The long-distance abscisic acid signal in the droughted plant: the fate of the hormone on its way from root to shoot

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Abstract

Abscisic acid (ABA) is a potent molecule that certainly modifies stomatal behaviour and plant water loss and probably acts to modify the growth of leaves. The hormone is synthesized both in the leaves and the roots of the plant and in the soil and may move freely from plant to soil and soil to plant. It can also move rapidly through the plant in both the xylem and the phloem and will partition between different compartments in different tissues largely as a function of pH. It is described here how perturbations in soil conditions around the roots and the water status of the air can modify the fluxes of ABA around the plant and its accumulation in different compartments and different tissues. These fluxes can be interpreted as signals of different stresses imposed on the plant and consideration is given to how different perturbations can exert subtle changes which are manifest as modified shoot growth rates and functioning. Most emphasis in the discussion is placed upon the plant’s responses to the imposition of soil and atmospheric drought.

Key words: ABA, stress, nutrient deficiency, xylem and phloem transport.

Introduction

The role of abscisic acid as a root-to-shoot stress signal is now well established. Transgenic plants with low capacity for ABA synthesis show disrupted stomatal behaviour and the grafting of deficient roots onto sufficient shoots fails to restore normal stomatal response (Borel et al., 2001). Further, the removal from the plant of roots in dry soil can result in reopening of stomata and increased shoot growth rates (Gowing et al., 1990). These responses are very unlikely to be a result of increased water supply from a reduced complement of roots.

The suggestion is that roots are able to ‘measure’ decreasing soil water availability during a period of drought (perhaps as a change in root water status), which results in an increased release of ABA from the stelar tissues of roots to the xylem vessels. After xylem transport to the shoot, guard cells respond rapidly and sensitively to increased ABA concentrations resulting in reduced transpirational water loss. A slower response by shoot meristems to extra ABA (much of which will arrive in the shoot from the root, Fig. 1, point 8) can be seen in inhibited leaf production, growth and development. There are many other documented changes in shoot morphology which take place in the droughted plant (e.g. changed stomatal differentiation, synthesis of epicuticular waxes, formation of thorns, hairs etc). Similar responses can be induced by external application of ABA (Trewavas and Jones, 1991). These may contribute to an improved adaptation to the long-term water shortage of plants growing in extreme and arid habitats. It will be argued here that because of the way that the ABA molecule partitions between different compartments of the plant, these responses will largely be due to chemical signals arriving in the shoot via the xylem stream.
Abscisic acid in the rhizosphere (1)

Roots do not grow in a hormone-free environment. The soil solution contains all the known plant hormones including abscisic acid (Frankenberger and Arshad, 1995). Much of the soil ABA originates from plant roots, which may release ABA, especially under non-transpiring conditions. Soil fungi are also sites of synthesis. Soil ABA is necessary to maintain an ABA equilibrium between roots and the external medium. Computer simulations (Slovik et al., 1995) predicted soil ABA concentrations in the low nanomolar range to keep this equilibrium and to prevent dramatic ABA loss to the rhizosphere. This has been confirmed experimentally (Hartung et al., 1996).

Without the ABA equilibrium with the soil, ABA leakage from the roots could greatly disrupt the root-to-shoot signalling process.

Roots can take up external ABA. The uptake is reduced if Casparian bands have been formed in the hypodermis (Freundl et al., 2000). The concentration of soil ABA is high in acid soils where ABA degradation by soil microorganisms is weak. In drying soils the ABA concentration increases above that level which must be expected by concentration as water is removed. It is not clear how much of this external ABA will be taken up under these conditions, as water and solute movement in the soil will slow dramatically as it dries. Localized soil drying around the roots will also restrict the uptake of hormones in the soil solution.

ABA loss to the surrounding medium (2)

As shown above ABA must be present in the soil to maintain the equilibrium between internal and external ABA. Further release must be expected when growing in...
ABA accumulation in roots by increased biosynthesis (3)

Roots are fully equipped with all the enzymes and precursors that synthesize ABA. However, information is still lacking on the enzymatic step of ABA biosynthesis which is responsible for the ‘measurement’ of reduced soil water availability. Besides a reduced water content, soils of arid environments often exhibit high salt concentrations, high soil strength, alkaline pH values, and low amounts of nutrients. The effects of all these factors on ABA accumulation have been investigated in the past. Reduced soil water, increased salt concentrations (NaCl and CaCl2), ammonium supply, phosphate deficiency, and high mechanical impedance stimulate ABA accumulation in roots by increased ABAxyl (Jeschke et al., 1997a; Wolf et al., 1990; Hartung and Jeschke, 1999).

Recent papers (Hussain et al., 1999) show that increased mechanical impedance in soils increases ABA concentrations in the xylem which may explain stomatal limitations shown by these plants. The stimulus for this increase is not clear, but in much work it seems that increased soil strength can result in plant water deficit and the resulting synthesis of extra ABA (Hartung et al., 1994). This is not always the case, however, (Masle and Passioura, 1990). Root water deficits may be expected to increase the delivery of ethylene to shoots and this hormone can act to restrict shoot growth (Hussain et al., 2000). Synthesis of ethylene can also result from anaerobic conditions in the soil which also often accompany soil compaction. Oxygen stress will result in reduced ABA delivery to shoots (Else et al., 1995).

Radial ABA-transport in the roots (4)

ABA can be translocated to the xylem in the symplast as well as in the apoplast. If transported symplastically large amounts of the hydrophilic ABA anions have to be transported across the plasma membrane of the xylem parenchyma cells into the apoplast of the stele. In this case changes in water flow would dilute or concentrate ABAxyl rapidly resulting in dramatic concentration fluctuations within short time periods (Else et al., 1994). Freundl et al have shown that ABA can be dragged directly across the endodermis of maize and sunflower with the water (Freundl et al., 1998). Such an apoplastic bypass flow of ABA buffers these fluctuations of ABAxyl and plays an important role for an ABA homeostasis in xylem vessels in unstressed plants. Such homeostasis has been observed by Tardieu et al. (Tardieu et al., 1992) and others in plants growing in the field subjected to sometimes abrupt changes in evaporative demand.

ABA redistribution between stem tissue and xylem vessels (5)

Very little information is available about the fate of ABA during its long-distance xylem transport through stems. Above nodes ABAxyl tends to become lower because a significant amount of ABA is diverted to the leaves (see all the ABA transport models published by Jeschke et al., 1997a, b; Jokhan et al., 1999). However, ABAxyl is also affected by a redistribution between the xylem and the surrounding stem tissue.

Sauter and Hartung (unpublished results) have analysed ABA in the xylem sap and in tissues after its passage through an isolated bean stem internode. It seems that ABA can be fed from the stem parenchyma into the xylem if ABAxyl is low. When ABA metabolism was inhibited by substances such as tetcyclacis the ABA release from the stem tissues to the xylem was also increased. On the other hand ABA redistributes from the xylem to the stem tissues if ABAxyl occurs in the concentration range that may be expected under stress conditions.

While applying a vacuum to the upper cut surface of an isolated 8 cm long first internode of runner bean, an ABA free buffer was perfused. On its passage through the xylem vessels this buffer was enriched with ABA (13–23 nM) from the surrounding tissue. When 100 nM
ABA was added to the medium at pH 7 (Fig. 2). ABA rose to 80–100 nM within 10 min. Under slightly acid conditions (pH 6) ABA increased gradually, reaching 50 nM after 40–60 min indicating that some of the xylem sap ABA has been taken up by the internode tissue. Neutral or slightly alkaline xylem saps have been detected in stressed plants (Wilkinson et al., 1998), whereas pH of well-watered plants is lower by 1 pH unit. Figure 2 shows that during transport at pH 6 (unstressed pH), substantial amounts of ABA are taken up by the surrounding tissue, at pH 7 (stressed pH and a more effective anion trap) xylem sap is enriched with ABA originating from the stem parenchyma cells.

ABA in leaves (6, 10)

Leaf ABA biosynthesis is increased only when leaves fall to the point where turgor approaches zero, suggesting that ABA transport via xylem is necessary for the regulation of growth and stomatal behaviour under conditions of mild drought stress. Henson et al. (Henson et al., 1989) and others show that there are many circumstances where shoot growth and functioning are influenced under circumstances where shoot water relations are not changed. Very mild soil water deficits can have profound effects on community composition and functioning and it has been suggested that root-to-shoot chemical signalling can explain many of these observed changes (Davies and Gowing, 1999). However, import of ABA to the leaf does not necessarily result in an accumulation of ABA in the leaves. Under phosphate deficiency imported xylem sap ABA is degraded after having acted on the stomata. As a result ABA is not accumulated in the leaves despite a stimulated import via the xylem.

The flux of ABA to the leaves of even well-watered plants may be enough to close the stomata if ABA is able to reach the sites of action on the guard cells. In well-watered plants xylem sap pH and therefore apoplastic ABA arriving in the leaf will be rapidly taken up into the alkaline compartments in the leaf (the symplast). Wilkinson and Davies have shown that a small alkalinization of the xylem sap (less than a pH unit) can be enough to close stomata significantly, without extra ABA (Wilkinson and Davies, 1997), and these workers have hypothesized that this is because the hormone arriving in the leaf resides for longer in the apoplast at these pHs and therefore gains access to the guard cells (Fig. 3). pH changes of this magnitude are routinely observed in plants in drying soil in response to quite small reductions in soil water content (Wilkinson et al., 1998). Bacon et al. have shown a similar subtle modification of the leaf growth response to ABA as a function of apoplastic ABA (Bacon et al., 1998). At relatively alkaline pHs (found under stress), even very low ABA concentrations found in the xylem of well-watered plants (around 10 nM) will limit leaf growth rate. At acidic pHs, these concentrations are ineffective as the arriving ABA is partitioned to the symplast, presumably away from sites of action for leaf growth. Large amounts of ABA will also be loaded into the alkaline phloem cells in the leaf and transported away from the leaf. This process can be restricted under stress.

Fig. 2. A suction experiment with an isolated 1st internode of runner bean. After 40 min 100 nM (+)ABA was added to the external buffer and the ABA content of the perfused buffer was analysed for ABA.

Fig. 3. Diagram of a leaf section demonstrating ABA flows from the xylem to the guard (= target) cells. Under well-watered conditions (pH 6.3; right hand side) ABA is redistributed to the mesophyll and the epidermis by anion trapping. In a more alkaline xylem sap (left hand side) ABA remains high, mesophyll ABA may even be released to the xylem. (Figure modified from Taiz and Zeiger, 2000.)
where the apoplast becomes more alkaline. This process will also result in the export of previously trapped ABA from the organelles of the leaf to the apoplast. This hormone may contribute to the control of stomatal behaviour and leaf growth, but modelling suggests that this is only after the hormone has been loaded into the phloem and transported to the roots from where it is returned to the shoots in the xylem (see Section 9).

**ABA in fruits (7)**

It is well known that ABA can accumulate in fruits of plants in drying soil, again often because these are relatively alkaline compartments. Such measurements of accumulation are often made at the end of an extended drought period while more dynamic estimates of hormonal accumulations in fruit are not common in the literature. Davies et al. have suggested that the accumulation of ABA in the epidermis of growing tomato fruit may be restricted under mild soil drying because of limited water flux between the developing fruit and the vegetative part of the plant (Davies et al., 2000). Tomato fruits and grapes are two fruits which can show no shrinkage even when the water potential of the leaves is lower than that of the fruit. This argues for a very limited xylem connection between fruits and shoots. This may restrict fluxes of hormones into fruits with the result that mild soil drying can restrict shoot growth with little effect on fruit growth and development. There are now several examples of experiments where plants are subjected to partial drying of the roots and where this treatment can greatly restrict water use as a result of limited leaf growth and stomatal behaviour while fruits grow and develop without limitation. This increase in water use efficiency (fruit produced per unit of water used) is highly desirable in dryland agriculture and may simply be a result of differential deposition of chemical signals due to the hydraulic architecture of the plant.

**Meristems (8)**

The water relations of meristems of droughted plants are probably relatively well buffered against large variation as soil dries or as transpiration increases (Barlow, 1986). This is because meristems of broad-leaved plants lose relatively little water directly while meristematic regions of grass leaves are well protected inside the base of the grass tiller. Assessment of turgors in the leaf bases of growing grass leaves (using the micro-pressure probe) suggests that turgors can be maintained at or close to the values shown by well-watered plants, even when seedlings are subjected to quite low soil water potentials (Thomas et al., 1988). As meristem water potential falls, solutes accumulate (Munns et al., 1979) to maintain turgor and much of this occurs because leaf growth rates are reduced by soil drying (Kuang et al., 1990). Solute regulation in roots is less effective at turgor maintenance (Spollen et al., 1993), but water relations of growing cells are still buffered from severe dehydration.

If turgors in meristematic regions are sustained by restricted leaf growth as soil dries then it must be asked why growth is restricted. There is often a good relationship between ABA accumulation in meristems and restriction in leaf growth rate (Dodd and Davies, 1994) and even the apparent temperature sensitivity of the leaf growth response to applied ABA can be explained by differential accumulation of ABA in meristems at different temperatures. Results of this kind show how important it can be to assess the ABA contents of the growing cells themselves, if a case is to be made for growth regulation. In much older work, growth rates of grass leaves have been related to ABA accumulation in leaf laminae where cells are all fully expanded. Any relationship that is found between these two variables will have little bearing on the regulation of growth.

The effects of ABA on growth in meristems where the water relations are tightly regulated may contrast markedly with regulation in cells which dehydrate as water availability is reduced. Spollen et al. have noted that the influence of ABA on root growth can vary markedly with the water status of the root cells (Spollen et al., 1993). In root cells where turgor is not fully maintained by solute regulation, it is clearly unrealistic to assess the role of ABA in growth regulation by applying the hormone to roots with high turgors. Recent work (Sharp et al., 2000) has suggested that in shoots where ethylene synthesis rates are high, as they may be as soil dries and shoot water potentials decline, ABA may act to sustain shoot growth rates by suppressing ethylene synthesis. At high turgors in protected meristems, ABA may act more directly to restrict leaf growth rate.

**ABA recirculation (9)**

Increase of root tissue ABA and of ABA recirculation is not only a result of an enhanced ABA biosynthesis in roots. ABA that has been synthesized in leaves can be loaded to the phloem, transported to the roots where it can be deposited or recirculated to the xylem vessels. ABA recirculation has been detected in white lupins, in castor beans and in maize (Jeschke et al., 1997a, b; Wolf et al., 1990). Recirculation becomes more important under conditions of increased ABA phloem transport, particularly under salt stress (11), phosphate deficiency and ammonium nutrition (Jeschke et al., 1997a; Peuke et al., 1994).

It is important to stress that ABA recirculation is a quantitatively significant component of the root-to-shoot signalling process. In much writing there is an assumption
that ABA in the xylem stream is root-sourced and has been synthesized there in response to some form of root perturbation. This is clearly not the only contribution to the xylem signal and recirculated ABA will also play an important role. The extent of recirculation from the root will depend upon root water status and other variables. Presumably any stress which results in an alkalization of the apoplastic compartment will increase recirculation from the root and enhance the strength of the root signal.

Conclusion

Long-distance ABA signalling in droughted plants is often thought to be seductively simple: ABA synthesis in roots and transport to leaves is increased as a function of soil drying and shoot growth and physiology is modified as a function of this increased delivery. It should be apparent from the above that this is far from the case. There is now a good understanding of how the fluxes of ABA about the plant can be regulated, often as result of changes in the pH relations of different compartments of the leaf. There is also much that is still not known. There is little information on the link between a change in soil conditions and the generation of the ABA signal and the crucial changes in apoplastic hormone concentrations from indirect measurements of other variables can only be inferred. In addition to this there is comparatively little information on soil drought on the long-distance signalling via other chemicals, although evidence is accumulating on the interactions between ethylene and ABA (Hussain et al., 2000) and cytokinins and ABA (Stoll et al., 2000) in response to soil drying. What is now clear is that chemical signalling is an important component of the plant’s stress response and as such is a prime target for agronomic (Davies et al., 2000) or genetic manipulation. The target genes for manipulation may not be the apparently obvious ones such as the ABA synthesis genes, but rather genes that might influence the pHs of different compartments of the leaf.

References


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