5 Calcium

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5.1 HISTORICAL INFORMATION

5.1.1 Determination of Essentiality

The rare earth element calcium is one of the most abundant elements in the lithosphere; it is readily available in most soils; and it is a macronutrient for plants, yet it is actively excluded from plant cytoplasm.

In 1804, de Saussure showed that a component of plant tissues comes from the soil, not the air, but it was considerably later that the main plant nutrients were identified. Liebig was the first person to be associated strongly with the idea that there are essential elements taken up from the soil (in 1840), although Sprengel was the first person to identify calcium as a macronutrient in 1828 (1). Calcium was one of the 20 essential elements that Sprengel identified.

Salm-Horstmar grew oats (*Avena sativa* L.) in inert media with different elements supplied as solutions in 1849 and 1851 and showed that omitting calcium had an adverse effect on growth (2). However, it was the discovery that plants could be grown in hydroponic culture by Sachs (and almost simultaneously Knop) in 1860 that made investigation of what elements are essential for plant growth much easier (2). Sachs' first usable nutrient solution contained $CaSO_4$ and $CaHPO_4$.

It has been well known since the early part of the twentieth century that there is a very distinct flora in areas of calcareous soils, comprised of so-called calcicole species. There are equally distinctive groups of plant species that are not found on calcareous soils, the calcifuge species (see Section 5.3.2.3).

5.2 FUNCTIONS IN PLANTS

Calcium has several distinct functions within higher plants. Bangerth (3) suggested that these functions can be divided into four main areas: (a) effects on membranes, (b) effects on enzymes, (c) effects on cell walls, and (d) interactions of calcium with phytohormones, although the effects on enzymes and the interactions with phytohormones may be the same activity. As a divalent ion, calcium is not only able to form intramolecular complexes, but it is also able to link molecules in intermolecular complexes (4), which seems to be crucial to its function.

5.2.1 Effects on Membranes

Epstein established that membranes become leaky when plants are grown in the absence of calcium (5) and that ion selectivity is lost. Calcium ions (Ca^{2+}) bridge phosphate and carboxylate groups of phospholipids and proteins at membrane surfaces (6), helping to maintain membrane structure. Also, some effect occurs in the middle of the membrane, possibly through interaction of the calcium and proteins that are an integral part of membranes (6,7). Possibly, calcium may link adjacent phosphatidyl-serine head groups, binding the phospholipids together in certain areas that are then more rigid than the surrounding areas (8).

5.2.2 Role in Cell Walls

Calcium is a key element in the structure of primary cell walls. In the primary cell wall, cellulose microfibrils are linked together by cross-linking glycans, usually xyloglucan (XG) polymers but also glucoarabinoxylans in Poaceae (Gramineae) and other monocots (9). These interlocked microfibrils are embedded in a matrix, in which pectin is the most abundant class of macromolecule. Pectin is also abundant in the middle lamellae between cells.

Pectin consists of rhamnogalacturonan (RG) and homogalacturonan (HG) domains. The HG domains are a linear polymer of (1→4)-α′-linked D-galacturonic acid, 100 to 200 residues long, and are deposited in the cell wall with 70 to 80% of the galacturonic acid residues methyl-esterified at the C6 position (9). The methyl-ester groups are removed by pectin methylesterases, allowing calcium ions to bind to the negative charges thus exposed and to form inter-polymer bridges that hold the backbones together (9). The whole structure can be thought of as resembling an eggbox (Figure 5.1).

Pectin is a highly hydrated gel containing pores; the smaller the size of these pores, the higher the Ca^{2+} concentration in the matrix and more cross-linking of chains occurs (11). This gel holds the XG molecules in position relative to each other, and these molecules in turn hold the cellulose microfibrils together (Figure 5.2). The presence of the calcium, therefore, gives

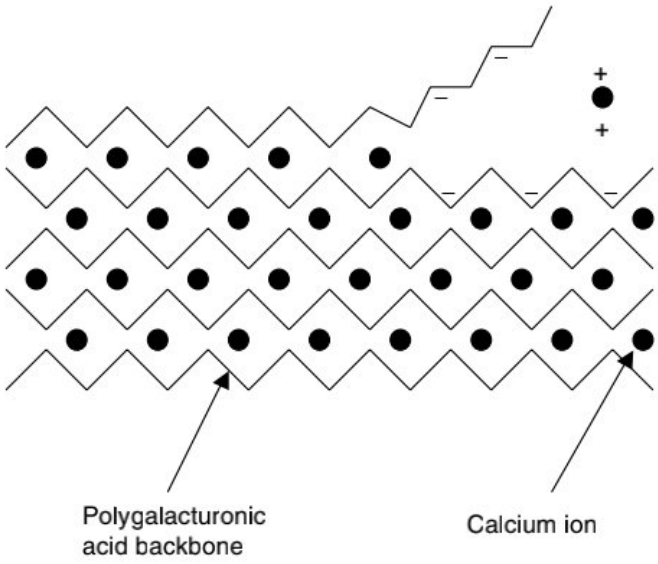

FIGURE 5.1 The 'eggbox' model of calcium distribution in pectin. (Based on E.R. Morris et al., *J. Mol. Biol.* 155: 507–516, 1982.)

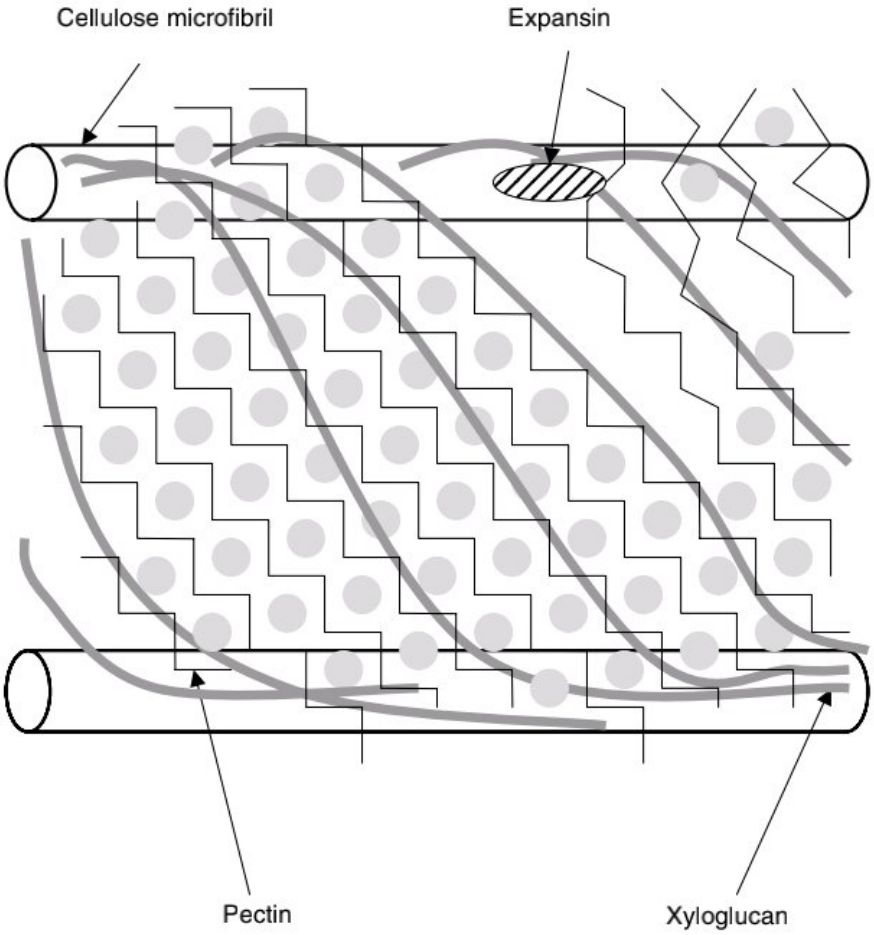

FIGURE 5.2 Diagrammatic representation of the primary cell wall of dicotyledonous plants. (Based on E.R. Morris et al., *J. Mol. Biol.* 155:507–516, 1982; F.P.C. Blamey, *Soil Sci. Plant Nutr.* 49:775–783, 2003; N.C. Carpita and D.M. Gibeaut, *Plant J.* 3:1–30, 1993.) To the right of the figure, Ca^{2+} ions have been displaced from the HG domains by H^+ ions, so that the pectin is no longer such an adhesive gel and slippage of the bonds between adjacent XG chains occurs and expansin is able to work on them. This loosens the structure and allows the cellulose microfibrils to be pushed further apart by cell turgor.

some load-bearing strength to the cell wall (13). It is suggested that when a primary cell wall is expanding, localized accumulation of H^+ ions may displace Ca^{2+} from the HG domains, thereby lowering the extent to which the pectin holds the XG strands together (11). In a root-tip cell, where the cellulose microfibrils are oriented transversely, slippage of the XG chains allows the cellulose microfibrils to move further apart from each other, giving cell expansion in a longitudinal direction.

Cell-to-cell adhesion may also be given by Ca^{2+} cross-linking between HG domains in the cell walls of adjacent cells, but this action is less certain as experimental removal of Ca^{2+} leads to cell separation in a only few cases (9). In the ripening of fruits, a loosening of the cells could possibly occur with loss of calcium. It has been postulated that decrease in apoplastic pH in ripening pome fruits may cause the release of Ca^{2+} ions from the pectin, allowing for its solubilization (14). However, in an experiment on tomato (*Lycopersicon esculentum* Mill.), the decline in apoplastic pH that occurred was not matched by a noticeable decrease in apoplastic Ca^{2+} concentration, and the concentration of the ion remained high enough to limit the solubilization of the pectin (15). It certainly seems that calcium inhibits the degradation of the pectates in the cell wall by inhibiting the formation of polygalacturonases (16), so the element has roles in possibly holding the pectic components together and in inhibiting the enzymes of their degradation. In a study on a ripening and a nonripening cultivar of tomato (Rutgers and *rin*, respectively), there was an increase in calcium concentration after anthesis in the *rin* cultivar, whereas in the Rutgers cultivar there was a noticeable fall in the concentration of bound calcium and an increase in polygalacturonase activity (17). In a study on calcium deficiency in potato (*Solanum tuberosum* L.), deficient plants had more than double the activity of polygalacturonase compared with normal plants (18).

5.2.3 Effects on Enzymes

Unlike K^+ and Mg^{2+}, Ca^{2+} does not activate many enzymes (19), and its concentration in the cytoplasm is kept low. This calcium homeostasis is achieved by the action of membrane-bound, calcium-dependent ATPases that actively pump Ca^{2+} ions from the cytoplasm and into the vacuoles, the endoplasmic reticulum (ER), and the mitochondria (20). This process prevents the ion from competing with Mg^{2+}, thereby lowering activity of some enzymes; the action prevents Ca^{2+} from inhibiting cytoplasmic or chloroplastic enzymes such as phosphoenol pyruvate (PEP) carboxylase (21) and prevents Ca^{2+} from precipitating inorganic phosphate (22).

Calcium can be released from storage, particularly in the vacuole, into the cytoplasm. Such flux is fast (23) as it occurs by means of channels from millimolar concentrations in the vacuole to nanomolar concentrations in the cytoplasm of resting cells (24). The calcium could inhibit cytoplasmic enzymes directly, or by competition with Mg^{2+}. Calcium can also react with the calcium-binding protein calmodulin (CaM). Up to four Ca^{2+} ions may reversibly bind to each molecule of calmodulin, and this binding exposes two hydrophobic areas on the protein that enables it to bind to hydrophobic regions on a large number of key enzymes and to activate them (25). The Ca^{2+}–calmodulin complex also may stimulate the activity of the calcium-dependent ATPases (26), thus removing the calcium from the cytoplasm again and priming the whole system for further stimulation if calcium concentrations in the cytoplasm rise again.

Other sensors of calcium concentration are in the cytoplasm, for example, Ca^{2+}-dependent (CaM-independent) protein kinases (25). The rapid increases in cytoplasmic Ca^{2+} concentration that occur when the channels open and let calcium out of the vacuolar store and the magnitude, duration, and precise location of these increases give a series of calcium signatures that are part of the responses of a plant to a range of environmental signals. These responses enable the plant to respond to drought, salinity, cold shock, mechanical stress, ozone and blue light, ultraviolet radiation, and other stresses (24).

5.2.4 Interactions with Phytohormones

An involvement of calcium in the actions of phytohormones seems likely as root growth ceases within only a few hours of the removal of calcium from a nutrient solution (22). The element appears to be involved in cell division and in cell elongation (27) and is linked to the action of auxins. The loosening of cellulose microfibrils in the cell wall is controlled by auxins, giving rise to excretion of protons into the cell wall. Calcium is involved in this process, as discussed earlier. Furthermore, auxin is involved in calcium transport in plants, and treatment of plants with the indoleacetic acid (IAA) transport inhibitor, 2,3,5-triiodobenzoic acid (TIBA), results in restricted calcium transport into the treated tissue (28). As the relationship is a two-way process, it cannot be confirmed easily if calcium is required for the action of IAA or if the action of IAA gives rise to cell growth, and consequent cell wall development, with the extra pectic material in the cell wall then acting as a sink for calcium. It is also possible that IAA influences the development of xylem in the treated tissue (29).

Increase in shoot concentrations of abscisic acid (ABA) following imposition of water-deficit stress leads to increased cytoplasmic concentration of Ca^{2+} in guard cells, an increase that precedes stomatal closure (24). Further evidence for an involvement of calcium with phytohormones has come from the observation that senescence in maize (*Zea mays* L.) leaves can be slowed by supplying either Ca^{2+} or cytokinin, with the effects being additive (30). There is also a relationship between membrane permeability, which is strongly affected by calcium content and ethylene biosynthesis in fruit ripening (31).

5.2.5 Other Effects

It has been known for a long time that calcium is essential for the growth of pollen tubes. A gradient of cytoplasmic calcium concentration occurs along the pollen tube, with the highest concentrations being found in the tip. The fastest rate of influx of calcium occurs at the tip, up to 20 pmol cm^{-2} s^{-1}, but there are oscillations in the rate of pollen tube growth and calcium influx that are approximately in step (32). It seems probable that the calcium exerts an influence on the growth of the pollen tube mediated by calmodulin and calmodulin-like domain protein kinases (25), but the growth and the influx of calcium are not directly linked as the peaks in oscillation of growth precede the peaks in uptake of calcium by 4 s (32). Root hairs have a high concentration of Ca^{2+}, and root hair growth has a similar calcium signature to pollen tube growth (24). Slight increases in cytoplasmic Ca^{2+} concentration can close the plasmodesmata in seconds, with the calcium itself and calmodulin being implicated (33). Many sinks, such as root apices, require symplastic phloem unloading through sink plasmodesmata, so this action implies that calcium has a role as a messenger in the growth of many organs.

It seems that calcium can be replaced by strontium in maize to a certain extent (34), but despite the similarities in the properties of the two elements, this substitution does not appear to be common to many plant species. In general, the presence of abundant calcium in the soil prevents much uptake of strontium, and in a study on 10 pasture species, the concentration of strontium in the shoot was correlated negatively with the concentration of calcium in the soil (35).

5.3 DIAGNOSIS OF CALCIUM STATUS IN PLANTS

5.3.1 Symptoms of Deficiency and Excess

Plants deficient in calcium typically have upper parts of the shoot that are yellow-green and lower parts that are dark green (36) (Figure 5.3). Given the abundance of calcium in soil, such a condition is unusual, although it can arise from incorrect formulation of fertilizers or nutrient solutions.

FIGURE 5.3 Calcium-deficient maize (*Zea mays* L.). The younger leaves which are still furled are yellow, but the lamina of the older, emerged leaf behind is green. (Photograph by Allen V. Barker.) (For a color presentation of this figure, see the accompanying compact disc.)

However, despite the abundance of calcium, plants suffer from a range of calcium-deficiency disorders that affect tissues or organs that are naturally low in calcium. These include blossom-end rot (BER) of tomato (Figure 5.4 and Figure 5.5), pepper (*Capsicum annuum* L.), and water melon (*Cucumis melo* L.) fruits, bitter pit of apple (*Malus pumila* Mill.), black heart of celery (*Apium graveolens* L.), internal rust spot in potato tubers and carrot (*Daucus carota* L.) roots, internal browning of Brussels sprouts (*Brassica oleracea* L.), internal browning of pineapple (*Ananas comosus* Merr.), and tip burn of lettuce (*Lactuca sativa* L.) and strawberries (*Fragaria x ananassa* Duch.) (22,37,38). Recently, it has been suggested that the disorder 'crease' in navel and Valencia oranges (*Citrus aurantium* L.) may be caused by calcium deficiency in the albedo tissue of the rind (39).

In these disorders, the shortage of calcium in the tissues causes a general collapse of membrane and cell wall structure, allowing leakage of phenolic precursors into the cytoplasm. Oxidation of polyphenols within the affected tissues gives rise to melanin compounds and necrosis (40). With the general breakdown of cell walls and membranes, microbial infection is frequently a secondary effect. In the case of crease, calcium deficiency may give less adhesion between the cells of the rind, as the middle lamella of these cells is composed largely of calcium salts of pectic acid (39).

Local excess of calcium in the fruit gives rise to goldspot in tomatoes, a disorder that mostly occurs late in the season and that is pronounced with high temperature (41). The disorder 'peteca'

FIGURE 5.4 Fruit of tomato (*Lycopersicon esculentum* Mill. cv Jack Hawkins) (Beefsteak type) showing blossom-end rot (BER). (Photograph by Philip S. Morley.) (For a color presentation of this figure, see the accompanying compact disc.)

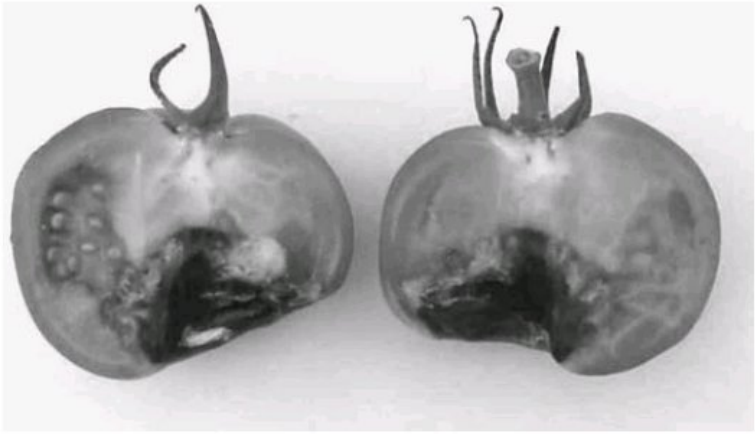

FIGURE 5.5 Cross section of fruit of tomato (*Lycopersicon esculentum* Mill. cv Jack Hawkin) showing advanced symptoms of BER. (Photograph by Philip S. Morley.) (For a color presentation of this figure, see the accompanying compact disc.)

that gives rise to brown spots on the rind of lemons (*Citrus limon* Burm. f.) is associated with localized high concentrations of calcium (as calcium oxalate crystals) and depressed concentrations of boron, although this phenomenon has not yet been shown to be the cause of the disorder (42).

Given the suggestion that calcium may be involved in cell-to-cell adhesion and in the ripening of fruit, it is hardly surprising that in pome fruits, firmness of the fruit is correlated positively with the concentration of calcium present (43). However, this relationship is by no means straightforward; in a study of Cox's Orange Pippin apples grown in two orchards in the United Kingdom, there were lower concentrations of cell wall calcium in the fruit from the orchard that regularly produced firmer fruits than in fruits from other orchards (44). The fruits from this orchard contained higher concentrations of cell wall nitrogen.

Other studies have shown no relationship between calcium concentration in apples at harvest and their firmness after storage, but it is definitely the case that fruit with low Ca^{2+} concentrations are more at risk of developing bitter pit while in storage (45).

5.3.2 Concentrations of Calcium in Plants

5.3.2.1 Forms of Calcium Compounds

Within plants, calcium is present as Ca^{2+} ions attached to carboxyl groups on cell walls by cation-exchange reactions. As approximately one third of the macromolecules in the primary cell wall are pectin (9), it can be seen that a large proportion occurs as calcium pectate. Pectin may also join with anions, such as vanadate, and serve to detoxify these ions. The Ca^{2+} cation will also join with the organic anions formed during the assimilation of nitrate in leaves; these anions carry the negative charge that is released as nitrate is converted into ammonium (46). Thus, there will be formation of calcium malate and calcium oxalacetate and, also very commonly, calcium oxalate in cells.

Calcium oxalate can occur within cells and as extracellular deposits. In a study of 46 conifer species, all contained calcium oxalate crystals (47). All of the species in the Pinaceae family accumulated the compound in crystalliferous parenchyma cells, but the species not in the Pinaceae family had the compound present in extracellular crystals.

This accumulation of calcium oxalate is common in plants in most families. Up to 90% of total calcium in individual plants is in this form (48,49). Formation of calcium oxalate crystals occurs in specialized cells, crystal idioblasts, and as the calcium oxalate in these cells is osmotically inactive their formation serves to lower the concentration of calcium in the apoplast of surrounding cells without affecting the osmotic balance of the tissue (48). A variety of different forms of the crystals occur (49), and they can be composed of calcium oxalate monohydrate or calcium oxalate dihydrate (50).

5.3.2.2 Distribution of Calcium in Plants

Calcium moves toward roots by diffusion and mass flow (51,52) in the soil. A number of calcium-specific ion channels occur in the membranes of root cells, through which influx occurs, but these channels appear to be more involved in enabling rapid fluxes of calcium into the cytoplasm and organelles as part of signalling mechanisms (53). This calcium is then moved into vacuoles, endoplasmic reticulum, or other organelles, with movement occurring by means of calcium-specific transporters (20).

The bulk entry of calcium into roots occurs initially into the cell walls and in the intercellular spaces of the roots, giving a continuum between calcium in the soil and calcium in the root (54). For calcium to move from the roots to the rest of the plant, it has to enter the xylem, but the Casparian band of the endodermis is an effective barrier to its movement into the xylem apoplastically. However, when endodermis is first formed, the Casparian band is a cellulosic strip that passes round the radial cell wall (state I endodermis), so calcium is able to pass into the xylem if it passes into the endodermal cells from the cortex and then out again into the pericycle, through the plasmalemma abutting the wall (55). This transport seems to occur, with the calcium moving into the endodermal cells (and hence into the symplasm) through ion channels and from the endodermis into the pericycle (and ultimately into the much higher concentration of calcium already present in the xylem) by transporters (56,57). Highly developed endodermis has suberin lamellae laid down inside the cell wall around the entire cell (state II endodermis), and in the oldest parts of the root, there is a further layer of cellulose inside this (state III) (55). Although some ions such as K^+ can pass through state II endodermal cells, Ca^{2+} cannot. There are plasmodesmata between endodermis and pericycle cells, even where the Casparian band is well developed, but although phosphate and K^+ ions can pass, the plasmodesmata are impermeable to Ca^{2+} ions.

This restriction in effect limits the movement of calcium into the stele to the youngest part of the root, where the endodermis is in state I. Some movement occurs into the xylem in older parts of the root, and this transport can occur by two means. It is suggested that movement of calcium through state III endodermis might occur where it is penetrated by developing lateral roots, but the Casparian band rapidly develops here to form a complete network around the endodermal cells of the main and lateral roots (55). The second site of movement of calcium into the stele is through passage cells (55). During the development of state II and state III endodermis some cells remain in state I. These are passage cells. They tend to be adjacent to the poles of protoxylem in the stele, and they are the site of calcium movement from cortex to pericycle.

In some herbaceous plants (e.g., wheat, barley, oats), the epidermis and cortex are lost from the roots, especially in drought, so the passage cells are the only position where the symplast is in contact with the rhizosphere (55). Most angiosperms form an exodermis immediately inside the epidermis, and the cells of this tissue also develop Casparian bands and suberin lamellae, with passage cells in some places (55). These passage cells are similarly the only place where the symplasm comes in contact with the rhizosphere.

Because of this restricted entry into roots, calcium enters mainly just behind the tips, and it is mostly here that it is loaded into the xylem (Figure 5.6). Absorption of calcium into the roots may be passive and dependent on root cation-exchange capacity (CEC) (58). Transfer of calcium into roots is hardly affected by respiratory uncouplers, although its transfer into the xylem is affected (54,59).

Once in the xylem the calcium moves in the transpiration stream, and movement around the plant is restricted almost entirely to the xylem (60,61) as it is present in the phloem only at similarly low concentrations to those that occur in the cytoplasm.

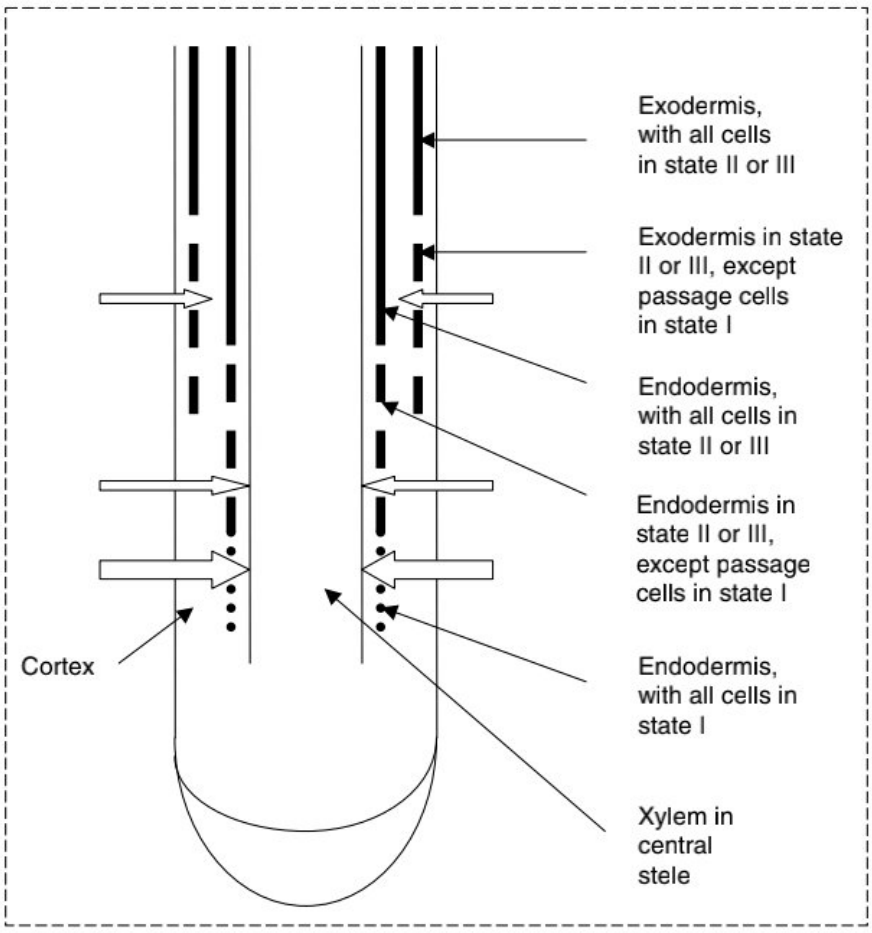

FIGURE 5.6 Diagrammatic representation of longitudinal section of root, showing development of endodermis and exodermis, and points of entry of calcium. (Based on C.A. Peterson and D.E. Enstone, *Physiol. Plant* 97: 592–598, 1996.)

As calcium is not mobile in the phloem, it cannot be retranslocated from old shoot tissues to young tissues, and its xylem transport into organs that do not have a high transpiration rate (such as fruits) is low (22). Its flux into leaves also declines after maturity, even though the rate of transpiration by the leaf remains constant (62), and this response could be related to a decline in nitrate reductase activity as new leaves in the plant take over a more significant assimilatory role (22,63). When a general deficiency of calcium occurs in plants, because of the low mobility of calcium in phloem, it is the new leaves that are affected, not the old leaves, as calcium in a plant remains predominantly in the old tissues (Figure 5.7).

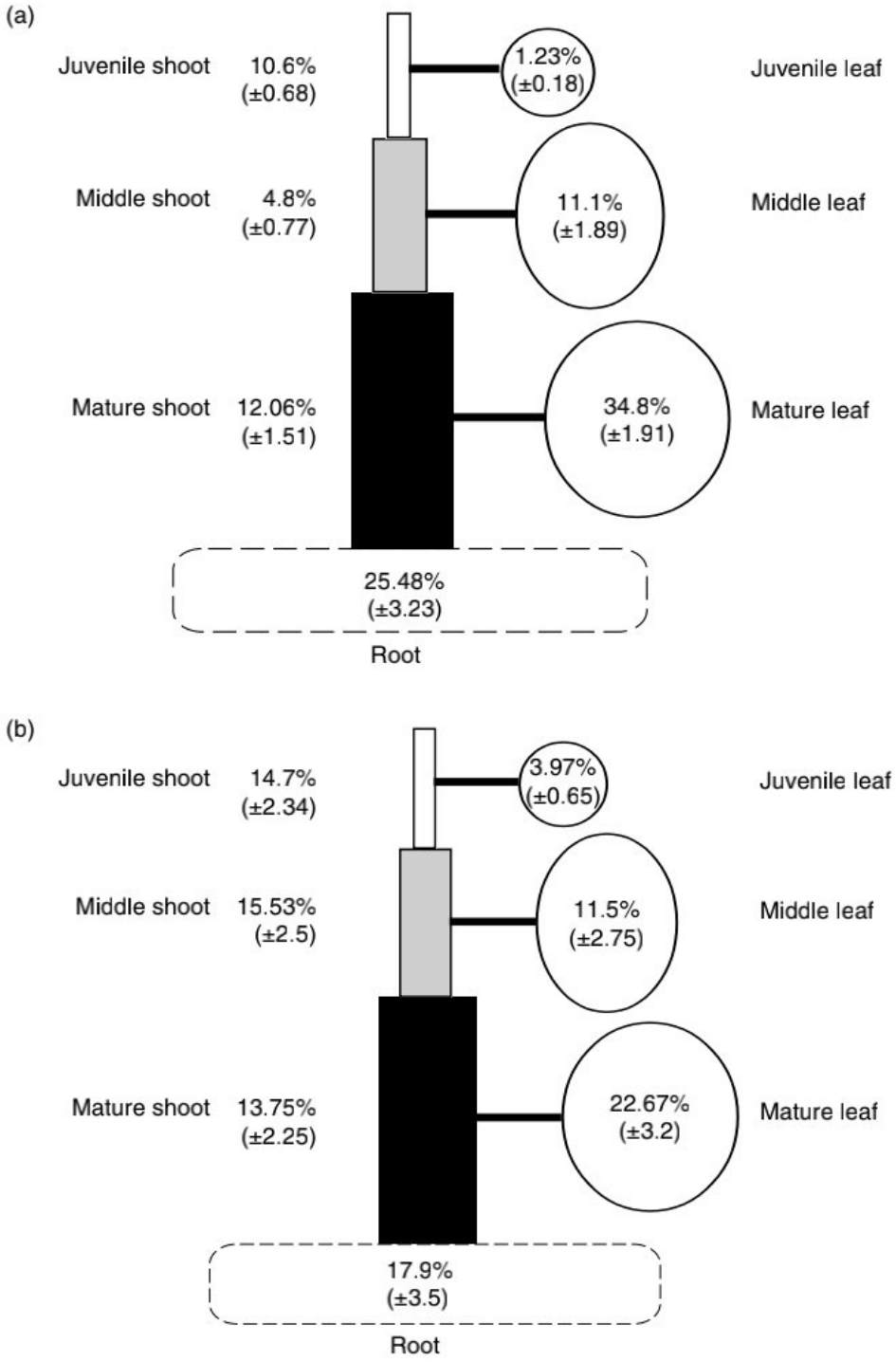

FIGURE 5.7 Distribution of calcium (a) and distribution of dry mass (b) in *Capsicum annuum* cv Bendigo plants grown for 63 days in nutrient solution (values are means of values for nine plants ± standard error).

It was long thought that a direct connection occurs between the amount of transpiration that a plant carries out and the amount of Ca^{2+} that it accumulates. For example, in a study of five tomato cultivars grown at two levels of electrical conductivity (EC) there was a linear, positive relationship between water uptake and calcium accumulation over 83 days (64). However, with the movement of Ca^{2+} in the symplasm of the endodermis apparently being required for xylem loading, it became accepted that Ca^{2+} is taken up in direct proportion to plant growth, as new cation-exchange sites are made available in new tissue. The link with transpiration could therefore be incidental, because bigger plants transpire more. Thus the plant acts as a giant cation exchanger, taking up calcium in proportion to its rate of growth.

Supplying calcium to decapitated plants at increased ion activity (concentration) leads to increased uptake of the ion, a process that appears to contradict this concept. However, in intact plants, the rate of uptake is independent of external ion activity, as long as the ratios of activities of other cations are constant relative to the activity of Ca^{2+} (65,66).

The theory that calcium travels across the root in the apoplastic pathway, until it reaches the Casparian band of the endodermis and at which its passage to the xylem becomes symplastic, is not entirely without problems. White (56,67) calculated that for sufficient calcium loading into xylem, there must be two calcium-specific ion channels per μm^2 of plasmalemma on the cortex side of the endodermis. This possibility is plausible. However, for the flux of calcium to continue from the endodermis into the pericycle there must be 0.8 ng Ca^{2+}-ATPase protein per cell, equivalent to 1.3 mg per gram of root fresh weight. This concentration is greater than the average total root plasmalemma protein concentration in plants. Furthermore, there is no competition between Ca^{2+}, Ba^{2+}, and Sr^{2+} for transport to mouse-ear cress (*Arabidopsis thaliana* Heynh.) shoots, as would be expected if there was protein-mediated transport in the symplast. Some apoplastic transport to the xylem cannot be ruled out.

The walls of xylem vessels have cation-exchange sites on them; in addition to the whole plant having a CEC, the xylem represents a long cation-exchange column with the Ca^{2+} ions moving along in a series of jumps (54). The distance between each site where cation exchange occurs depends on the velocity of the xylem sap and the concentration of Ca^{2+} ions in it (54). Thus, for transpiring organs such as mature leaves, the calcium moves into them quickly, but for growing tissues such as the areas close to meristems, the supply of calcium is dependent on the deposition of cell walls and the formation of new cation-exchange sites (54). It has been suggested that transpiring organs receive their calcium in the transpiration stream during the day, and growing tissues receive their calcium as a result of root pressure during the night (54).

The restriction in movement of calcium to the xylem gives rise to most of the calcium-deficiency disorders in plants. For example, BER (Figure 5.4 and Figure 5.5) in tomatoes occurs because the developing fruits are supplied solutes better by phloem than by xylem as the fruits do not transpire. Xylem fluid goes preferentially to actively transpiring leaves, giving a lower input of calcium into developing fruits (68). A period of hot, sunny weather not only gives rise to so much transpiration that calcium is actively pulled into leaves, but gives rates of photosynthesis that are enhanced to the extent that fruits expand very rapidly. Under these conditions, it is likely that localized deficiencies of calcium will occur in the distal end of the fruits, furthest from where the xylem enters them (the 'blossom' end) (Figure 5.4 and Figure 5.5). Typically, tomatoes grown for harvest in trusses are more susceptible to BER than 'single-pick' types, presumably because the calcium has to be distributed to several developing sinks at the same time. Conditions that promote leaf transpiration, such as low humidity, lower the import of calcium into developing fruits and increase the risk of BER.

It has also been thought in the past that salinity, which increases water potential in the root medium, would likewise restrict calcium import into the fruit, accounting for increased incidence of BER that is known to occur under saline conditions. This effect of salinity could be important in some natural soils, but is also important in glasshouse production of tomatoes as high-electroconductivity (EC) nutrient solutions are sometimes used because they increase dry matter production in fruits and improve flavor. However, it has been observed that if the ion activity ratios $a_K/\sqrt{(a_{Ca} + a_{Mg})}$ and a_{Mg}/a_{Ca} are kept below critical values, the risks of BER developing in high-EC nutrient solutions are

lowered (69). It seems as if one of the causes of increased BER with salinity is normally due to increased uptake of K^+ and Mg^{2+}, which restricts the uptake and distribution of Ca^{2+} ions.

Cultivars differ in susceptibility to BER, with beefsteak and plum types of tomato being particularly susceptible. Susceptibility is related partly to fruit yield, and two susceptible cultivars of tomato (Calypso and Spectra) were shown to have a higher rate of fruit set than a nonsusceptible cultivar (Counter) (70). The so-called calcium-efficient strains of tomato do not have lower incidence of BER, since although they accumulate more dry matter than Ca-inefficient strains, this accumulation is predominantly in the leaves (64). Cultivars with relatively small fruits, such as Counter (70), and with xylem development in the fruit that is still strong under saline conditions (71), are able to accumulate comparatively high proportions of their calcium in the distal end of the fruits under such conditions and are less susceptible to BER (64). However, cultivars with low yields of fruits per plant may show even lower incidence of BER than those with high yields (64).

Losses of tomatoes to BER in commercial horticulture can reach 5% in some crops, representing a substantial loss of potential income. The main approaches to prevent BER are to use less-susceptible cultivars and to cover the south-facing side of the glasshouse (in the northern hemisphere) with white plastic or whitewash to limit the amount of solar radiation of the nearest plants and prevent their fruits from developing too quickly in relation to their abilities to accumulate calcium.

5.3.2.3 Calcicole and Calcifuge Species

In general, calcicole species contain high concentrations of intracellular calcium, and calcifuge species contain low concentrations of intracellular calcium. The different geographic distributions of these plants seem to be largely determined by a range of soil conditions other than just calcium concentration in the soil *per se*. In the calcareous soils favored by calcicoles, in addition to high concentration of Ca^{2+}, pH is high, giving low solubility of heavy metal ions and high concentrations of nutrient and bicarbonate ions. In contrast, the acid soils favored by calcifuges have low pH, high solubility of heavy metal ions, and low availability of nutrients (5).

The growth of calcicole species is related strongly to the concentration of calcium in the soil, but the inability of calcicole species to grow in acid soils is linked strongly to an inability to tolerate the high concentrations of ions of heavy metals, in particular Al^{3+}, Mn^{2+}, and Fe^{3+} (5,72). For calcifuge species, the difficulty in growing in a calcareous soil stems from an inability to absorb iron, although in some calcareous soils low availability of phosphate may also be a critical factor.

In an experiment with tropical soils in which the sorption of phosphate from $Ca(H_2PO_4)_2$ solution (and its subsequent desorption) were measured, pretreating the soil with calcium sulfate solution increased the sorption of phosphate (73). In the most acid of the soils tested, sorption of phosphate was increased by 93%. Because the extracts of the soil became more acid following calcium sulfate treatment, it appears that the calcium was attracted to the sites previously occupied by H^+ ions, and when present, itself offered more sites for sorption of phosphate ions. Where the supply of phosphorus to plants is limited because it is sorbed to soil inorganic fractions, it seems as if sorption to calcium is more difficult to break than sorption to other components. In an experiment in which wheat (*Triticum aestivum* L.) and sugar beet (*Beta vulgaris* L.) were grown in a fossil Oxisol, with mainly Fe/Al-bound P, and in a Luvisol, a subsoil from loess with free $CaCO_3$ and mainly Ca-bound P, both species (but particularly the sugar beet) were able to mobilize the Fe/Al-bound P more than the Ca-bound P (74).

Some plants are much more efficient than others at taking up phosphate from calcium-bound pools in the soil. One efficient species is buckwheat (*Fagopyrum esculentum* Moench). In a comparison of this species and wheat, the buckwheat took up 20.1 mg P per pot compared with 2.1 mg P per pot for wheat if nitrogen was supplied as nitrate (75). Changing the nitrogen supply to ammonium nitrate increased phosphorus accumulation by the wheat largely, with very little effect on the buckwheat, indicating that it is the capacity of buckwheat to acidify the rhizosphere even when the nitrogen supply is nitrate that makes buckwheat able to utilize this firmly bound source of phosphorus.

For calcifuge species growing on calcareous soils, it seems as if the availability of iron is the most significant factor affecting plant growth, with chlorosis occurring due to iron deficiency. However, this deficiency is caused largely by immobilization of iron within the leaves, not necessarily a restricted absorption of iron (76,77). Calcicole species seem to make iron and phosphate available in calcareous soils by exudation of oxalic and citric acids from their roots (78). The high concentrations of bicarbonate ions in calcareous soils seem to be important in inhibition of root elongation of some calcifuge species (79).

5.3.2.4 Critical Concentrations of Calcium

The concentrations of calcium in plants are similar to the concentrations of potassium, in the range 1 to 50 mg Ca g^{-1} dry matter (Mengel, this volume). Most of the calcium is located in the apoplast, and where it is present in the symplast, it tends to be stored in organelles or vacuoles or is bound to proteins. The concentration of free Ca^{2+} in a root cortical cell is of the order of 0.1 to 1.0 mmol m^{-3} (54).

In general, monocotyledons contain much less calcium than dicotyledons. In an experiment comparing the growth of ryegrass (*Lolium perenne* L.) and tomato, the ryegrass reached its maximum growth rate when the concentration of calcium supplied gave a tissue concentration of 0.7 mg g^{-1} dry mass, whereas tomato reached its maximum growth rate only when tissue concentration was 12.9 mg g^{-1} (80,81). This difference between monocotyledons and dicotyledons is dictated by the CEC of the two groups of plants. In algal species, where the cell wall is absent and CEC is consequently low, calcium is required only as a micronutrient (82).

Tissue concentrations of calcium can vary considerably according to the rate of calcium supply. In a study by Loneragan and Snowball (81), internal Ca^{2+} concentrations were reasonably constant for 0.3, 0.8, and 2.5 µM calcium in the flowing nutrient solutions for each plant species tested, but with 10, 100, or 1000 µM Ca^{2+} supply, internal Ca^{2+} concentrations were noticeably higher. In a recent study of chickpea (*Cicer arietinum* L.), nine different Kabuli (large-seeded) accessions had a mean concentration of Ca^{2+} in nodes 4 to 7 of the shoot of 17.4 mg g^{-1} dry mass after 33 days of growth, and 10 different Desi (small-seeded) accessions had a mean Ca^{2+} concentration of 17.1 mg g^{-1} dry mass (83). In the Kabuli accessions, the range was between 13.5 and 20.6 mg g^{-1}, compared with between 13.1 and 19.0 mg g^{-1} in the Desi accessions, so different genotypes of the same species grown under the same conditions seem to contain very similar shoot calcium concentrations.

There are considerable amounts of data regarding what the critical concentrations of calcium are in different plants and different species. For data on these concentrations in a large number of species, the reader is referred to some special publications (84,85).

In a study of three cultivars of bell pepper, mean tissue concentrations ranged only from 1.5 to 1.8 mg g^{-1} dry mass in the proximal parts and from 0.95 to 1.3 mg g^{-1} dry mass in the distal part of healthy fruits. concentrations in fruits suffering BER were between 0.6 and 1.0 mg g^{-1} (86). Concentrations of calcium in fruits of cucumber (*Cucumis sativus* L.), a plant that is not susceptible to BER, are typically three to seven times these values (87).

There is one important exception to the finding that internal calcium concentrations are relatively constant regardless of how plants are grown. Plants supplied with nitrogen as ammonium tend to have much lower concentrations of cations, including calcium, than plants supplied with nitrate (22). Thus, tomato plants supplied with ammonium-N are more prone to BER than plants grown on nitrate.

5.3.2.5 Tabulated Data of Concentrations by Crops

Concentrations of Ca^{2+} in shoots and fruits of some crop species are reported in Table 5.1 and Table 5.2.

TABLE 5.1
Deficient and Adequate Concentrations of Calcium in Leaves and Shoots of Various Plant Species

Plant Species	Plant Part	Type of Culture	Concentration in Dry Matter (mg kg^{-1})		Reference	Comments
			Deficient	Adequate		
Avena sativa L. (oat)	Tops	Pot culture, soil	1100–1400	2600	88	Plants at flowering
	Straw	Sand culture	1000–1400	3600–6400	88	At harvest
Bromus rigidus Roth	Shoot	Flowing nutrient solution	900	1010	81	Plants grown in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively
Capsicum annuum L. (pepper)	Leaves	Nutrient solution		Up to 30000 5000	89	Mature leaves Juvenile leaves
Citrus aurantium L. (orange)	Leaves Shoots	Sand culture	1400–2000 2300–2800	14800 11700	88	Measurements taken in September
Ficus carica L. (fig)	Leaves	Orchard		30000 30000 29000 35000	90	Values for May, July, September and October. 10 trees surveyed in 9 areas of 2 orchards, for 3 years
Fragaria x ananassa Duchesne (strawberry)	Leaves	Sand culture	2300/9000	15000	91	'Adequate' plants had 1% of leaves with tipburn. 'Deficient' plants had 33.2% of leaves with tipburn (plants supplied 1/40th control Ca and 3x K) or 9% of leaves with tipburn (plants supplied control Ca and 3x K)
Hordeum vulgare L. (barley)	Shoots	Flowing nutrient solution	1100	7300	81	Plants grown in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively
Linum usitatissimum L. (flax)	Tops	Field	2000–4500	3700–5200	88	
Lolium perenne L. (perennial ryegrass)	Shoots	Flowing nutrient solution	600	10800	81	Plants grown in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively
Lupinus angustifolius L.	Shoots	Flowing nutrient solution	1400	13900	81	Plants grown in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively
Lycopersicon esculentum Mill. (tomato)	Leaf blade	Sand culture	1700	16100	36	Upper leaves (yellow in deficient plants)
	Leaf blade		11000	38400		Lower leaves (still green in deficient plants)
	Petioles		1100	10800		Upper petioles
	Petioles		2600	22300		Lower petioles
	Stem		Trace	6700		Upper stems

TABLE 5.1 (*Continued*)

Plant Species	Plant Part	Type of Culture	Concentration in Dry Matter (mg kg^{-1})		Reference	Comments
			Deficient	Adequate		
	Stem		5300	9900		Lower stems
	Shoots	Flowing nutrient solution	2700	24900	81	Plants grown in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively
Malus pumila Mill. [*M. domestica* Borkh.] (apple)	Leaves		7200		88	Leaves of terminal shoot, stated value below which deficiency symptoms occur
Medicago sativa L. (alfalfa)	Shoots	Flowing nutrient solution	1100	15000	81	One cultivar, in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively
Nicotiana tabacum L. (tobacco)	Leaves	Field trial	9400–13000	13300–24300	88	
Phaseolus lunatus L. (lima bean)	Stem		6000	9000	88	Poor seed set below first value, good seed set above second value
Prunus persica (L.) Batsch (peach)	Leaves	Orchard		14500 17000 18200	92	Soil pH 5.6 Soil pH 5.9 Soil pH 6.2
Prunus insititia L. *Prunus domestica* L. *Prunus salicina* (Lindl.) × *Prunus cerasifera* (Ehrh.) (plum)	Leaves	Nutrient solution		5300/8200 6600/10300 6300/10100	93	Values for days 45 and 96
Secale cereale L. (rye)	Shoots	Flowing nutrient solution	900	8300	81	Plants grown in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively
Solanum tuberosum L. (potato)	Young leaves	Nutrient solution	Below 900	Above 4500	18	21-day-old plants
Trifolium subterraneum L. (subterranean clover)	Shoots	Flowing nutrient solution	1400	19100	81	One cultivar, in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively
Triticum aestivum L. (wheat)	Shoots	Flowing nutrient solution	800	4700	81	One cultivar, in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively
Zea mays L. (corn)	Shoots	Flowing nutrient solution	300	9200	81	Plants grown in 0.3 and 1000 mmol m^{-3} Ca^{2+}, respectively

Note: Values in dry matter.

5.4 ASSESSMENT OF CALCIUM STATUS IN SOILS

5.4.1 Forms of Calcium in Soil

Calcium is the main exchangeable base of clay minerals and, as such, is a major component of soils. One of the most important natural sources of calcium is underlying limestone or chalk, where it occurs as calcium carbonate (calcite). Calcium in rocks also occurs as a mixture of calcium and magnesium carbonates (dolomite). Soils over such rocks often contain large amounts of calcium carbonate, although not invariably so. The soils may not have been derived from the rock, but have

TABLE 5.2
Deficient and Adequate Concentrations of Calcium in Fruits of Various Plant Species

Plant Species	Plant Part	Type of Culture	Concentration in Fresh Matter (mg kg^{-1})		Reference	Comments
			Deficient	Adequate		
Capsicum annuum L. (pepper)	Fruits	Nutrient solution		1500–1800 (dry wt)	86	Proximal pericarp tissue
				1000–1200 (dry wt)		Distal pericarp tissue (healthy)
			600 (dry wt)			Distal pericarp tissue (BER-affected)
Cucumis sativus L. (cucumber)	Fruits	Rockwool and nutrient solution		3000–6000 (dry wt)	87	Range of values according to salinity treatment and size of fruit
Fragaria x ananassa Duchesne (strawberry)	Fruits	Sand culture		65/120/201 (559/1192/2060) (dry wt)	91	Values from left to right for plants that had 33.2% of leaves with tipburn (plants supplied 1/40th control Ca and 3x K), 9% of leaves with tipburn (plants supplied control Ca and 3x K) 1% of leaves with tipburn (control)
Lycopersicon esculentum Mill. (tomato)			210/240 (dry wt)	280 (dry wt)	94	For 'deficient' values, first value is for an experiment in which 44.5% of fruit had BER, second value for an experiment in which 18.9% of fruit had BER. For 'adequate' value 0.9% of fruit had BER
Malus pumila Mill. [*M. domestica* Borkh.] (apple) cv Jonagold	Fruitlets in July	34 different orchards	105	190	95	Fruitlets with 'deficient' concentration showed much higher incidence of physiological disorders in storage
cv Cox's Orange Pippin	Fruit at harvest	Orchard grown	33 36 38	64 64 62	45	Range found in fruit harvested in 3 consecutive years. Fruit with the lower values had higher incidence of bitter pit
cv Cox's Orange Pippin			45		96	Minimum level for recommending fruit for controlled atmosphere storage. Below this level bitter pit is common
Pyrus communis (pear)	Fruit	4 Orchards	60	76	97	Values of 60 and 67 mg kg^{-1} fresh weight in fruit from different orchards linked with high incidence of internal breakdown and cork spot

Note: Values in fresh matter, unless shown to contrary.

come from elsewhere and been deposited by glaciers, and furthermore, although calcium carbonate is sparingly water soluble, it can be removed by leaching so that the overlying soil may be depleted of calcium carbonate and be acidic.

Some soils contain calcium sulfate (gypsum), but mostly only in arid regions. A further source of calcium in soils is apatite [$Ca(OH_2).3Ca(PO_4)_2$] or fluorapatite [$Ca_5(PO_4)_3F$]. Chlorapatite [$Ca_5(PO_4)_3Cl$] and hydroxyapatite [$Ca_5(PO_4)_3OH$] also exist in soils (98). Calcium is also present in the primary minerals augite [$Ca(Mg,Fe,Al)(Al,Si)_2O_6$], hornblende [$NaCa_2(Mg,Fe,Al)_5(Si,Al)_8O_{22}(OH)_2$], and the feldspar plagioclase (any intermediate between $CaAl_2Si_2O_8$ and $NaAlSi_3O_8$) (98).

Within the fraction of soils where particles are as small as clay particles, calcium occurs in gypsum, calcite, hornblende, and plagioclase. Sherman and Jackson (99) arranged the minerals in the clay fractions of the A horizons of soils in a series according to the time taken for them to weather away to a different mineral. These calcium-containing minerals are all early in this sequence, meaning that calcium is lost from the minerals (and becomes available to plants) early in the weathering process, but has been entirely lost as a structural component in more mature soils (98). Any calcium present in these more mature soils will be present attached to cation-exchange sites, where it usually constitutes a high proportion of total exchangeable cations, so the amounts present depend on the CEC of the soil.

Concentrations of Ca^{2+} in soils may be affected by ecological disturbance. Acid depositions are known to decrease Ca^{2+} concentrations in soils, which while not necessarily affecting plant yields directly may have a big impact on ecosystem dynamics. Acid deposition on the coniferous forests of the Netherlands has been shown to give rise to fewer snails, and the birds that feed on the snails have fewer surviving offspring due to defects in their eggs (100). This effect seems to be related largely to the abundance of snails being depressed by low calcium concentrations in the plant litter. In terms of how serious this problem might prove to be, it should be noted that changes in soil Ca^{2+} concentration caused by acid rain are less than 1 g Ca^{2+} m^{-2} $year^{-1}$. This change is small compared with a transfer of 3.3 to 4.7 g Ca^{2+} m^{-2} $year^{-1}$ from mineral soil to young forest stands (101).

Experiments on the Hubbard Brook Experimental Forest in New Hampshire, USA, have shown that calcium is lost from ecosystems following deforestation. This loss is true for other cations and also for nitrate. In the Hubbard Brook experiment, during the 4 years following deforestation, the watershed lost 74.9 kg Ca^{2+} ha^{-1} $year^{-1}$ as dissolved substances in the streams, compared with 9.7 kg Ca^{2+} ha^{-1} $year^{-1}$ in a watershed where the vegetation had not been cut down (102). This increased loss was attributed partly to increased water flows due to decreased water loss by transpiration, but more importantly through the breakdown of the plant material enhancing the turnover of the nitrogen cycle and the consequent generation of H^+ ions, thereby releasing cations from the cation-exchange sites of the soil (102). Recent studies have shown that calcium loss continues for at least 30 years, with the longer-term loss possibly occurring because of the breakdown of calcium oxalate in the forest soil after removal of the trees (103).

5.4.2 Soil Tests

The main test for soil calcium is to calculate the amount of the limestone required for a particular crop on a particular soil (see 5.5.2 below).

5.4.3 Tabulated Data on Calcium Contents in Soils

Concentrations of Ca^{2+} in soils typical of a range of soil orders are shown in Table 5.3.

5.5 FERTILIZERS FOR CALCIUM

5.5.1 Kinds of Fertilizer

The most common application of calcium to soils is as calcium carbonate in chalk or lime. This practice occurred in Britain and Gaul before the Romans (Pliny, quoted in Ref. (105)). It does not

TABLE 5.3
Calcium Concentration, Cation Exchange Capacity and pH of Top Layers of Some Representative Soils

Soil	Soil Order	Ca^{2+} Concentration (mmol kg^{-1})	CEC ($cmol_c$ kg^{-1})	pH
Typic Cryoboralf, Colorado, 0–18 cm depth	Alfisol	30.5	13.3	5.9
Typic Gypsiorthid, Texas, 5–13 cm depth	Aridisol	100.0	21.6	7.9
Typic Ustipsamment, Kansas, 0–13 cm depth	Entisol	9.5	52.0	6.6
Typic Dystrochrept, West Virginia, 5–18 cm depth	Inceptisol	5.0	11.4	4.9
Typic Argiustoll, Kansas, 0–15 cm depth	Mollisol	73.5	23.8	6.6
Typic Acrustox, Brazil, 0–10 cm depth (low CEC below 65 cm)	Oxisol	2.1	20.5	5.0
Typic Haplorthod, New Hampshire, 0–20 cm depth	Spodosol	14.5	25.7	4.9
Typic Umbraquult, North Carolina, 0–15 cm depth	Ultisol	2.0	26.2	3.9
Typic Chromoxerert, California, 0–10 cm depth	Vertisol	84.0	24.6	7.8

Source: Data from USDA, *Soil Taxonomy: A Basic System of Soil Classification for Making and Interpreting Soil Surveys*. Agricultural Handbook Number 436. Washington, DC: USDA, 1975.

come strictly under the definition of fertilizer, as the main functions of the calcium carbonate are to make clay particles aggregate into crumbs, thereby improving drainage, and to lower soil acidity.

Despite the observation that addition of gypsum to tropical soils may increase the sorption of phosphate (73), it seems as if this effect is not universal, and it is the change in pH brought about by limestone or dolomite that is more important in aiding phosphate sorption than the provision of Ca^{2+} ions. In an experiment on addition of calcium carbonate, dolomite, gypsum, and calcium chloride to the Ap horizon of a Spodosol, all additions increased the retention of phosphorus in the soil except the calcium chloride (106). The order of this increase was calcium carbonate > dolomite > gypsum, which followed the order of increase in pH. Gypsum is not expected to increase pH of soil, but it is likely that this pH change, and the consequent effect on phosphorus sorption, was due to impurities, likely lime, in the gypsum used.

Following an addition of lime, Ca^{2+} from the calcium carbonate ($CaCO_3$) exchanges for $Al(OH)_2^+$ and H^+ ions on the cation-exchange sites. The $Al(OH)_2^+$ ions give rise to insoluble $Al(OH)_3$ that precipitates; the H^+ ions react with bicarbonate $(HCO_3)^-$ that arises during the dissolution of calcium carbonate in the soil water. This reaction leads to the formation of carbon dioxide, lost from the soil as a gas, and water, both of which are neutral products (107).

In very acid soils, there is a shortage of available calcium, and application of calcium carbonate will help rectify this problem. One of the outcomes of adding calcium would be to displace Al^{3+} and H^+ ions from the root plasmalemma, where they would otherwise be displacing Ca^{2+} ions (108). Experiments with alfalfa (*Medicago sativa* L.) grown on acid soils showed that while application of lime increased calcium concentrations in the shoots, it also decreased concentrations of aluminum, manganese, and iron. As those cultivars that were the least sensitive to the acid soil had

lower concentrations of these three elements anyway, it seems as if the beneficial effect of the lime was in modifying soil pH rather than supplying additional Ca (109).

The more neutral or alkaline pH brought about by liming gives a more favorable environment for the microorganisms of the nitrogen cycle, enhancing the cycling of nitrogen from organic matter. It also increases the availability of molybdenum, and it restricts the uptake of heavy metals (107).

Another action of lime is to decrease the concentration of fluoride in tea (*Camellia sinensis* L.) plants. This crop accumulates high concentrations of fluoride from soils of normal fluoride concentration. The action of liming in limiting fluoride concentrations in tea plants is surprising given that the uptake of fluoride is higher from more neutral soil than from acid soil and given that liming may increase the water-soluble fluoride content of the soil (110). In this case, it appears that the Ca^{2+} in the lime either affects cell wall and plasmalemma permeability or changes the speciation of the fluoride in the soil.

In some instances calcium sulfate (gypsum) may be applied as a fertilizer, but this application is more for a source of sulfur than calcium or to improve soil structure. Apatite (applied as rock phosphate) and superphosphate contain twice as much calcium by weight as the phosphorus that they are used primarily to supply, and triple superphosphate contains two thirds as much calcium as phosphorus (98). One situation where gypsum is particularly useful is in the reclamation of sodic soils, where the calcium ions replace the sodium on the cation-exchange sites and the sodium sulfate that results is leached out of the soil (107).

Calcium nitrate and calcium chloride are regularly used as sprays on developing apple fruits to prevent bitter pit (111). Of the two calcium forms, nitrate is less likely to cause leaf scorch, but some varieties of apple are susceptible to fruit spotting with nitrate. Dipping the fruit in $CaCl_2$ immediately after harvest supplements the regular sprays (111). Spraying apple trees with calcium nitrate during the cell expansion phase of fruit growth increases the nitrogen and the calcium concentrations in the fruit at harvest and gives firmer fruit at harvest and after storage (112).

Application of calcium salts to sweet cherry (*Prunus avium* L.) fruits just before harvest may also decrease the incidence of skin cracking that follows any heavy rainfall at this time (43). Multiple applications throughout the summer give better protection, and $CaCl_2$ is better than $Ca(OH)_2$, as the latter can cause fruit to shrivel in hot seasons (113). Recent research has shown that spraying $CaCl_2$ and boron with a suitable surfactant on strawberry plants at 5-day intervals from the time of petal fall gives fruits that are firmer and more resistant to botrytis rot at harvest, or after 3 days storage, than untreated fruits; after the 3 days, they have a higher concentration of soluble solids and more titratable acidity (114). Treating pineapples with lime during their growth seems to lower the incidence of internal browning that arises in the fruit in cold storage, and increases their ascorbic acid content (38). The fruit of tomato cultivars particularly susceptible to BER (e.g., the beefsteak cultivar Jack Hawkins) may be sprayed with calcium salts, although the efficacy of this treatment is doubtful.

There are also calcium treatments for improving shelf life and fruit quality that are used after harvest. For example, dipping cherry tomatoes in 25 mM $CaCl_2$ after harvest increases apoplastic calcium concentrations and decreases incidence of skin cracking (115). Vacuum infiltration of Ca^{2+} increases the time of ripening of peaches, so that they can be stored for longer periods before sale, and such use of calcium salts is common for tomatoes, mangoes (*Mangifera indica* L.), and avocadoes (*Persea americana* L.) (116). The firmness of plums (*Prunus domestica* L.) is increased by pressure infiltration of 1 mM $CaCl_2$ (117).

There is some evidence that supply of supplementary calcium nitrate partially alleviates the effects of NaCl salinity in strawberry in hydroponic culture (118) and in cucumber and melon (*Cucumis* spp. L.) in irrigated fields (119).

5.5.2 Application of Calcium Fertilizers

Liming is carried out by application of $CaCO_3$ in limestone, a process that is described in some detail in Troeh and Thompson (98). The neutralizing capacity of the limestone used is measured by

comparing it to calcite, which is $CaCO_3$, with a *calcium carbonate equivalent* (CCE) of 100%. The fineness of the lime affects its efficiency for liming, and the CCE and fineness and hardness of the lime together give the *effective calcium carbonate equivalent* or *reactivity*. Application should occur when the soil is dry or frozen, to avoid damage to the soil by the vehicles carrying the lime. Although soil testing will determine if an application is required, it is often the practice to apply lime a year ahead of a crop in a rotation that has a strong lime requirement (often a legume). An application once every 4 to 8 years is usually effective. Limestone, burned lime (CaO), or slaked lime [$Ca(OH)_2$] can also be used. Burned lime has a CCE of 179% and slaked lime a CCE of 133%.

The amount of lime required is determined from soil analysis, either by a pH base saturation method or a buffer solution method (98,120). The soil requirement for lime, defined, for example, as the number of tonnes of calcium carbonate required to raise the pH of a hectare of soil 200 mm deep to pH 6.5 (120), will depend on the initial pH and also on CEC of the soil. Most soils have a much greater proportion of their cations attached to cation-exchange sites than in solution, meaning that a high proportion of the H^+ ions present are not measured in a simple pH test. Adding lime to the soil neutralizes the acidity in the soil solution, but the Ca^{2+} ions displace H^+ ions from the exchange sites, with the potential to make the pH of the soil acidic once more, and this acidity is neutralized by reaction of the H^+ with the lime. The H^+ in soil solution is called the *active acidity*, and the H^+ held to the exchange sites on soil colloids is called the *reserve acidity* The greater the CEC, the greater the reserve acidity and the greater the lime requirement (98).

In the pH-base saturation method, the percent base saturation of the soil, the CEC of the soil and the initial pH all have to be measured. To calculate how much lime should be added the percent base saturation at the initial and at the target pH value are read off a graph, and the amount of $CaCO_3$ to be added is calculated from the difference in percent base saturation at the two pH values multiplied by the CEC (98).

In the buffer solution method, a sample of the soil is mixed with a buffer, and the amount of lime required is read off a table from the value of decrease in buffer pH on adding the soil (120).

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