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Influence of Calcium on Sodium and Potassium Absorption by Fresh and Aged Bean Stem Slices

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ABSTRACT

The influence of Ca on the aging processes of bean stem (*Phaseolus vulgaris*) slices and on the absorption of K and Na by fresh and aged slices was investigated. In the presence of Ca, fresh tissue showed a preferential Na uptake. The preference for Na over K resulted from a differential depressive effect of Ca on absorption of these two ions. In aged tissue Na uptake was also depressed, but K absorption was accelerated, with a net result of a much greater absorption of K than Na.

The presence of Ca in the aging medium promoted the development of K-absorbing capacity as well as an increase in the rate of respiration but did not influence the loss of capacity to absorb Na as tissue aged. This, along with the demonstration that protein synthesis is involved in the development of K-absorbing capacity by aging tissue, suggests that Ca may have an effect on basic physiological processes concerned with development of ion absorption by aging tissue. The influences that Ca may have on the physical and physiological aspects of ion transport are discussed.

Bean stem slices change in ion absorption characteristics as they are kept (aged) in aerated Ca solutions for various periods (30). Freshly sliced tissue absorbs Na readily, much more so than K. In contrast, after aging for 18 hr in $CaSO_4$ solutions, the slices absorb less Na while their capacity to absorb K is much enhanced (30).

The phenomenon of aging involves a myriad of subtle and gross changes in the physiology and morphology of plant cells. It has been demonstrated that by washing slices of aging tuberous roots or stem tissues in various solutions, ion absorption (1, 2, 15, 17, 21, 30, 33, 36–38), respiratory patterns (14, 15, 17, 19, 26, 33, 38), endoplasmic reticulum (9), protein and RNA metabolism (3, 4, 12, 20, 27), and phospholipid turnover in membrane fractions (40) are altered. Plant hormones have also been found to influence the response of isolated tissues as they are aged (30, 34, 39).

When the process of aging is studied in stem slices or any responsive tissue, the samples are usually placed in an aerated solution for various periods. A review of the literature indicated that one of the variables in such investigations was the solution in which the tissue was aged. For example, studies on aging have been carried out in distilled water (1, 17) and in solutions consisting of tap water (19, 21), tris-buffer (36, 37), and various salts (2, 17, 27, 30, 40).

The influence of Ca on ion absorption is well documented (5, 6, 10, 13, 18, 23, 31, 32, 41). It has been suggested that Ca promotes selective ion transport (6, 10, 13, 18, 31, 32) and is involved in the maintenance of membrane integrity (22–25). Correlation between ion leakage and the development of ion-absorbing capacity in storage tissue (9, 21), as well as changes in endoplasmic reticulum (9) and membrane phospholipid (40), suggested that alterations in membrane integrity might be responsible for certain aspects of the aging phenomenon. Since it has been proposed that Ca plays a specific role in the maintenance of structural integrity and selective ion transport, a study was made of the effect that the composition of the solution might have on the development of aging processes and of how Ca influences these processes in bean stem slices.

MATERIALS AND METHODS

The materials and methods have been described in detail (29, 30) except for minor alterations. Briefly, the procedures were as follows.

Brittle wax beans, *Phaseolus vulgaris*, were grown in the greenhouse in a dilute Johnson's solution (11, 30) for approximately 3 weeks, at which time stem tissue consisting of both the epicotyl and hypocotyl was harvested. The stem was sliced transversely at 400-μ intervals (see Refs. 29, 30, 35 for details). Approximately 40 slices made up a sample weighing 80 to 110 mg.

Samples were placed in aging solutions of various compositions for predetermined periods. The solutions were maintained at 30 C unless otherwise specified. Subsequently ion absorption was studied by placing stem slices in solutions at 30 C containing specified concentrations of K or Na as the Cl salt. Calcium (as $CaSO_4$), when added, was present at a concentration of 0.5 mM in the absorption or aging solutions. Sodium was labeled with ^{22}Na, and K with ^{86}Rb. The validity of the use of ^{86}Rb to trace K has been established (28, 30).

Absorption periods were terminated by exposing samples for 15 min to a cold (4 C) solution containing 1 mM NaCl or KCl and 0.5 mM $CaSO_4$. This desorption procedure was carried out to remove diffusible and superficially adsorbed ions (7). At the end of desorption the samples were rinsed in water, blotted, weighed, and ashed in nickel-plated planchets. The ash was wetted, spread with detergent, and dried under infrared heat lamps. Radioactivity was determined with a gas flow counter; amounts of Na and K are expressed as nanomoles per gram fresh weight.

Chemical determinations were made with an atomic absorption spectrophotometer and Na and K content are expressed as micromoles per gram fresh weight.

Respiration was determined in a closed system in which the

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disappearance of O_2 from a solution containing the tissue was measured with a Beckman O_2 electrode and a Beckman model 777 O_2 analyzer connected to an Autograph recorder. Respiration is expressed as nanomoles of O_2 consumed per gram fresh weight per minute.

Bacteria have been shown to influence certain aspects of the aging of plant tissue (4, 16); however, this influence appears to be negligible in bean stem slices (30). Nevertheless, 50 μg ml^{-1} chloramphenicol, an effective bacteriostat (16, 40), was used routinely as a precautionary measure to maintain a low bacterial count in the aging solution.

RESULTS

The effect of various divalent cations on K absorption by fresh and aged stem slices was investigated. The results are presented in Figure 1. The concentration of divalent cations was 0.5 mM in both the aging and absorption solutions with the exception of the control, in which no divalent cations were added. The absorption solutions contained 0.1 mM K. In the absence of divalent cations in the aging and absorption solutions, freshly sliced tissue absorbed K considerably faster than did aged tissue. When divalent cations were present in the absorption and aging solutions, the results were reversed; K absorption was much greater by aged tissue than by fresh tissue in every treatment except Ba. It should be noted that of all the divalent cations studied Ca added to the aging and absorption solutions was the most effective in promoting K absorption by aged tissue.

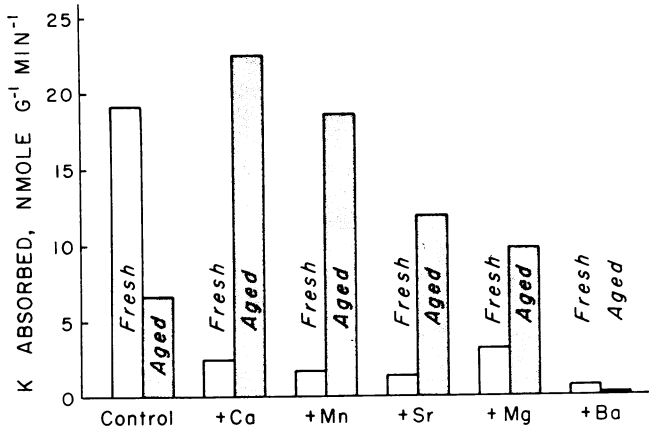

FIG. 1. Absorption of K by fresh and aged bean stem slices in the presence or absence of divalent cations. Potassium was present at a concentration of 0.1 mM, and divalent cations, when added, at 0.5 mM. Both absorption and aging solutions contained the divalent cations indicated in the figure except the control solutions, in which no divalent cations were added to either solution. Fresh tissue was placed in absorption solutions within 10 min of slicing, and aged tissues were washed in distilled water or 0.5 mM divalent cation solutions for 18 hr before absorption. Absorption period, 30 min; temperature, 30 C.

The data in Figure 1 suggest two questions. First, why is the absorption of K by freshly sliced tissue much greater in the absence of Ca than in its presence? Second, in what way does Ca influence aging so that subsequent K absorption is greater?

Results in Figure 2 show the effect on Na absorption of aging in either 0.5 mM Ca or distilled water. Absorption solutions consisted of 0.1 mM Na and 0.5 mM Ca. As previously shown (30), the absorption of Na decreased as stem slices were washed. The presence or absence of Ca in the aging solutions had little effect on subsequent Na absorption.

When K absorption was studied in a similar experiment, the

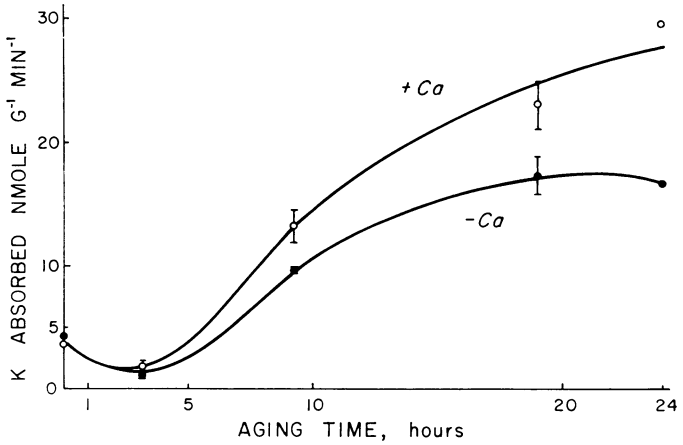

FIG. 3. Absorption of K as a function of aging time in the presence and absence of Ca. Potassium concentration, 0.1 mM; Ca, 0.5 mM in absorption solutions. All other conditions and conventions as indicated for Figure 2.

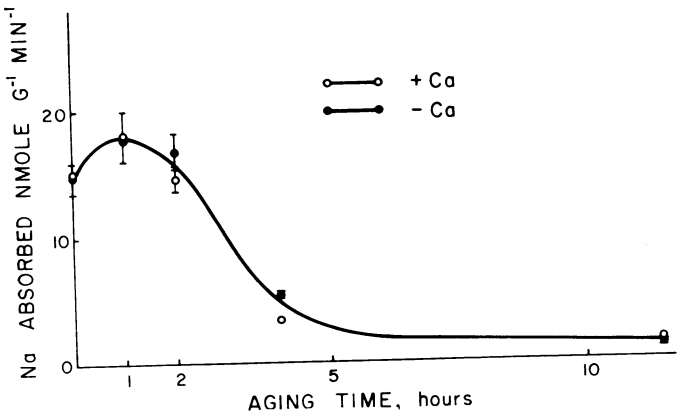

FIG. 2. Absorption of Na as a function of aging time in the presence and absence of Ca in aging solutions. Sodium was present at a concentration of 0.1 mM and Ca at 0.5 mM in all absorption solutions. Aging solutions were either distilled water or 0.5 mM $CaSO_4$. Absorption period, 30 min; temperature, 30 C. Where horizontal lines are drawn, the point represents a mean of two values indicated by the horizontal lines. If horizontal lines not drawn, difference not greater than symbol.

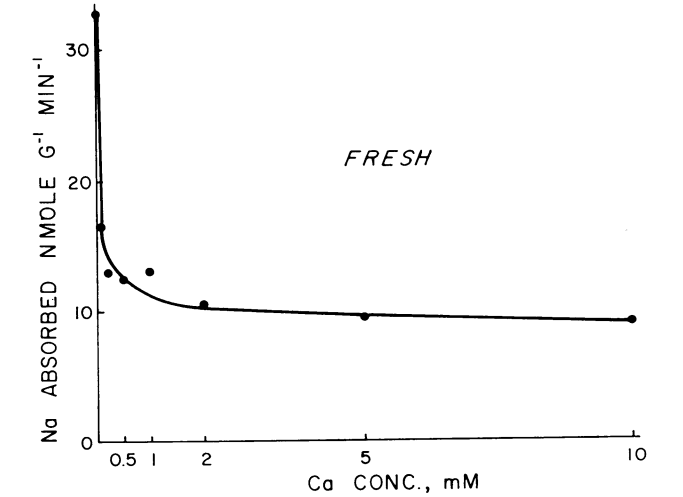

FIG. 4. Absorption of Na by fresh tissue as a function of increasing concentrations of Ca. Sodium was present at a concentration of 0.1 mM, and Ca varied from 0 to 10 mM. Absorption period, 30 min; temperature, 30 C. No replicates.

results were somewhat different (Fig. 3). The aging solutions were either water or 0.5 mM Ca and the absorption solutions were 0.1 mM K and 0.5 mM Ca. The development of K-absorbing capacity by aging tissue was less when no Ca was added during aging.

The results in Figure 4 demonstrate the effect that increasing concentrations of Ca have on the absorption of Na by freshly sliced stem tissue. Sodium was present at a concentration of 0.1 mM, and Ca was increased from 0 to 10 mM. Small concentrations of Ca (0.1 mM) decreased Na uptake 50%. Increasing the concentration of Ca up to 10 mM had little further effect.

A similar experiment on K absorption is shown in Figure 5. Calcium depressed the absorption of K by fresh tissue more severely than the absorption of Na (*cf.* Fig. 4). Absorption of K decreased to a value approaching 1 nmole g^{-1} min^{-1} at the highest Ca concentrations, while Na absorption was decreased to approximately 8 nmoles g^{-1} min^{-1}.

Results were quite different when tissues were aged and absorption of Na and K was compared as a function of increasing Ca concentrations.

Figure 6 shows the results from an experiment on the absorption of Na by aged stem slices. Stem slices were aged for 18 hr in the presence or absence of 0.5 mM Ca and rinsed with water;

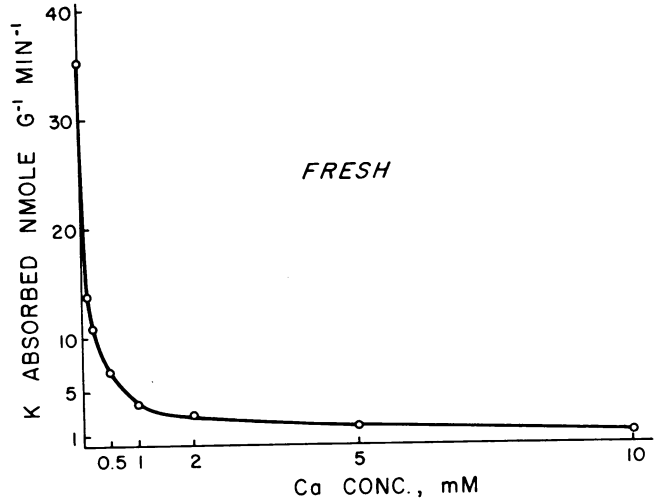

FIG. 5. Absorption of K by fresh bean stem slices as a function of increasing Ca concentration. Potassium present at a concentration of 0.1 mM, and all other conventions and conditions as in Figure 4.

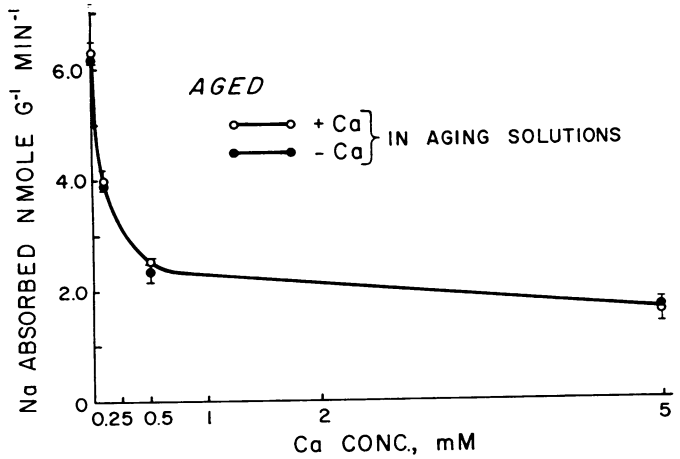

FIG. 6. Absorption of Na by aged tissue as a function of Ca concentration. Tissue aged for 18 hr in presence or absence of 0.5 mM Ca previous to absorption period. Absorption period, 30 min; temperature, 30 C. All other conditions and conventions as in Figure 2.

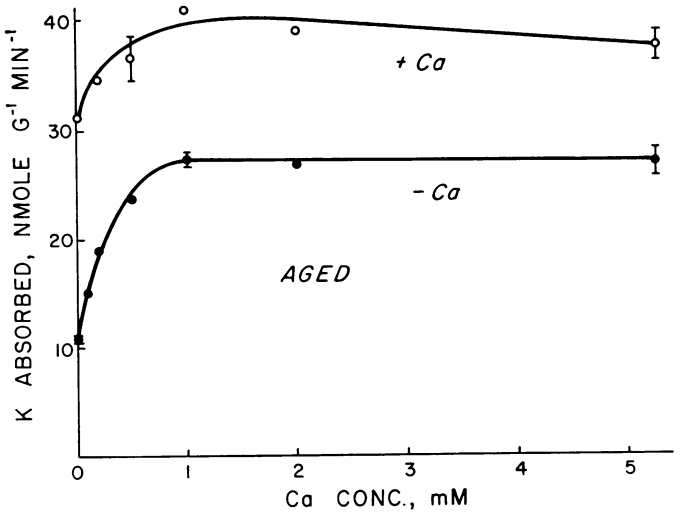

FIG. 7. Absorption of K (0.1 mM) by aged tissue as a function of Ca concentration. All other conditions and conventions as indicated in Figure 6.

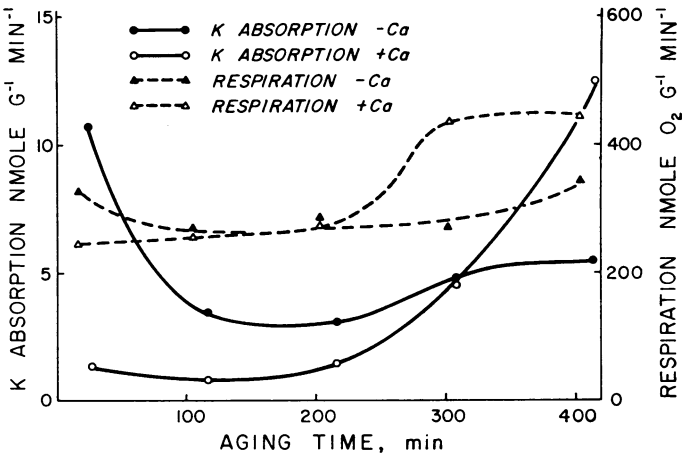

FIG. 8. Absorption of K and respiratory rate of tissue as a function of aging time in the presence and absence of Ca in aging solutions. Measurements of absorption and respiration made on the same sample. Potassium concentration was 0.1 mM, and Ca, when added, was 0.5 mM. Respiratory measurements were made over a 15-min period, and the absorption period was 10 min. No replicates.

then the absorption of Na (0.1 mM) was studied as a function of increasing concentrations of Ca. As shown earlier with fresh tissue (*cf.* Fig. 4), increasing concentrations of Ca decreased Na absorption. It should also be noted that the presence or absence of Ca in the aging solutions had no effect on subsequent Na absorption by aged tissue.

A similar experiment studied the effect of Ca on K absorption by tissue aged for 18 hr in the presence and absence of Ca (Fig. 7). Increasing the level of Ca in the absorption media resulted in a greater percentage enhancement of K absorption by tissue aged in water than in Ca solutions. However, in comparison with tissue aged in water, the absorption of K by tissue aged in Ca solutions was higher at all levels of Ca added in the absorption solutions.

Figure 8 shows results of an experiment on the effect of Ca on respiration and K absorption by aging stem slices. These are typical results of several experiments. Measurements of respiration and of K absorption were done on the same sample. Potassium was present at a concentration of 0.1 mM, and Ca was 0.5

mM when added. When tissue was aged in a Ca solution, an increase in K absorption was observed with a certain lag period, and this paralled the observed increase in respiration of the tissue. When Ca was omitted from the aging and absorption solutions, the increase in respiration rate was slight, as was the increase in K absorption. An interesting aspect of the respiratory studies was the observation that, although the increase in respiration paralleled the rise in K absorption capacity, this enhanced absorption capacity continued to develop long after the higher level of respiration had been established. In fact absorption continued to increase while respiration was declining, slightly, with time.

The development of ion absorption capacity along with increased respiration has been suggested to involve basic alterations in physiological processes of plant tissue (1, 9, 14, 20, 40) and especially in protein metabolism (4, 12, 20, 27).

Figure 9A shows results of an experiment on the effect of cycloheximide, a known inhibitor of protein synthesis (4), on the development of K absorption capacity. Potassium was present at 0.1 mM, and Ca was 0.5 mM. Cycloheximide, 10 μM, was present during aging but not during absorption. The capacity to absorb K does not develop to any great extent when cycloheximide is added during aging. The results in Figure 9B show the effect of cycloheximide on the development of respiration as a function of time. Respiration is not enhanced when 10 μM cycloheximide is in the aging solutions; this corresponds to the lack of development of K absorption capacity (Fig. 9A).

The changes in K and Na content of stem slices as a function of time were studied in solutions containing 0.1 mM K and 0.5 mM Ca or 0.1 mM Na and 0.5 mM Ca. Tissue was sliced, washed in water for 15 sec, and then placed in the respective solutions.

The K content decreased within 30 min after the tissue was immersed in K solution (Fig. 10A). The K content then remained fairly constant for several hours. After 8 hr an increase in K content was detectable and by 20 hr the K content had risen above the initial level.

Conversely, the Na content increased immediately upon immersion of the tissue in Na solution (Fig. 10B). After 3 hr it leveled off. Although not shown in this figure, Na content remains virtually constant at the level obtained after 3 hr for up to 24 hr in Na solutions.

One point should be made about the effective Ca concentration required for development of the aging phenomenon. A series of experiments showed that 1 to 2 mM Ca in the aging solutions resulted in a maximal aging response. In other words, the absorption of K by tissues aged at these concentrations of Ca was as much as 20% higher than that by tissue aged in solution containing 0.5 mM Ca. Qualitatively, however, the relationships were the same.

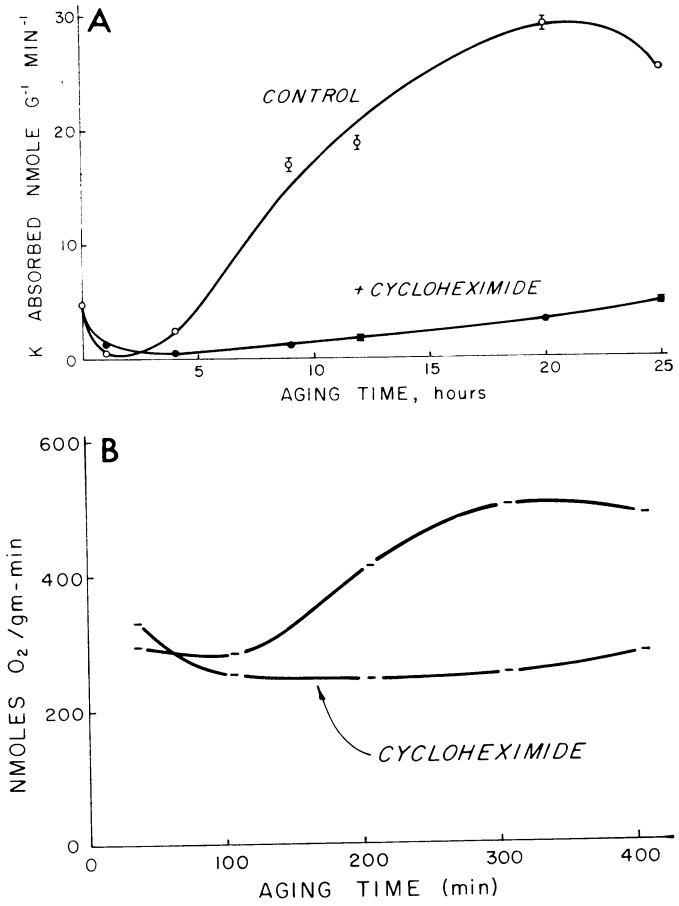

FIG. 9. A: Absorption of K as a function of aging time in the presence and absence of cycloheximide. Aging solutions contained ± 10 μM cycloheximide, and 0.5 mM $CaSO_4$. Absorption solutions contained 0.1 mM K and 0.5 mM Ca. Absorption period, 30 min; temperature, 30 C. Other conditions and conventions as in Figure 2. B: Respiration as a function of aging time in presence and absence of cycloheximide. Other conditions and conventions as in part A.

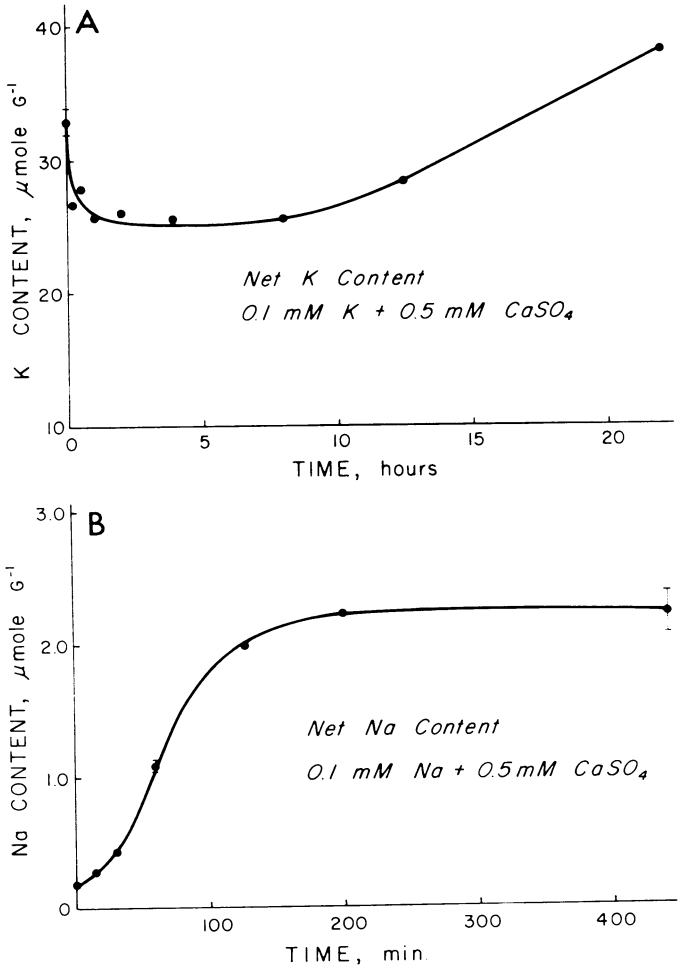

FIG. 10. A: Potassium content of tissue as a function of time in solution containing 0.1 mM K and 0.5 mM Ca. B: Sodium content of tissue as a function of time in solution containing 0.1 mM Na and 0.5 mM Ca.

DISCUSSION

This paper is concerned with some aspects of aging bean stem slices. The data suggest that the solutions in which aging treatments are carried out must be known and controlled. Variability in their composition may lead to quantitative and qualitative differences in the characteristics of an aged tissue.

Evidence presented in Figure 10A suggests that slicing stem tissue results in an initial efflux of K, possibly resulting from partial damage or complete destruction of cells close to the cut surface. The rinsing procedure prior to analyses or absorption studies may not entirely remove this labile fraction, particularly from damaged cells. The higher initial rates of K absorption measured by radioisotopes (Figs. 1, 8) may represent isotopic exchange. Even though a desorption procedure is carried out at the end of the absorption period (see "Materials and Methods"), ions may leak slowly enough from partially damaged cells so that this constitutes a measurable fraction in the freshly sliced tissue. When the tissue is washed during aging, this exchangeable fraction is reduced. Calcium may accelerate this process (Figs. 1, 8). The loss of K could "trigger" the subsequent development of K absorption capacity. Although the kinetics of the respective processes do not indicate this (30 min for initial loss of K and 4 hr or more for development of K absorption capacity), the possibility should not be ruled out. Of major interest, however, is the alteration of physiological processes paralleling the development of K absorption capacity (Figs. 8, 9).

The immediate increase in Na content of stem tissue (Fig. 10B) is the result of substantial rates of Na absorption by fresh tissue (Figs. 2, 4). As bean stem tissue ages, the rate of Na absorption declines (Fig. 2, Ref. 30), as does the rate of increase in Na content. When the rate of Na absorption becomes 0, an increase in Na content is no longer detectable. It is unlikely that the increase in Na content is an exchange with K. The absorption of Na by fresh tissue is fairly specific (29), it is inhibited by antimetabolites (30), and kinetically it does not mirror the loss of K.

The importance of Ca in the transport of ions by plant tissue is well known (5, 6, 10, 18, 23, 31, 32), and it has been shown to promote selective ion transport (6, 10, 18, 23, 31, 32, 41). A similar observation was made in this study. Preferential absorption of Na by freshly sliced tissue appeared to be promoted by the presence of Ca. Potassium and Na absorption are both reduced (*cf.* Figs. 4 and 5), with K uptake reduced to a value of approximately one-eighth of the Na uptake at the highest Ca level. The net result is what appears to be a preferential Na-absorbing mechanism.

The effect of Ca on Na uptake in aged stem tissue is qualitatively similar to that found in fresh tissue. The reduction of Na absorption in the presence of Ca is in addition to an already substantially reduced capacity for Na uptake as the result of aging (*cf.* Figs. 2, 4, and 6). Hence, Na absorption by aged tissue is almost 0 in the presence of Ca.

The effect of Ca on K absorption differs in aged and fresh tissues (*cf.* Figs. 5 and 7). Increasing concentrations of Ca accelerated K absorption by aged tissue but depressed it in fresh tissue. The net result, then, on K and Na absorption by aged tissue in the presence of Ca is a higher rate of K absorption and a reduced rate of Na absorption, with the tissue preferentially absorbing K.

The positive response of K absorption to Ca, as well as the requirement of Ca for maximal development of K absorption, indicates that K uptake by aged tissue is mediated by a mechanism different from the one responsible for Na uptake, and it is proposed that a new mechanism for K transport develops with aging. These data are in accord with previous speculations (30).

Alteration of respiration in response to aging was demonstrated in this study (*cf.* Fig. 8). The increase of respiration as bean stem slices are aged parallels the observed increase in K absorption. It is interesting to note the necessity for Ca as well as protein synthesis for the increased respiration and K uptake. Calcium has been shown previously to influence respiration in maize roots (8). Evidence that major physiological changes attend or control the increase in K uptake during aging is presented in Figure 9. It can be seen that cycloheximide eliminates most of the increase in K uptake and respiration induced by aging. These results taken in conjunction with the observation that the absence of Ca in the aging solution also reduced development of K absorption capacity and respiration suggest that the presence of Ca has a modifying effect on the aging process.

One such alteration in aging tissue is the turnover of membrane constituents, as shown in potato discs (40). Calcium could conceivably be important in this process. A close connection has been demonstrated (22, 24, 25) between Ca and membrane integrity. Marinos (22) found that membranes broke down when barley roots were deficient in Ca. The requirement for Ca appeared to be fairly specific for the maintenance of membrane structure. Alterations of membrane fractions as tissue aged were demonstrated by Jackman and van Steveninck (9). The endoplasmic reticulum of beet root slices went through a complete cycle of breakdown and resynthesis during an aging period, undoubtedly involving a substantial turnover of membrane components (9). The Ca requirement for maximal development of K-absorbing capacity during aging (*cf.* Figs. 1, 3, and 7), and the involvement of protein synthesis, along with findings already discussed (9, 22, 40), lead one to speculate that Ca may influence membrane metabolism in aging stem slices.

LITERATURE CITED

1. APREES, T. 1966. Evidence for the widespread occurrence of induced respiration in slices of plant tissue. Aust. J. Biol. Sci. 19: 981–990.
2. BIELESKI, R. L. 1966. Accumulation of phosphate, sulfate and sucrose by excised phloem tissue. Plant Physiol. 41: 447–454.
3. CLICK, R. E. AND D. P. HACKETT. 1963. The role of protein and nucleic acid synthesis in the development of respiration in potato slices. Proc. Nat. Acad. Sci. U.S.A. 50: 243–250.
4. ELLIS, R. J. AND I. R. MACDONALD. 1967. Activation of protein synthesis by microsomes from aging beet disks. Plant Physiol. 42: 1297–1302.
5. ELZAM, O. E. AND T. K. HODGES. 1967. Calcium inhibition of potassium absorption by corn roots. Plant Physiol. 42: 1483–1488.
6. EPSTEIN, E. 1961. The essential role of calcium in selective cation transport by plant cells. Plant Physiol. 36: 437–444.
7. EPSTEIN, E., W. E. SCHMID, AND D. W. RAINS. 1963. Significance and technique of short-term experiments on solute absorption by plant tissue. Plant Cell Physiol. 4: 79–84.
8. HANDLEY, R. AND R. OVERSTREET. 1962. Sodium chloride, calcium chloride, and the respiration of maize root sections. Science 135: 731–732.
9. JACKMAN, M. E. AND R. F. M. VAN STEVENINCK. 1967. Changes in the endoplasmic reticulum of beetroot slices during aging. Aust. J. Biol. Sci. 20: 1063–1068.
10. JACOBSON, L., R. HANNAPEL, D. P. MOORE, AND M. SCHAEDLE. 1961. Influence of calcium on selectivity of ion absorption process. Plant Physiol. 36: 58–61.
11. JOHNSON, C. M., P. R. STOUT, T. C. BROYER, AND A. B. CARLTON. 1957. Comparative chlorine requirements of different plant species. Plant Soil 8: 337–353.
12. KAHL, G., H. LANGE, AND G. ROSENSTOCK. 1969. Substratspiegel, Enzymaktivitäten und genetische Regulation nach Derepression in pflanzlichen Speichergeweben. Z. Naturforsch. 243: 911–918.
13. LAHAYE, P. A. AND E. EPSTEIN. 1969. Salt toleration by plants: enhancement by calcium. Science 166: 395–396.
14. LATIES, G. G. 1957. Respiration and cellular work and the regulation of the respiration rate in plants. *In:* B. Glass, ed., Survey of Biological Progress, Vol. 3. Academic Press, New York. pp. 216–299.
15. LATIES, G. G. 1967. Metabolic and physiological development in plant tissues. Aust. J. Sci. 30: 193–203.
16. LEAVER, C. J. AND J. EDELMAN. 1965. Antibiotics as a means of control of bacterial contamination of storage tissue discs. Nature 207: 1000–1001.
17. LOUGHMAN, B. C. 1960. Uptake and utilization of phosphate associated with respiratory changes in potato tuber slices. Plant Physiol. 35: 418–424.
18. MAAS, E. V., D. P. MOORE, AND B. J. MASON. 1969. Influence of calcium and magnesium on manganese absorption. Plant Physiol. 44: 796–800.
19. MACDONALD, I. R. 1967. Oxygen tension a determining factor in the respiration of potato disks of varying thickness. Plant Physiol. 42: 227–232.

20. MacDonald, I. R., J. S. D. Bacon,. D. Vaughan, and R. J. Ellis. 1966. The relation between ion absorption and protein synthesis in beet disks. J. Exp. Bot. 17: 822–837.
21. MacDonald, I. R., P. C. DeKock, and A. H. Knight. 1960. Variations in the mineral content of storage tissue disks maintained in tap water. Physiol. Plant. 13: 76–89.
22. Marinos, N. G. 1962. Studies on submicroscopic aspects of mineral deficiencies. I. Calcium deficiency in the shoot apex of barley. Amer. J. Bot. 49: 834–841.
23. Marschner, H. 1964. Einfluss von Calcium auf die Natriumaufnahme und die Kalciumabgabe isolierter Gerstenwurzeln. Z. Pflanzenernaehr. Dueng. Bodenk. 107: 19–32.
24. Marschner, H. and I. Günther. 1964. Ionenaufnahme und Zellstruktur bei Gerstenwurzeln in Abhängigkeit von der Calcium-Versorgung. Z. Pflanzenernaehr. Dueng. Bodenk. 107: 118–136.
25. Marschner, H. and K. Mengel. 1966. Der Einfluss von Ca- und H-ionen bei unterschiedlichen Stoffwechselbedingungen auf die Membranpermeabilatät junger Gerstenwurzeln. Z. Pflanzenernaehr. Dueng. Bodenk. 112: 39–49.
26. Palmer, J. M. and B. C. Loughman. 1964. Some factors affecting the rate of absorption and utilization of ions by stem segments. New Phytol. 63: 217–231.
27. Polya, G. M. 1968. Inhibition of protein synthesis and uptake in beetroot tissue by cycloheximide and cryptopleurine. Aust. J. Biol. Sci. 21: 1107–1118.
28. Rains, D. W. 1968. Kinetics and energetics of light-enhanced potassium absorption by corn leaf tissue. Plant Physiol. 43: 394–400.
29. Rains, D. W. 1969. Cation absorption by slices of stem tissue of bean and cotton. Experientia 25: 215–216.
30. Rains, D. W. 1969. Sodium and potassium absorption by bean stem tissue. Plant Physiol. 44: 547–554.
31. Rains, D. W. and E. Epstein. 1967. Sodium absorption by barley roots: role of the dual mechanisms of alkali cation transport. Plant Physiol. 42: 314–318.
32. Rains, D. W., W. E. Schmid, and E. Epstein. 1964. Absorption of cations by roots. Effects of hydrogen ions and essential role of calcium. Plant Physiol. 39: 274–278.
33. Robertson, R. N. 1960. Ion absorption and respiration. Biol. Rev. 35: 231–264.
34. Shaw, M. and M. S. Manocha. 1965. Fine structure in detached senescing wheat leaves. Can. J. Bot. 43: 747–755.
35. Smith, R. C. and E. Epstein. 1964. Ion absorption by shoot tissue: technique and first findings with excised leaf tissue of corn. Plant Physiol. 39: 338–341.
36. van Steveninck, R. F. M. 1962. Potassium fluxes in red beet tissue during its "lag phase". Physiol. Plant. 15: 211–215.
37. van Steveninck, R. F. M. 1961. The lag-phase in salt uptake of storage tissue. Nature 190: 1072–1075.
38. Steward, F. C., R. Wright, and W. E. Berry. 1932. The respiration of cut disks of potato tuber in air and immersed in water, with observations upon surface: volume effects and salt accumulation. Protoplasma 16: 576–611.
39. Sugiura, M. 1963. Inhibitory effect of kinetin on respiration of tobacco leaf disks. Bot. Mag. 76: 359–362.
40. Tang, W. J. and P. A. Castelfranco. 1968. Phospholipid synthesis in aging potato tuber tissue. Plant Physiol. 43: 1232–1238.
41. Viets, F. G. 1944. Calcium and other polyvalent cations as accelerators of ion accumulation by excised barley roots. Plant Physiol. 19: 466–480.