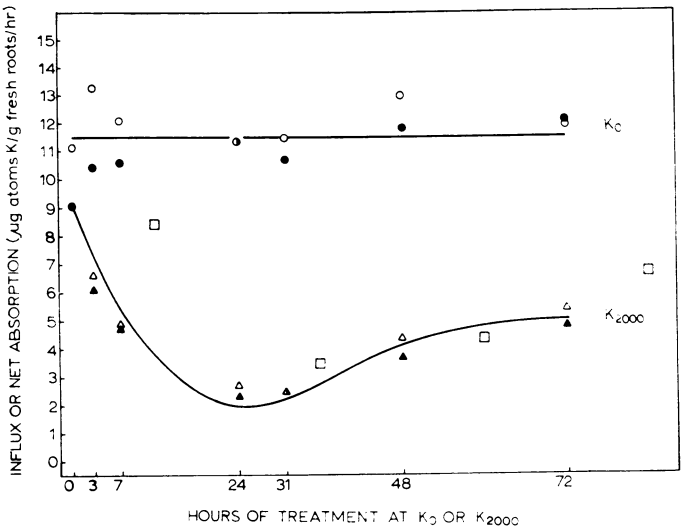

FIG. 1. Effect of time on ^{86}Rb influx into intact (○, △) and excised (●, ▲) roots of barley seedlings after transfer to K_0 and K_{2000} and on net K absorption rates into seedlings (□) after transfer to K_{2000}. Seedlings were grown in full nutrient solutions without K before transferring to K_0 (○, ●), or K_{2000} (△, ▲) solutions. Net K absorption rates by intact barley seedlings were calculated from daily K analyses by equation 1 (see text).

severely depressed ($P < 0.05$) when compared with influx into roots of plants grown continuously at K_0 (Fig. 1). Values for influx into K_{2000} roots dropped to a minimum of 20% of those of K_0 roots within 1 day after transfer to K_{2000}. This decline in influx accompanied an increase in K concentration of K_{2000} plants (Table I). After 1 day, influx into K_{2000} roots increased slightly with time but still remained at values of only 30 to 40% of the influx into K_0 roots. Influx into K_0 roots remained reasonably constant over the period studied, even though K_0 plants showed increasingly severe K deficiency symptoms from 48 hr following transfer (Day 7) and grew less than plants transferred to K_{2000} (Table I).

Negligible activity appeared in the tops of either K_0 or K_{2000} plants during the 20-min influx and subsequent desorption period.

Net K Absorption Rate and Its Relation to Influx. Transferring plants from K_0 to K_{2000} depressed their average daily net K absorption rates similarly to influx (Fig. 1). This suggests that the approach to equilibrium K concentrations in intact barley seedlings results primarily from a reduced K influx rather than an increased K efflux as the concentration of K within the plant builds up. Efflux of K appeared relatively unimportant (Fig. 1) even when the K concentrations of plants reached relatively high values (Table I).

Discrepancies with Excised Root Data. The results of the present work conflict with published conclusions that roots approaching equilibrium K concentrations have a large K efflux component (8, 9, 14). These earlier experiments determined K efflux from the difference between ^{42}K or ^{86}Rb influx and net K absorption by excised roots. The conflict in estimates of the importance of K efflux has arisen from errors in the measurement of K influx and of net K absorption in earlier studies with excised roots.

Earlier workers have made serious errors in estimates of efflux by assuming that net K retention measured net K absorption by excised roots. When excised roots lose K in the xylem exudate during the absorption period, net K retention will underestimate net K absorption. Data from intact plants indicate that only K-starved roots retain K against movement to the xylem. For example, during the first 3 hr after transfer to high K solutions, roots of K-starved barley plants absorbed K rapidly but lost little K to their tops; after 3 hr, with increasing K content of the roots, an increasing proportion of K moved from roots to tops (Table II). Indeed, when K reached a constant high concentration in plants grown in continuously maintained nutrient solutions containing 2000 μM K, the roots retained only 20% of the total K absorbed (10). It seems reasonable to assume that root K status would influence K retention and K movement into the xylem in the same manner in excised roots as in intact plants.

To obtain a valid measure of net K absorption for excised roots, account must be taken of the K exuded from the cut end of the xylem. For example, the data of Pitman *et al.* (17) show that K influx was very much higher than net K absorption rates into excised roots when net K absorption represented only that K retained by roots. However, when an amount equivalent to the K transported to the tops is included in the calculation of net K absorption rate, then K influx becomes equal to net K absorption rate.

Influx of K into excised roots may give results valid for extrapolation to whole plants since, in the present experiment, values of influx into both excised and intact roots were identical, showing an increasing depression of influx with increasing K status of the plant (Fig. 1). Good agreement between influx values into excised roots and roots of whole plants is to be expected if the excised roots are freshly excised and the influx period is short enough to preclude any loss of label in the xylem exudate. The excised roots used in the present experiment satisfy these conditions. Earlier failures to show effects of K status on

Table I. *Changes in Fresh Weight and K Concentration of Barley Seedlings after Transfer from K_0 to K_0 or K_{2000}*

Time after Transfer	Fresh Weight of Whole Plant		K Concentration in K_{2000} Plants	
	K_0	K_{2000}	Tops	Roots
hr	*g*		*μg-atoms/g fresh wt.*	
0	0.16	...	31	22
3	0.15	0.16	36	41
7	0.16	0.15	61	57
24	0.17	0.18	107	85
31	0.17	0.18	118	83
48	0.19	0.19	134	94
72	0.19	0.22	153	96
96	0.22	0.29	172	103

Table II. *Transport of K to Tops of Barley Seedlings after Transfer from K-free Solutions to Solutions Containing K*

Data from Present Experiment		Data from Pitman *et al.* (17)	
Time period	Transport[1]	Time period	Transport[1]
hr	%	*hr*	%
0–3	20.0	1–2	0
3–7	69.7	2–4	33
7–24	69.8	4–11.5	64
24–48	77.3	11.5–23	92
48–72	77.0		
72–96	75.7		

[1] Transport = amount of K moving to tops in a given time period per total amount of K absorbed from the external solution in the same time period.

influx (8, 9, 14) may have arisen from differences in the techniques of measuring K influx into excised roots. For example, influx measurements in very short time periods (8) would have given exaggerated values for absorption because of failure to remove labeled K from cell wall exchange sites: exchangeable K would have accounted for a large fraction of the total root K since no other cations were present in the experimental solutions. In experiments over longer periods of time (9), differences between the published and the present excised root experiments would occur if excision and bathing of roots in KCl solutions in the absence of calcium for long periods caused cell membranes to deteriorate. Values of efflux across these membranes could then assume undue prominence.

K Efflux from Intact Plants. The results of the present experiment establish that efflux of K from roots of intact plants is relatively unimportant compared to influx even when concentrations of K in the plant are quite high. However, this conclusion may only apply when, as in the present experiment, K absorption predominantly occurs through the operation of the high affinity mechanism (2). At higher K concentrations, in the region of the low affinity mechanism of K absorption (2), efflux might assume some prominence. Indeed, accurate determinations of K efflux from root tissues bathing in solutions of high K concentrations could resolve whether K movement across the plasmalemma is predominantly passive (19) or active (20, 21) when the low affinity mechanism of K absorption dominates.

Quantitative, direct measurements of efflux of ions from homogeneous plant tissues have been made by three-compartment analysis (13, 15, 16) in which the change in specific activity of the cytoplasm (the compartment in which incoming K mixes freely) with time after transfer of plant tissue from labeled to unlabeled solutions can be accounted for. Unfortunately, this direct approach cannot be applied to intact plants growing with a continuous supply of K primarily because of the transport to plant tops of a large proportion of K absorbed by the root. Quantitative determinations of the efflux from intact plants must therefore continue to rely on comparisons of influx with net absorption rates. With this technique, the present results establish that, at concentrations of K commonly found in soil solutions, rates of K absorption into intact plants are governed by influx mechanisms and efflux of K is negligible in comparison with influx.

LITERATURE CITED

1. Dean, J. A. 1960. Flame Photometry. McGraw Hill, New York.
2. Epstein, E. 1966. Dual pattern of ion absorption by plant cells and by plants. Nature 212: 1324–1327.
3. Epstein, E. and C. E. Hagen. 1952. A kinetic study of the absorption of alkali cations by barley roots. Plant Physiol. 27: 457–474.
4. Epstein, E., D. W. Rains, and O. E. Elzam. 1963. Resolution of dual mechanisms of potassium absorption by barley roots. Proc. Nat. Acad. Sci. U.S.A. 49: 684–692.
5. Hoagland, D. R. and T. C. Broyer. 1936. General nature of the process of salt accumulation by roots with description of experimental methods. Plant Physiol. 11: 471–507.
6. Humphries, E. C. 1951. The absorption of ions by excised root systems. II. Observations on roots of barley grown in solutions deficient in phosphorus, nitrogen or potassium, J. Exp. Bot. 2: 344–379.
7. Jackson, P. C. and H. R. Adams. 1963. Cation-anion balance during potassium and sodium absorption by barley roots. J. Gen. Physiol. 46: 369–386.
8. Jackson, P. C. and D. G. Edwards. 1966. Cation effects on chloride fluxes and accumulation levels in barley roots. J. Gen. Physiol. 50: 225–241.
9. Jackson, P. C. and K. J. Stief. 1965. Equilibrium and ion exchange characteristics of potassium and sodium accumulation by barley roots. J. Gen. Physiol. 48: 601–616.
10. Johansen, C., D. G. Edwards, and J. F. Loneragan. 1968. Interactions between potassium and calcium in their absorption by intact barley plants. II. Effects of calcium and potassium concentration on potassium absorption. Plant Physiol. 43: 1722–1726.
11. Johnson, C. M. and A. Ulrich. 1959. Analytical methods for use in plant analysis. Calif. Agric. Exp. Sta. Bull. No. 266.
12. Loneragan, J. F. 1968. Nutrient concentration, nutrient flux, and plant growth. 9th Internatl. Congress Soil Sci. Trans. II: 173–182.
13. Macrobbie, E. A. C. and J. Dainty. 1958. Ion transport in *Nitellopsis obtusa*. J. Gen. Physiol. 42: 335–353.
14. Mengel, K. and B. Schneider. 1965. Die K-Aufnahme als Funktion der Influxrate und der Zellpermeabilität, mathematisch und experimentell an der K-Aufnahme junger Gerstenwurzeln dargestellt. Physiol. Plant. 18: 1105–1114.
15. Pallaghy, C. K. and B. I. H. Scott. 1969. The electrochemical state of cells of broad bean roots. II. Potassium kinetics in excised root tissue. Aust. J. Biol. Sci. 22: 585–600.
16. Pitman, M. G. 1963. The determination of the salt relations of the cytoplasmic phase in cells of beet root tissue. Aust. J. Biol. Sci. 16: 647–668.
17. Pitman, M. G., A. C. Courtice, and B. Lee. 1968. Comparison of potassium and sodium uptake by barley roots at high and low salt status. Aust. J. Biol. Sci. 21: 871–881.
18. Sutcliffe, J. F. 1952. The influence of internal ion concentration on potassium accumulation and salt respiration of red beet root tissue. J. Exp. Bot. 3: 59–76.
19. Torii, K. and G. G. Laties. 1966. Dual mechanisms of ion uptake in relation to vacuolation in corn roots. Plant Physiol. 41: 863–870.
20. Welch, R. M. and E. Epstein. 1968. The dual mechanisms of alkali cation absorption by plant cells: Their parallel operation across the plasmalemma. Proc. Nat. Acad. Sci. U.S.A. 61: 447–453.
21. Welch, R. M. and E. Epstein. 1969. The plasmalemma: Seat of the type 2 mechanisms of ion absorption. Plant Physiol. 44: 301–304.
22. Williams, R. F. 1948. The effects of phosphorus supply on the rates of intake of phosphorus and nitrogen upon certain aspects of phosphorus metabolism in gramineous plants. Aust. J. Sci. Res. (B) 1: 333–361.