Electrical Signals in Plants: Facts and Hypotheses

Chapter · January 2006

DOI: 10.1007/978-3-540-37843-3_17

1 author:

Eric Davies
North Carolina State University
138 PUBLICATIONS **4,010** CITATIONS

SEE PROFILE

17 Electrical Signals in Plants: Facts and Hypotheses

ERIC DAVIES

17.1 What is the context?

Electrical signals were first described over 200 years ago in both plants (Berthelon 1783) and animals (Galvani 1791), and had become an important line of study in plants over 140 years ago (Burdon-Sanderson 1873; Darwin 1875); thus they are far from being a novel phenomenon. Most of the earlier work (Burdon-Sanderson 1873; Darwin 1875), involved insect-trapping plants, which, like the sensitive plant, *Mimosa pudica*, have very rapid and visually striking responses to touch (reviewed in Simons 1981; Braam 2005) and were the preferred organisms for study for over a century. Until about 1880, there seemed to have been general acceptance of electrical signals as common to plants and animals; indeed, they were the only mechanism known for intercellular communication in any living system. However, shortly after his work on electrical signals, Darwin (1875) postulated the existence of chemical signals in plants (Darwin 1881), thus, plants were recognized as having both electrical and chemical signals, while animals had only electrical. Then, with the discovery of animal hormones and the emergence of the field of endocrinology at the turn of the (twentieth) century, it was accepted that both animals and plants had both types of signal.

Work on electrical signals in plants continued in the twentieth century, especially in India (e.g. Bose and Das, 1925), and to a lesser extent the USA (e.g. Pickard 1973), yet there seems to have been a paradigm shift, the existence of electrical signals, at least in "normal" plants was questioned. There are at least two reasons for this. First, almost concomitant with the publication of Pickard's (1973) review of action potentials in plants, there appeared a book (Tompkins and Bird 1973), aimed at the general public, which was based to a large extent on irreproducible results reported by an FBI lie detector expert (Backster 1968). This work caught the public's imagination (at least in the USA), but caused immense consternation among genuine plant scientists, since the entire field of plant electrophysiology was rendered suspect (Galston and Slayman 1979). Second, there was an underlying assumption that there was no real need for rapid signals in organisms as sluggish as

Botany Department, North Carolina State University, Raleigh NC 27695–7612, USA (e-mail: eric_davies@ncsu.edu)

Plant Electrophysiology – Theory & Methods (ed. by Volkov)

plants. Thus, research on electrical signals in plants hit an impasse, particularly in the USA, where funding effectively stopped for 2 decades. However (and despite this lack of funding), the field has opened up again over the last 25 years, and there is increasing consensus that electrical signals in plants do, indeed, exist, not only in those with rapid movements, but in all plants. This recognition has come about at least in part from the realization that "normal" plants can have very rapid systemic responses on fundamental processes such as gene expression (Davies and Schuster 1981a,b) and this requires the generation and transmission of even more rapid systemic signals (Davies 1987a,b). This recognition of the need for electrical signals in plants is also manifested by the very recent "First Symposium in Plant Neurobiology" (see: http//izmb.de/zellbio/volkmann/index.html) and the publication of a book with articles from that Symposium (Baluska et al. 2006) as well as the publication of this present textbook, the first ever devoted to plant electrophysiology. To summarize, the main "fact" emerging from the first section is that electrical signals do, indeed, occur in most, if not all organisms (including plants), not just those exhibiting rapid and visibly-obvious responses.

17.2 What are major definitions and types of signal?

For the purposes of this article, a stimulus is anything that evokes a response within the plant (termed "stimulus–response coupling" in the animal literature), while a signal is anything which is generated and transmitted by the plant in response to that stimulus. Thus the stimulus might be applied from outside the plant, while the signal *must* be generated within the plant. Further, the signal itself becomes a stimulus when it arrives at its destination and provokes its own response.

As an example to expound on this point (and references will be furnished later in the appropriate sections), when voltage is applied to a plant (electrical stimulation) it can evoke the generation of an electrical signal (action potential, or AP), which is transmitted through the plant. This traveling AP almost certainly involves calcium influx into the cytoplasm followed by chloride and potassium efflux. These sub-components of the AP signal, such as increased cytosolic calcium, become stimuli when they evoke responses further downstream, such as the activation of phospholipase C. Phospholipase C can be thought of a signal in the vicinity of the plasma membrane, where it becomes a stimulus and evokes the release of inositol phosphate metabolites. These metabolic signals (especially IP_3), can then act as stimuli to evoke release of more calcium from intracellular stores. Finally, these signals (IP_3, calcium and/or their associated protein phosphorylation events) can then act as stimuli to modulate gene expression (Davies and Stankovic 2006). This sequence of

$$\text{stimulus} \rightarrow \text{signal} = \text{stimulus} \rightarrow \text{signal} = \text{stimulus} \rightarrow \text{signal}$$

continues, and a major goal of research on electrical signals in plants is to decipher as many of the steps as possible.

The array of signals in plants for which the term "electrical" has been used includes: action potentials (AP), including spontaneous action potential (SAPS); variation potentials (VP), also called slow wave (SW; Stahlberg and Cosgrove 1996); voltage transients (VT) or voltage spikes (VS); and rhythmical electrical activity (REA). By definition, all of these electrical activities must involve changes in membrane potential (MP). However, this review will be restricted to those changes in MP which are transmitted long distances and which have known stimulus and response. The major reason behind this is that any flux of ions across the plasma membrane will elicit some change in MP, and since all cells (especially root cells involved in ion uptake) have ions traversing the plasma membrane virtually all the time, then almost all of metabolism would be linked to "electrical" signals.

Here, we define "signal" as having some aspect of transmission, and, depending on the distance transmitted, it might be systemic, i.e. throughout the plant (thus, by definition, long distance), intercellular, or intracellular. Here we will focus our attention on the two major long-distance signals, AP and VP, especially in "normal" vascular plants, i.e. those without obvious rapid visible manifestations, and limited consideration will be given to plants with rapid movements (Braam 2005), and to non-vascular plants, especially algae with very large cells and with which most of the early intracellular recordings were made (Wayne 1994). For more information on giant algal cells, I suggest reading the chapters in this text by Bisson et al. (this volume) and Shimmen (this volume). This review will also not deal with local changes in MP with little or no transmission, such as REA (Mitsumo and Sibaoka 1989; Antkowiak et al. 1991; Davies et al. 1991), or VT (Pickard 1984a,b; Krol and Trebacz 1999). Nor will it deal with changes in MP induced by symbionts (Assmann 1995; Felle et al. 1995), pathogens (Kurusu et al. 2005; Pike et al. 2005), insects (Volkov and Haack 1995; Maffei et al. 2004); gravity, or those involved in tip growth (Harold and Caldwell 1990). The role of biotic agents will be dealt with in this book by Maffei and Bossi (2006), gravity by Stankovic (2006) and in roots by Takamura (2006). This review will also not discuss the so-called "spontaneous" action potentials (SAPS), since they have no known stimulus or consequence (Zawadzki et al. 1995).

17.3 What are action potentials?

The AP is the only long distance signal that can be considered to be a "genuine" electrical signal (reviewed by Pickard 1973; Gradmann and Mummert 1980; Simons 1981; Wayne 1994; Ksenzhek and Volkov 1998; Davies 2004) The fundamental properties of a "pure" AP are that it be self-propagating,

all-or-nothing, and transmitted at essentially constant velocity and amplitude (Zawadzki et al. 1991; Dziubinska 2003). Since they are self-propagating, AP must depend on ion channels that respond (open) to changes in MP, i.e. voltage-gated channels (VGC), thus any cell containing voltage-gated channels can generate and transmit an AP. The primary VGC seems to be a plasma membrane-associated calcium channel that causes a sharp increase in the normally very low cytosolic calcium levels (Tester 1990; Wayne 1994, Fisahn et al. 2004). It has, however, been suggested that the increased cytosolic Ca^{2+} does not come through the plasma membrane, but is released from internal stores, perhaps triggered by IP3 (Plieth et al. 1998; Biskup et al. 1999; Wacke and Thiel 2001), but this view has been challenged recently (Tazawa and Kikuyama 2003). Nevertheless, there does seem to be cross-talk between PIP derivatives and K^+ channels (Liu et al. 2005) and the K^+ channel does appear to be a voltage sensing component of the membrane, at least in animal tissues (MacKimmon 2004; Horn 2005). In either case, the increase in calcium seems to trigger opening of a calcium-dependent chloride channel, which allows chloride efflux into the apoplast, and then the original (resting) MP is restored by an outward rectifying potassium channel. This sequence of ion fluxes has been described for giant algal cells (Wayne 1994; Shimmen 1997), a liverwort (Trebacz et al. 1994), the sensitive plant Mimosa (Samejima and Sibaoka 1980), and even trees such as willow (Fromm and Spanswick 1993), reviewed by Tester (1990). The transient change in MP causes the same sequence of events (Ca^{2+} influx, Cl^- efflux and K^+ efflux) to be repeated along the series of cells in the pathway.

In vascular plants, the primary location for long distance AP transmission is the phloem, especially the sieve tubes (Eschrich et al. 1988; Fromm and Spanswick 1993; Dziubinska et al. 2001; Dziubinska 2003), but there is no reason why lateral spread cannot occur through other cell types that contain VGC. In this case, all cells with symplastic connection to the phloem are likely to be "informed" by, and respond to, the passage of an AP. Indeed, in non-vascular plants such as algae (Wayne 1994) and liverworts (Trebacz et al. 1994). The AP must, by definition, be transmitted though non-phloem tissues.

The AP in plants seems to be very similar to the AP in animal cells such as heart, epithelium, etc, differing primarily in their rapidity (Sussman 1992), but they are very different from the AP in neurons, which rely on Na^+/K^+ exchange (Davies 1987b). The AP is made possible by the fact that the resting MP of plant cells is very negative (approximately 200 mV) to the inside. Calcium influx causes depolarization (less negative inside), this is enhanced by chloride efflux, but then neutralized by potassium efflux. Once an AP has passed, there is a period of delay (the refractory period) during which another AP cannot be generated or transmitted. This is most likely the result of temporary inactivation of one or other of the ion channels, most likely the calcium VGC.

17.4 What are variation potentials?

Unlike the AP which is a self-propagating, transmitted change in MP, the VP is a local change in MP resulting from the passage of some other signal (Malone 1994, 1996; Malone et al. 1994a,b; Stankovic et al. 1998; Mancuso 1999). Nevertheless, the local change in MP must again be the result of ion fluxes across the plasma membrane, but not through VGC, otherwise this would generate an AP. Further, long distance AP transmission occurs primarily in the phloem, primarily in the living but enucleate sieve tubes, whereas the VP is transmitted in the dead xylem (Davies et al. 1991; Malone 1994, 1996; Mancuso 1999).

There are two main candidates for the xylem-transmitted component: changes in pressure/tension and transport of chemicals. Both types of primary signal most likely exist and evidence for both of them is good. Since VP must depend on ion channels (but not VGC), the former explanation (pressure/tension changes) must be based on mechanosensitive channels (MSC), whereas the latter (chemicals in the xylem) must be based on ligand-activated channels (LAC). The evidence for rapid loss of tension in the xylem comes primarily from work using excision, heat wound and/or a pressure bomb as stimuli and employing electrodes, position sensing transducers and an analytical balance to assess changes in MP, tissue deformation and water uptake, respectively (Roblin and Bonnemain 1985; Davies et al. 1991; Stahlberg and Cosgrove 1995, 1996; Stankovic et al. 1997, 1998; Stankovic and Davies 1997b; Stahlberg et al. 2005). When one leaf is heat-wounded there is an essentially instantaneous relaxation of the entire stem, followed by changes in MP which appear both smaller and more delayed with increasing distance from the point of wounding (Davies et al. 1991; Stankovic et al. 1997; Stankovic and Davies 1997b; Mancuso 1999) These changes in MP and stem relaxation can be mimicked by transient or prolonged application of slight pressure to one leaf (Stankovic et al. 1997). Similar changes were also seen in wheat leaves (Malone and Stankovic 1991), who at that time evoked a mechanosensory explanation. More recently, Malone and co-workers have come to favor a role for chemicals transmitted in the xylem (Malone et al. 1994a; Malone 1996), which, by definition would have to involve LAC.

17.5 What stimuli evoke AP and VP and what are the consequences?

The brief answer to the first question is “non-damaging stimuli evoke AP, while damaging stimuli evoke VP” (Dziubinska 2003). Non-damaging stimuli that evoke AP include electrical stimulation, light/dark transitions (Trebacz and Zawadzki 1985), brief cooling (Pyatygin et al. 1992) and pollination

(Fromm et al. 1995), while damaging stimuli that evoke VP include severe wounding (reviewed in Dziubinska 2003). However, the distinction is not too clear. Sometimes excision results in AP (Pickard and Davies, unpublished results), while in the same organism excision is known to evoke a VP/SW (Stahlberg and Cosgrove 1995). The major consequences of electrical signals are provided in the chapter of this textbook (Fromm, this volume). Here I will briefly discuss the rapid responses, since it is the rapidity of response that necessitates a rapidly-generated and transmitted signal. These initial responses will be evoked by any of the primary events occurring as sub-components of the AP (or VP) including Ca^{2+} influx, Cl^- efflux, K^+ efflux, and the concomitant change in MP. As discussed earlier, these include changes in enzyme activity, gene expression, and in the status of the cytoskeleton (Davies 1987a,b, 1993).

Here I will emphasize responses at the level of gene expression, primarily transcription, but also translation, since it was the incredibly rapid changes in polyribosome formation and protein synthesizing capacity (Davies and Schuster 1981a) that led us to conclude that ultra-rapid signals must be involved (Davies and Schuster 1981b), and this led us to examine the potential role of electrical signals in systemic wound responses. We have focused our efforts on the putative role of elevated cytoplasmic Ca^{2+}, the channels through which it enters, their association with the cytoskeleton, and the effects of Ca^{2+} on modulating enzyme activity via phosphorylation. We showed early on that the *increase* in polyribosomes was accompanied by a paradoxical *reduction* in protein synthesis as well as a concomitant cessation of cytoplasmic streaming (Davies 1990). This reduction in protein synthesis along with formation of polysomes could be explained by phosphorylation of elongation factor, which would cause ribosomes to pile-up on mRNA (Davies 1993). Similarly, cessation of cytoplasmic streaming could result from phosphorylation of myosin and the simultaneous effect on both processes could be explained if polysomes were attached to the cytoskeleton (Davies et al. 1998; Davies and Stankovic 2006).

In addition to affecting translation, both kinds of electrical signal modify transcription. Both electrical stimulation (AP) and heat-wounding (VP), evoke systemic accumulation of protease inhibitor transcripts, but only flame-wounding (VP) induces calmodulin transcripts (Stankovic and Davies 1996, 1997a,b). The VP also causes accumulation of transcripts encoding a chloroplast mRNA binding protein (Vian et al. 1999), a leucine zipper transcription factor, bZIP (Stankovic et al. 2000), a vacuolar ATPase (Coker et al. 2003) and many others (Coker et al. 2005), including some that peak within 2 min in a leaf 5 cm distant from the wound site (Davies et al. 1997; Davies 2004). Again, Ca^{2+}-induced phosphorylation of a specific protein, in this case of RNA polymerase 2, could explain this very rapid systemic transcript accumulation (Davies and Stankovic 2006).

We have tried to understand how the different electrical signals might evoke the accumulation of different transcripts and our explanation is summarized in Fig. 17.1 and the pertinent references are cited in Davies (1993) and

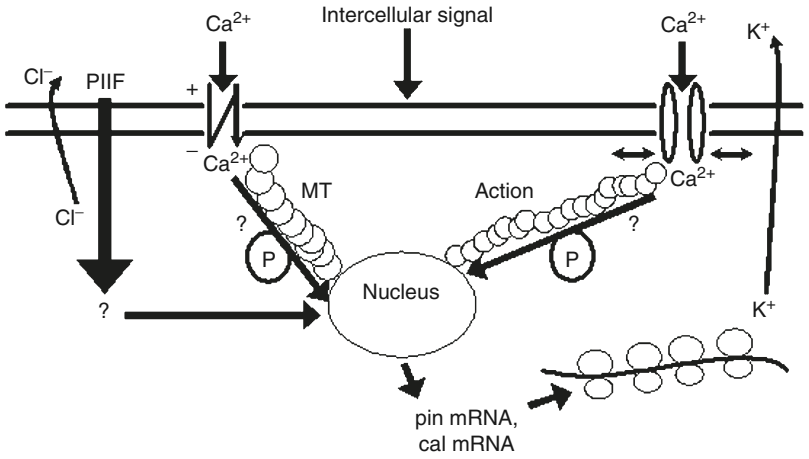

Fig. 17.1. Transduction of intercellular signals into cellular responses involving gene expression in tomato. In the case of an AP, voltage-gated channels associated with microtubules (MT) will allow calcium entry, while in the case of a VP, mechano-sensing channels associated with microfilaments will allow entry. Since the channels are associated with different cytoskeletal elements, are located in different parts of the cell, and are open for different durations, the downstream effects of calcium will be quite distinct. The encircled P denotes a phosphorylation wave, caused by cytoskeleton-associated protein kinases, and transmitted along the microtubules and microfilaments. With microfilaments, in particular (involved in the heat-wound-induced VP), this wave will continue to the nucleus where RNA synthesis will be enhanced and transcript accumulation will occur. This is because actin in the nucleus associates with RNA polymerase 2 which becomes activated on phosphorylation. These newly-synthesized transcripts might not be translated, either because potassium leaves the cell, lowering it to sub-optimal levels for translation, or because cytoskeleton-associated polysomes become non-functional because of phosphorylation (inactivation) of elongation factor. Finally, chloride passing into the cell wall might activate cell wall enzymes, including those that liberate PIIF (protease-inhibitor inducing factor). References in support of these events are elsewhere (Davies and Schuster 1981a,b; Davies 1987a,b, 1990, 1993, 2004; Davies et al. 1998; and especially Davies and Stankovic 2006). Figure from Stankovic and Davies (unpublished)

Davies and Stankovic (2006). Briefly, the hypothesis suggests that Ca^{2+} entering through a mechanosensitive channel (VP) will encounter Ca^{2+}-binding proteins, such as calcium-dependent protein kinases, associated with microfilaments, In the cytoplasm, these will phosphorylate myosin and thus inhibit cytoplasmic streaming, and elongation factor and thus inhibit ribosome movement on cytoskeleton-bound polysomes, while in the nucleus, phosphorylation of RNA polymerase 2 will enhance transcription. These phosphorylation responses can be extremely rapid (Bogre et al. 1996). In contrast, Ca^{2+} entering a voltage gated channel (AP) will encounter Ca^{2+}-binding proteins on the microtubules also possibly leading to enhanced transcription (Mori et al. 2004). Interestingly, in terms of changes in transcript accumulation, the transmitting tissue, although "informed", is unable to respond, thus making the

system more nerve-like. This is obvious with the VP, since it is transmitted in dead xylem cells, which are incapable of any metabolic response. However, it must also be true of the electrically stimulated AP, since it is transmitted in sieve tubes which have no nuclei, thus cannot synthesize RNA.

17.6 How do you measure AP and VP?

To measure any change in MP, some kind of electrode in contact with the plant must be attached to some kind of recording device. Since the electrical changes are small, the signals usually have to be amplified and the recording device must have high impedance, otherwise the electrical output from the plant is used to drive the recording device and the signal "disappears". The simplest electrodes to use are extracellular ones, which can either be surface contact electrodes or wire (piercing) electrodes. Surface contact electrodes (akin to the lie detector electrodes used by Backster 1968) have the advantage of not inflicting tissue damage, which is important when studying wound responses. However, since these electrodes contain KCl and they tend to dry out, this changes the ionic status of the region being measured, so these electrodes can be used for only short-term (a few hours) recordings. Piercing electrodes (silver, platinum) have the disadvantage of inflicting damage, but this problem can be circumvented by allowing the tissue to recover from the wound. They have the advantage of being usable for days and perhaps weeks and they do not appreciably alter the ionic status of the tissue (Zawadzki et al. 1995). Both electrodes measure the apoplastic ion concentration in the region of the electrode, and we have found essentially identical recordings with both types of electrodes placed close together on the same plant.

In order to take measurements, however, there must be a complete circuit, and so another electrode is needed. This may be a genuine ground electrode (i.e. in the soil), or a reference electrode at some other part of the plant. With a ground electrode, one measures the change in MP at the measuring electrode compared with (presumably) no change in the ground electrode, while with a reference electrode at some other point on the plant one measures the difference in MP between what is happening at the measuring electrode compared with what is happening at the reference. This is clearly seen in Fig. 17.2, where the signal going through successive measuring electrodes is upwards, but this gains the opposite sign when passing through the reference.

A more difficult type of electrode to use is a microelectrode (see Shabala 2006, this volume for details). The microelectrode must be inserted (with great care) into the cytoplasm of an individual cell making sure it is not in the much more voluminous vacuole. The reference electrode is put in the bathing medium, and so this method measures the actual MP of that individual cell, as well as any change in MP that results from an electrical signal. These electrodes were first used in giant algal cells even before they were used in animal cells

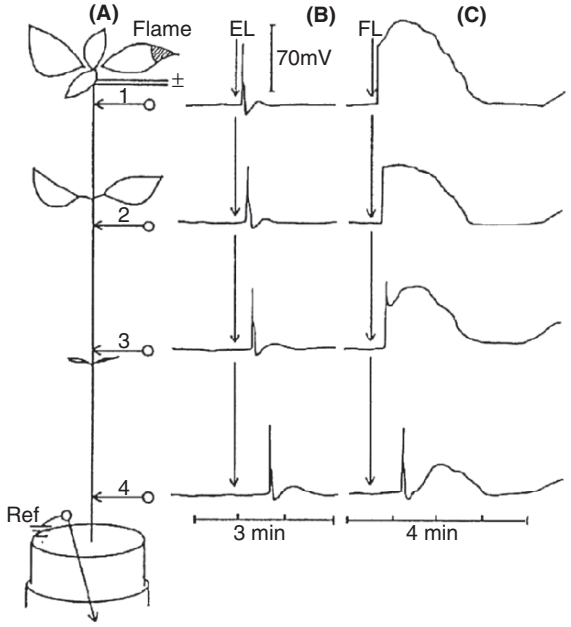

Fig. 17.2. Typical action potentials and variation potentials measured in sunflower. The plant diagrammed to the left (**A**) was stimulated electrically (5 V for 1 s) at a point about 5 cm below the lowest petiole (+/–) or heat wounded with a gentle flame applied to the tip of a leaf (*W*). Measuring electrodes (inserted silver wires) were placed along the stem, and a reference electrode was placed in the pot. The electrical responses to electrical stimulus are action potential that are shown in the middle (**B**). The electrical responses to the heat wounding stimulus are variation potentials that are shown to the right (**C**). From Davies et al. (1991), with permission. Note: electrical stimulation evoked a pure AP, while flame-wounding evoked a combined AP/VP, with the former "traveling" faster than the latter

(Wayne 1994). The main limitation of this method for vascular plants is that it is generally restricted to large, accessible cells, either epidermal or subepidermal, or individual cells such as root hairs or pollen tubes (Tester 1990). In such instances the tissue, organ, or intact plant has to be in the bathing medium, thus preventing their measurement in any but small seedlings. More recently, several workers have taken advantage of an aphid's ability to probe phloem sieve tubes. Generally, the aphid is allowed to penetrate the phloem sieve tube, its head is severed, leaving its mouthparts in the phloem, and then electrodes are (very carefully) inserted into the aphid's mouthparts giving the microelectrode direct access to the sieve tube (Koziolek et al. 2003) This permits the measurement of MP in phloem sieve tube cells of essentially any plant (attacked by aphids) growing under any condition, including trees such as poplar (Lautner et al. 2005). This technique has given unequivocal evidence

that the AP does travel in the phloem sieve tube, but has not ruled out its transmission in other tissues. Yet another electrode that can measure changes in MP is the vibrating probe electrode (see Feijo 2006, this volume for details), but it, too, needs to be in a bathing medium. Both of these techniques have the additional advantage of being able to measure specific ions, not just MP, thus the specific ion fluxes involved in AP and VP can be identified (in the accessible cell types).

The signal emerging from the electrode system needs to be converted into a visible representation, and for many years this was done using a chart recorder. These were satisfactory for most purposes, except they would suffer from baseline drift, an overly large signal, or some other factor that would cause the pen to go off-scale. Furthermore, rapid chart speeds could not be used or else entire rolls of chart paper would be consumed in minutes. In the past 15 years, there has been increasing use of computers especially equipped with data acquisition cards (analog/digital converters) and data manipulating programs such as LabView. Such programs store all the data, which can then be visualized (and printed) with the appropriate time intervals and voltage parameters. Furthermore, the number electrodes that can be used is limited only by computer memory rather than by the number of channels on the recorder(s). Recently, additional kinds of electrical activity in plants have been identified as result of taking exceedingly rapid (frequent) data sampling (Volkov 2000; Pickard 2001; Volkov et al. 2005).

17.7 How do you differentiate between AP and VP?

The most convenient way to differentiate between these signals is by their shape, velocity and constancy. The AP consists normally of a sharp rise, a brief peak, and sharp return to near baseline, whereas the VP (as its name suggests) is far more variable, normally appearing as a sharp rise followed by a lingering decline, often with spikes (AP?) superimposed and/or interspersed. In addition, the AP stays essentially the same as it passes through the plant, while the VP gets smaller and "slower". "Slower" is put in quotes, since it is not the VP itself that travels, but either a loss of tension (hydraulic signal) or perhaps chemicals in the xylem. In analogy to identification of "ducks" analogy, "If it looks like an AP, moves like an AP, and keeps going like an AP, it is most likely an AP".

There are reports in the literature (e.g. Wildon et al. 1992) claiming that flame-wounding evokes AP, even though the signals had shapes typical of VP, and a series of electrodes was not used to determine whether the signal diminished with passage. More recently others (Kozoliek et al. 2003; Lautner et al. 2005) have shown that flame-wounding evokes VP-like signals, but based on other evidence (cold blockage of signal) they called them AP. The situation is made very complex because a VP (resulting from a

hydraulic signal transmitted in the xylem) can evoke an AP in the phloem. Cold blocks would remove the phloem component, but not the xylem component. However, since the VP is fundamentally different from the AP it should be possible to differentiate between them quite clearly. This is because the VP results (usually) from a loss of tension in the xylem and, by using position sensing transducers (PST), this change in tension can be measured indirectly by changes in tissue deformation (contraction/relaxation) throughout the plant (Davies et al. 1991; Stahlberg and Cosgrove 1995; Mancuso 1999).

In our hands, these changes in stem length and diameter *always* precede changes in MP and are accompanied by almost immediate cessation of water uptake (Davies et al. 1991; Stankovic et al. 1998), whereas an AP is *never* preceded by tissue deformation (although it may be followed by very small fluctuations in stem length/diameter). To clearly distinguish between the AP and VP, experiments employing a combination of several electrodes in a row, PSTs, and balances (to measure water uptake), in conjunction with stripping the bark, phloem-girdling and/or applying a cold block (to prevent phloem AP), and/or under high humidity (to presumably reduce the VP) would be needed. I am unaware of any such "complete" reports, but such a study would be a welcome addition to the literature. The experiments will, however, need to be done carefully, since there is cross-talk between the VP and AP, insofar as any change in MP, including that evoked by a VP, can evoke an AP. Indeed, pressure, presumably detected by MSC and normally evoking a VP can be transduced to elicit an AP (Shimmen, this volume).

17.8 Why do plants have electrical signals and why are there two types?

The fundamental reason why plants have electrical signals is that they permit very rapid systemic information transmission, so that the entire plant is informed almost instantly even though only one region may have been perturbed. This is especially important for detection and response to danger, which is detected at one location, but where the entire plant needs to be warned so it can mount an essentially immediate response. Such an ultrarapid response is of particular importance with viruses, which can enter a wound and then launch a systemic invasion very rapidly, annihilating the plant unless it mounts an effectively rapid counter-attack. Not only do responses to putative viral attack need to be rapid and systemic, they also need to be highly coordinated, so that the appropriate cells exhibit the appropriate response. As described recently (Davies and Stankovic 2006), the wounded cells send a signal to both adjacent and distant cells, which respond by stopping cytoplasmic streaming, thereby slowing down intracellular flow and virus movement, clog plasmodesmata, thereby stopping cell-to-cell

transmission, inhibit translation elongation/termination, preventing ribosome movement along mRNA and translation of viral mRNA, and also by activating genes that can help in the defense response. Obviously, some of these responses cannot occur in the signal transmitting cells, since these are either dead (xylem) or lack nuclei and major cytoskeleton elements (sieve tubes). In contrast to these defense responses, insectivorous plants employ a pre-emptive strategy by making the first strike, although rapidity and coordinated activity of several cells are again essential, this time to prevent escape of the prey.

One plausible explanation for why there are two different electrical signals is that they provide back-up for each other, furnishing at least some overlap in the responses they elicit. Indeed, there may be cross-talk between the pressure-sensing VP and the voltage-sensing AP (Shimmen, this volume), especially if plants possess force/voltage sensors as have been described in prokaryotes (Bezanilla and Perozo 2002). Another reason could be that the two signals also evoke different responses, so that their effects can be additive and this might be why heat-wounding often evokes a combination of VP and AP (Stankovic et al. 1998). Another explanation is that since the VP is transmitted via the xylem, it will be more effective during the day, especially under conditions of high transpiration, whereas (at least in our hands with sunflower and tomato), the electrically-induced AP is more easily evoked at night. It is to be expected that any highly successful biological entity (and plants are undoubtedly that) will have developed back-up systems to cope with an immense diversity of threats and opportunities.

References

Antkowiak B, Mayer WE, Engelmann W (1991) Oscillations in the membrane potential of pulvinar motor cells in situ in relation to leaflet movements of *Desmodium motorium*. J Exp Bot 42:901–910

Assmann S (1995) Electrifying symbiosis. Proc Natl Acad Sci USA 92:1795–1796

Backster C (1968) Evidence of a primary perception in plant life. Int J Parapsychol 10:329–348

Baluska F, Mancuso S, Volkmann D (2006) Communications in plants. Neuronal aspects of plant life. Springer, Berlin Heidelberg New York

Bertholon ML (1783) De l'Electricité des Végétaux, Alyon, Paris

Bezanilla F, Perozo E (2002) Force and voltage sensors in one structure. Science 298:1562–1563

Biskup B, Gradmann D, Thiel G (1999) Calcium release from InsP3-sensitive internal stores initiates action potential in Chara. FEBS Lett 453:72–76

Bogre L, Ligterink W, Heberle-Bors E, Hirt H (1996) Mechanosensors in plants. Nature 383:489–490

Bose JC, Das GP (1925) Physiological and anatomical investigations on *Mimosa pudica*. Proc R Soc B 98:290–312

Braam J (2005) In touch: plant responses to mechanical stimuli. New Phytol 165:373–389

Burdon-Sanderson J (1873) Note on the electrical phenomena which accompany stimulation of a leaf of *Dionaea muscipula*. Trans R Soc Lond 21:495–496

Coker JS, Jones DA, Davies E (2003) Identification, conservation, and relative expression of V-ATPase cDNAs in tomato plants. Plant Mol Biol Rep 21:145–158
Coker JS, Vian A, Davies E (2005) Identification, accumulation, and functional prediction of novel tomato transcripts systemically up-regulated after flame-wounding. Physiol Plant 124:311–322
Darwin C (1875) Insectivorous plants. Murray, London
Darwin C (1881) The power of movements in plants. Murray, London
Davies E (1987a) Wound responses in plants. Biochem Plants 12:243–264
Davies E (1987b) Action potentials as multifunctional signals in plants: a hypothesis attempting to unify apparently disparate wound responses. Plant Cell Environ 10:623–631
Davies E (1990) Plant wound signals and translation. Proceedings of the 13th international congress on plant growth substances, pp 519–530
Davies E (1993) Intercellular and intracellular signals in plants and their transduction via the membrane–cytoskeleton interface. Semin Cell Biol 4:139–147
Davies E (2004) Commentary: new functions for electrical signals in plants. New Phytol 161:607–610
Davies E, Schuster A (1981a) Intercellular communication in plants: Evidence for a rapidly-generated, bidirectionally-transmitted wound signal. Proc Natl Acad Sci USA 78:2422–2426
Davies E, Schuster A (1981b) Wounding, action potentials and polysome formation. Plant Physiol 67:538
Davies E, Stankovic B (2006) Electrical signals, the cytoskeleton, and gene expression: a hypothesis on the coherence of the cellular responses to environmental insult. In: Baluska F, Mancuso S, Volkmann D (eds) Communication in plants. Neuronal aspects of plant life. Springer, Berlin Heidelberg New York
Davies E, Zawadzki T, Witters D (1991) Electrical activity and signal transmission in plants: how do plants know? In: Penel C, Greppin H (eds) Plant signaling, plasma membrane and change of state, University of Geneva Press, Geneva, Switzerland, pp 119–137
Davies E, Vian A, Vian C, Stankovic B (1997) Rapid systemic up-regulation of genes after heat-wounding and electrical stimulation. Acta Physiol Plant 19:571–576
Davies E, Abe S, Larkins BA, Clore AM, Quatrano RS, Weidner S (1998) The role of the cytoskeleton in plant protein synthesis. In: Bailey-Serres J, Gallie DR (eds) A look beyond transcription: mechanisms determining mRNA stability and translation in plants. ASPP, pp 115–124, Rockville, MD, USA
Dziubinska H (2003) Ways of signal transmission and physiological role of electrical potentials in plants. Acta Soc Bot Pol 72:309–318
Dziubinska H, Trebacz K, Zawadzki T (2001) Transmission route for action potentials and variation potentials in Helianthus annuus. L. J Plant Physiol 158:1167–1172
Eschrich W, Fromm J, Evert RF (1988) Transmission of electric signals in sieve tubes of zucchini plants. Bot Acta 101:327–331
Felle HH, Kondorosi E, Kondorosi A, Schultze M (1995) Nod signal-induced plasma membrane potential changes are differentially sensitive to structural modification of the lipochitooligosacchraride. Plant J 7:939–947
Fisahn J, Herde O, Willmitzer L, Pena-Cortes H (2004) Analysis of the transient increase in cytosolic Ca2+ during the action potential of higher plants with high temporal resolution: requirement of Ca2+ transients for induction of jasmonic acd biosynthesis and PINII gene expression. Plant Cell Physiol 45:456–459
Fromm J, Spanswick R (1993) Characteristics of action potentials in willow (*Salix viminalis* L.). J Exp Bot 264:1119–1125
Fromm J, Hajirezaei M, Wilke I (1995) The biochemical response of electrical signaling in the reproductive system of Hibiscus plants. Plant Phys 109:375–384
Galston AW, Slayman CL (1979) The not-so-secret life of plants. Bioscience 29:337–344
Galvani L (1791) De viribus electricitatis in motu musculari commentarius. Bononiae Instituti Scientiarum, Bologna

Gradmann D, Mummert H (1980) Plant action potentials. In: Spanswick RM, Lucas WJ, Dainty J (eds) Membrane transport: current conceptual issues. Elsevier/North-Holland Press, Amsterdam, pp 333–344

Harold FM, Caldwell JD (1990) Tips and currents: electrobiology of apical growth. In: Heath IB (ed) Tip growth in plant and fungal cells. Academic Press, San Diego, pp 59–89

Horn R (2005) How ions channels sense membrane potential. Proc Natl Acad Sci USA 102:4929–4930

Koziolek C, Grams TEE, Schreiber U, Matyssek R, Fromm J (2003) Transient knockout of photosynthesis mediated by electrical signals. New Phytol 161:715–722

Krol E, Trebacz K (1999) Calcium-dependent voltage transients evoked by illumination in the liverwort *Conocephalum conicum*. Plant Cell Physiol 40:17–24

Ksenzhek OS, Volkov AG (1998) Plant energetics. Academic Press, San Diego

Kurusu T, Yagala T, Miyao A, Hirochika H, Kuchitsu K (2005) Identification of a putative voltage-gated Ca^{2+} channel as a key regulator of elicitor-induced hypersensitive cell death and mitogen-activated protein kinase activation in rice. Plant J 42:798–809

Lautner S, Grams TEE, Matyssek R, Fromm J (2005) Characteristics of electrical signals in poplar and responses in photosynthesis. Plant Physiol 138:2200–2209

Liu K, Li L, Luan S (2005) An essential function of phosphatylinositol phosphates in activation of plant shaker-type K^+ channels. Plant J 42:433–443

MacKimmon R (2004) Voltage sensor meets lipid membrane. Science 306:1303–1305

Maffei M, Bossi S, Spiteller D, Mithofer A, Boland W (2004) Effects of feeding *Spodoptera littoralis* on lima bean leaves. I. Membrane potentials, intercellular calcium variations, oral secretions, and regurgitate components. Plant Physiol 134:1752–1762

Malone M (1994) Wound-induced hydraulic signals and stimulus perception in *Mimosa pudica* L. New Phytol 128:49–56

Malone M (1996) Rapid, long-distance signal transmission in higher plants. Adv Bot Res 22:163–228

Malone M, Stankovic B (1991) Surface potentials and hydraulic signals in wheat leaves following localized wounding by heat. Plant Cell Environ 14:431–436

Malone M, Alarcon J-L, Palumbo L (1994) A hydraulic interpretation of rapid, long-distance wound signalling in the tomato. Planta 193:181–185

Malone M, Palumbo L, Boari F, Monteleone M, Jones HG (1994) The relationship between wound-induced proteinase inhibitors and hydraulic signals in tomato seedlings. Plant Cell Environ 17:81–87

Mancuso S (1999) Hydraulic and electrical transmission of wound-induced signals in *Vitis vinifera*. Aust J Plant Physiol 26:55–61

Mitsumo T, Sibaoka T (1989) Rhythmic electrical potential change of motor pulvinus in lateral leaflet of *Codariocalyx motorius*. Plant Cell Physiol 30:1123–1127

Mori MX, Erickson MG, Yue DT (2004) Functional stoichiometry and local enrichment of calmodulin interacting with Ca^{2+} channels. Science 304:432–435

Pickard B (1973) Action potentials in higher plants. Botanical Reviews 39:172–201

Pickard B (1984a) Voltage transients elicited by sudden step-up of auxin. Plant Cell Environ 7:171–178

Pickard B (1984b) Voltage transients elicited by brief chilling. Plant Cell Environ 7:679–681

Pickard WF (2001) A novel class of fast electrical events recorded by electrodes implanted in tomato shoots. Aust J Plant Physiol 28:121–129

Pike SM, Zhang X-C, Gassmann W (2005) Electrophysiological characterization of the *Arabidopsis avrRpt2*-specific hypersensitive response in the absence of other bacterial signals. Plant Physiol 138:1009–1017

Plieth C, Sattelmacher B, Hansen U-P, Thiel G (1998) The action potential in Chara: Ca^{2+} released from internal stores visualized by Mn^{2+}-induced quenching of fura-dextran. Plant J 13:167–175

Pyatygin SS, Opritov VA, Khudyakhov VA (1992) Subthreshold changes in excitable membranes of *Cucurbita pepo* L stem cells during cooling-induced action potential generation. Planta 186:161–165

Roblin G, Bonnemain J-L (1985) Propagation in *Vicia faba* stem of a potential variation induced by wounding. Plant Cell Physiol. 26:1273–1283

Samejima M, Sibaoka T (1980) Changes in extracellular ion concentration in the main pulvinus of *Mimosa pudica* during rapid movement and recovery. Plant Cell Physiol 21:467–479

Shimmen T (1997) Studies on mechano-perception in Characeae: effects of external Ca^{2+} and Cl^{-}. Plant Cell Physiol 38:691–699

Simons PJ (1981) The role of electricity in plant movements. New Phytol 87:11–37

Stahlberg R, Cosgrove DJ (1995) Comparison of electric and growth responses to excision in cucumber and pea seedlings. II. Long distance effects are caused by the release of xylem pressure. Plant Cell Environ 18:33–41

Stahlberg R, Cosgrove DJ (1996) Induction and ionic basis of slow wave potentials in seedlings of *Pisum sativum* L. Planta 200:416–425

Stahlberg R, Cleland RE, van Volkenburgh E (2005) Decrement and amplification of slow wave potentials during their propagation in *Helianthus annuus* L. shoots. Planta 220:550–558

Stankovic B, Davies E (1996) Both action potentials and variation potentials induce proteinase inhibitor gene expression in tomato. FEBS Lett 390:275–279

Stankovic B, Davies E (1997a) Intercellular communication in plants: electrical stimulation of proteinase inhibitor gene expression in tomato. Planta 202:402–406

Stankovic B, Davies E (1997b) Wounding evokes rapid changes in tissue deformation, electrical potential, transcription, and translation in tomato. Plant Cell Physiol 39:268–274

Stankovic B, Zawadzki T, Davies E (1997) Characterization of the variation potential in sunflower. Plant Physiol 115:1083–1088

Stankovic B, Witters DL, Zawadzki T, Davies E (1998) Action potentials and variation potentials in sunflower: an analysis of their relationships and distinguishing characteristics. Physiol Plant 105:51–58

Stankovic B, Vian A, Henry-Vian C, Davies E (2000) Molecular cloning and characterization of a tomato cDNA encoding a systemically wound-inducible bZIP DNA-binding protein. Planta 212:60–66

Sussman MR (1992) Shaking *Arabidopsis thaliana.* Science 256:619

Tazawa M, Kikuyama M (2003) Is Ca^{2+} release from internal stores involved in membrane excitation in characean cells. Plant Cell Physiol 44:518–526

Tester M (1990) Plant ion channels: whole cell and single channel studies. New Phytol 114:305–340

Tompkins P, Bird C (1973) The secret life of plants. Harper and Row, New York

Trebacz K, Zawadzki T (1985) Light-triggered action potentials in the liverwort *Conocephalum conicum.* Physiol Plant 64:482–486

Trebacz K, Simonias W, Schonknecht G (1994) Cytoplasmic Ca^{2+}, K^{+}, Cl^{-}, and NO_3^{-} activities in the liverwort *Conocephalum conicum* L, at rest and during action potentials. Plant Phys 106:1073–1084

Vian A, Henry-Vian C, Davies E (1999) Rapid and systemic accumulation of chloroplast mRNA binding protein transcripts after flame stimulus in tomato. Plant Physiol 121:517–524

Volkov AG, Haack RA (1995) Insect induced bioelectrochemical signals in potato plants. Bioelectrochem Bioenerg 35:55–60

Volkov AG (2000) Green plants/electrochemical interfaces. J Electroanal Chem 483:150–156

Volkov AG, Dunkley TC, Labady AJ, Brown C (2005) Phototropism and electrified interfaces in green plants. Electrochim Acta 504241–4247

Wacke M, Thiel G (2001) Electrically-triggered all-or-none liberation during action potential in the giant alga, Chara. J Gen Physiol 118:11–21

Wayne R (1994) The excitability of plant cells: with a special emphasis on characean internodal cells. Bot Rev 60:265–367

Wildon DC, Thain JF, Minchin PEH, Gubb IR, Reilly AJ, Skipper YD, Doherty HM, O'Donnell PJ, Bowles DJ (1992) Electrical signaling and systemic proteinase inhibitor induction in the wounded plant. Nature 360: 62-65.

Zawadzki T, DaviesE, Dziubinska H, Trebacz K (1991) Characteristics of action potentials in *Helianthus annuus* L. Physiol Plant 83:601–604

Zawadzki T, Dziubinska H, Davies E (1995) Characteristics of action potentials generated spontaneously in *Helianthus annuus*. Physiol Plant 93:291–297