

EXPERT VIEW

# Cation transporters in cell fate determination and plant adaptive responses to a low-oxygen environment

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## Abstract

Soil flooding creates low-oxygen environments in root zones and thus severely affects plant growth and productivity. Plants adapt to low-oxygen environments by a suite of orchestrated metabolic and anatomical alterations. Of these, formation of aerenchyma and development of adventitious roots are considered very critical to enable plant performance in waterlogged soils. Both traits have been firmly associated with stress-induced increases in ethylene levels in root tissues that operate upstream of signalling pathways. Recently, we used a bioinformatic approach to demonstrate that several Ca<sup>2+</sup> and K<sup>+</sup>-permeable channels from KCO, AKT, and TPC families could also operate in low oxygen sensing in *Arabidopsis*. Here we argue that low-oxygen-induced changes to cellular ion homeostasis and operation of membrane transporters may be critical for cell fate determination and formation of the lysigenous aerenchyma in plant roots and shaping the root architecture and adventitious root development in grasses. We summarize the existing evidence for a causal link between tissue-specific changes in oxygen concentration, intracellular Ca<sup>2+</sup> and K<sup>+</sup> homeostasis, and reactive oxygen species levels, and their role in conferring those two major traits enabling plant adaptation to a low-oxygen environment. We conclude that, for efficient operation, plants may rely on several complementary signalling pathway mechanisms that operate in concert and ‘fine-tune’ each other. A better understanding of this interaction may create additional and previously unexplored opportunities to crop breeders to improve cereal crop yield losses to soil flooding.

**Keywords:** Adventitious roots, aerenchyma, ethylene, hypoxia, NADPH oxidase, potassium, programmed cell death, reactive oxygen species.

## Introduction

### *Soil flooding as a major constraint for agricultural sustainability and food security*

The Sustainable Development Goals formulated by the United Nations aim to boost agricultural crop production to meet demands for global food security under current climate change. Of all natural hazards, floods and droughts have the biggest impact on the agriculture sector (FAO, 2015). Soil flooding creates low-oxygen (hypoxic) conditions in the root zone and severely hinders plant performance, resulting in substantial losses in crop production and even plant death. Urgency is enormous as global climate change is expected to increase the frequency and severity of flooding events in many regions worldwide (Tanoue *et al.*, 2016; Paprotny *et al.*, 2018; Box 1).

With the exception of rice, crops do not tolerate floods and are severely damaged when soil waterlogging occurs (Bailey-Serres and Voeselek, 2008). On a global scale, floods were the

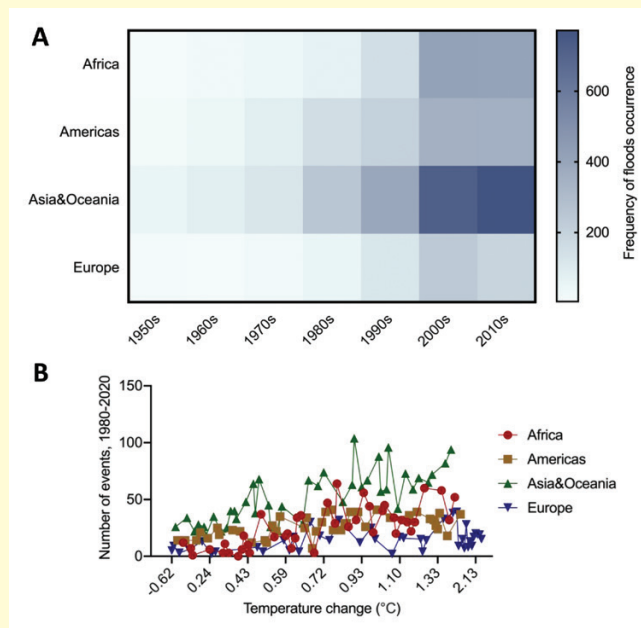
cause of almost two-thirds of all damage and loss to crops in the period between 2006 and 2016 (Fukao *et al.*, 2019). Over 1.7 billion ha of land is affected by soil flooding annually, with an estimated cost to the industry exceeding US\$80 billion year<sup>-1</sup> (Voeselek and Sasidharan, 2013). In Australia, the annual yield loss due to waterlogging ranges between 20% and 70% (depending on crop species and location; Shaw *et al.*, 2013); the same is true for other major crop production areas. Thus, the sustainability and profitability of agriculture in the 21st century is critically dependent on our ability to develop crop varieties that can maintain productivity when affected by soil hypoxia imposed by excessive water.

### *Plant adaptation to waterlogging*

The key constraint affecting plant performance in waterlogged soils is a lack of oxygen that creates hypoxic (or even anoxic—depending on duration and tissue type) conditions in plant

#### **Box 1. Flooding occurrence and effect in the context of global climate change.**

A heatmap showing the trend of global occurrence of climate-driven flooding events in different continents over the last 70 years (A). The colour scale indicates the number of flood events during corresponding periods. The presented information is extracted from the Emergency Events Database, EM-DAT (<https://public.emdat.be/>) provided by the Centre for Research on the Epidemiology of Disasters (CRED). The search terms are picked up as 'Natural', 'Hydrological', and 'Flood' for the corresponding disaster measure and time span.



The occurrence of flood events per year coincides with a simultaneous rise in the annual global temperature since 1980 (plotted on the x-axis in B). More frequent and intense flood events are likely to contribute to more severe damage and loss related to climate change. The presented analysis is based on the database from EM-DAT CRED, and the changes of temperature are reported in FAOSTAT.

cells. Hypoxia affects root metabolism, hampers aerobic respiration and ATP synthesis, and therefore has a severe impact on plant growth and yield (Gibbs and Greenway, 2003; Bailey-Serres and Colmer, 2014; Loreti *et al.*, 2016). Consequently, hypoxia also limits the availability of required energy to fuel the H<sup>+</sup>-ATPase pumps and severely hinders the plant's ability to deliver water and nutrients to the shoot (Bailey-Serres and Voesenek, 2008; Elzenga and van Veen, 2010; Gill *et al.*, 2018). Many plant species try to avoid hypoxic conditions by several means; two important traits found in grasses are formation of aerenchyma (Evans, 2003; Abiko *et al.*, 2012; Loreti and Perata, 2020; Yamauchi *et al.*, 2021) and development of adventitious roots (Steffens and Rasmussen, 2016; Lin and Sauter, 2020). Other key traits include formation of a barrier against radial oxygen loss and enhanced shoot elongation to overgrow deeper water layers to avoid hypoxia. These two traits are discussed extensively in other publications (Voesenek and Bailey-Serres, 2015; Mustroph, 2018; Pedersen *et al.*, 2021) and therefore are not covered here.

Formation of lysigenous aerenchyma in roots facilitates fast air diffusion between the atmosphere and the root tip, and allows the plant to function normally under flooded conditions (Jackson and Armstrong, 1999). This process requires the selective cell death and lysis of the cortical cells (Evans, 2003). Aerenchyma formation is constitutive in wetland species and in rice (the most flood-tolerant crop; Shiono *et al.*, 2011), and is inducible in other species upon onset of hypoxia (Colmer and Voesenek, 2009). Cell collapse during lysigenous aerenchyma formation resembles the canonical apoptotic pathway in animal cells (Evans, 2003; Yamauchi *et al.*, 2017; Ni *et al.*, 2019), and ethylene-mediated reactive oxygen species (ROS) signalling plays a critical role in this process (Colmer *et al.*, 2006; Rajhi *et al.*, 2011; Yamauchi *et al.*, 2014).

Formation of adventitious roots is considered as another key adaptive mechanism enabling plant adaptive responses to hypoxic conditions during waterlogging (Fukao and Bailey-Serres, 2004). The development of adventitious roots has been shown to play a very important role in water and nutrient uptake from the upper surface of the soil (Sauter, 2013; Steffens and Rasmussen, 2016) and is considered as the leading trait contributing to flood tolerance at the morphological level (Colmer and Voesenek, 2009; Manzur *et al.*, 2014). To emerge from their native organ, secondary roots must penetrate inner cell layers and the epidermis, which acts as a physical barrier to protect the organ (Steffens *et al.*, 2012). Thus, emergence of adventitious roots requires the cell death of epidermal cells that overlie adventitious root primordia. This process is known to be controlled by ethylene (Lorbiecke and Sauter, 1999; Steffens *et al.*, 2006; Dawood *et al.*, 2016), as ethylene insensitivity results in the formation of fewer adventitious roots (McDonald and Visser, 2003). The process also involves genes coding for cell wall-loosening enzymes, such as expansin and pectate lyase that are specifically expressed in front of emerging lateral root primordia (Neuteboom *et al.*, 1999). Adventitious root

formation is also controlled by GNOM, a guanine-nucleotide exchange factor which encodes a GDP/GTP exchange factor for small G proteins of the ADP-ribosylation factor class (ARF-GEF; Liu *et al.*, 2009).

### *Can ethylene do it all on its own?*

While most canonical models consider ethylene entrapment and signalling upstream of lysigenous aerenchyma formation (Yamauchi *et al.*, 2020), this is not necessarily true for all cases (e.g. aerenchyma formation in the root of the wetland plant *Juncus effusus*; Visser and Bögemann, 2006). Underwater growth of petioles/stems of *Nasturtium officinale* (watercress) also did not require ethylene (Muller *et al.*, 2021). Also, Parlanti *et al.* (2011) showed that in some rice genotypes, submergence led to an increase in the formation of aerenchyma without any noticeable increase in ethylene production; instead, it was causally related to hypoxia-induced elevation in H<sub>2</sub>O<sub>2</sub> levels. However, given the correlative nature of this study, to convincingly demonstrate the possibility of aerenchyma formation without involvement of ethylene signalling, experiments using available ethylene-insensitive rice mutants are required (Pandey *et al.*, 2021).

Furthermore, knocking out *RBOHH* (one of the key genes conferring NADPH oxidase activity) by CRISPR/Cas9 reduced ROS accumulation and inducible aerenchyma formation in rice roots (Yamauchi *et al.*, 2017). This suggests that (i) aerenchyma formation may occur independently of ethylene signalling in some cases (or plant species) and (ii) the latter process requires operation of an NADPH oxidase. So, ethylene *per se* could not do it all, and stress-induced modulation in ethylene production needs to be complemented by other signalling pathways.

Formation of the adventitious roots requires a mechanical rupture of the root cortex (Steffens *et al.*, 2012). It was shown that epidermal cells above root primordia that undergo cell death have a distinct molecular identity, with a transcriptome that is greatly different from that of other nodal epidermal cells (Steffens and Sauter, 2009). It was also shown that elevation in the ethylene content *per se* is not sufficient to trigger programmed cell death (PCD), and that reprogramming of epidermal cell fate by ethylene requires two signals: (i) mechano-sensing for spatial resolution and (ii) ROS for cell death signalling (Steffens *et al.*, 2012). According to the suggested model, adventitious root primordia create a mechanical force that is then sensed by some mechanosensitive channels (with as yet unknown molecular identity; Steffens *et al.*, 2012) that later regulate the cell's redox status and, thus, determines its fate.

### *Where do membrane transporters step into the picture?*

One of the most enigmatic questions in plant responses to flooding is the molecular identity of the oxygen sensors. In

Arabidopsis, hypoxia responses are controlled by five essential transcription factors belonging to the group VII ERFs: HYPOXIA RESPONSIVE ERF1 (HRE1) and HRE2, and RELATED TO APETALA2.2 (RAP2.2), RAP2.12, and RAP2.3 that are controlled by the cellular oxygen status. While some of the encoding genes react to hypoxic stress, RAP2.2, RAP2.3, and RAP2.12 are constitutively transcribed under normoxic conditions (Papdi *et al.*, 2008, 2015; Hinz *et al.*, 2010). In the light of this, plant low-oxygen sensing was attributed to the stability of these ERF-VII through the *PROTEOLYSIS6* (*PRT6*) N-degron pathway (Gibbs *et al.*, 2011; Licausi *et al.*, 2011; Weits *et al.*, 2014; van Dongen and Licausi, 2015). However, more recent evidence (Gibbs *et al.*, 2014; Hartman *et al.*, 2019) suggests that the ERF-VII can still be stabilized in the presence of oxygen through other means (Hartman *et al.*, 2019), and that the actual oxygen sensing could be performed by either plant cysteine oxidases (Weits *et al.*, 2014; White *et al.*, 2017; Masson *et al.*, 2019) or other molecules (Holdsworth, 2017). In mammalian systems, several K<sup>+</sup> and Ca<sup>2+</sup> channels were proposed to operate in oxygen sensing (Kemp and Peers, 2007), and our bioinformatics analysis has identified six channels from three families as potential oxygen sensors in plants. These comprise (Wang *et al.*, 2017b): four members of

the tandem-pore K<sup>+</sup> channel family (namely KCO1, KCO4, KCO5, and KCO6); an AKT2 Shaker K<sup>+</sup> channel; and a two-pore Ca<sup>2+</sup>-permeable (TPC1) channel. While these findings are waiting for experimental validation in direct patch-clamp experiments, they are indicative of the possible causal link between changes of intracellular Ca<sup>2+</sup> and K<sup>+</sup> homeostasis and plant adaptive responses to a low-oxygen environment (Box 2).

Anion channels also may play a critical role in plant adaptive responses to flooding. Soil flooding results in a significant cytosolic acidosis (Felle, 2005), and recently Lehmann *et al.* (2021) showed that the anion channel SLAH3 can sense changes in cytosolic acidification by specific histidine residues, thereby switching from a 'silent' dimer into active monomers, triggering membrane depolarization and activating signalling cascades boosting the plant's flooding stress response. Also, one of the most prominent biochemical alterations under flooded conditions is a dramatic (several orders of magnitude) increase in the  $\gamma$ -aminobutyric acid (GABA) content (Kreuzwieser *et al.*, 2009). It was recently shown that hypoxia-induced elevation in the GABA level may restore membrane potential by pH-dependent regulation of H<sup>+</sup>-ATPase and/or by generating more energy through the activation of the GABA shunt pathway and the tricarboxylic acid (TCA) cycle (Wu *et al.*, 2021).

### Box 2. Key developments in understanding the role of membrane transporters in plant adaptive responses to the low oxygen environment.

- **Ion channels may operate as low-oxygen sensors in plants**

The bioinformatics analysis has identified six cation channels from three families as potential oxygen sensors in plants (Wang *et al.*, 2017b). These comprise four members of the tandem-pore K<sup>+</sup> channel family (KCO1, KCO4, KCO5, and KCO6); an AKT2 Shaker K<sup>+</sup> channel; and a two-pore Ca<sup>2+</sup>-permeable (TPC1) channel.

- **Hypoxia-induced K<sup>+</sup> loss may trigger PCD in root cortex**

The onset of hypoxia activates outward-rectifying K<sup>+</sup> GORK channels (Gill *et al.*, 2018), resulting in cytosolic K<sup>+</sup> loss. The failure to maintain high cytosolic K<sup>+</sup> levels can induce cell elimination via programmed cell death (PCD) by unblocking activities of caspase-like proteases and endonucleases (Rubio *et al.*, 2020) and potentially leading to formation of a lysigenous aerenchyma.

- **Adventitious root development requires operation of mechano-sensitive ion channels**

The emergence of adventitious roots implies a mechanical rupture of the root cortex and requires involvement of ROS and mechano-sensitive ion channels (Steffens *et al.*, 2012). These channels are Ca<sup>2+</sup> permeable (Frachisse *et al.*, 2020).

- **NADPH oxidase and Ca<sup>2+</sup>-permeable channels form a 'hub' and amplify ROS signal required for adventitious root formation**

NADPH oxidase encoded by RBOH genes shapes hypoxia-specific Ca<sup>2+</sup> signatures via the modulation of apoplastic H<sub>2</sub>O<sub>2</sub> production (Wang *et al.*, 2017a). Ca<sup>2+</sup>-permeable mechano-sensitive ion channels operate in tandem with RBOH, forming a positive feedback loop (Demidchik *et al.*, 2018), and may confer PCD of epidermal cells around root primordia.



*Membrane transporters and aerenchyma development*

Earlier we showed that onset of hypoxia induces severe membrane depolarization that activates outward-rectifying K<sup>+</sup> GORK channels (Gill *et al.*, 2018), resulting in the cytosolic K<sup>+</sup> loss. We have also shown that elevation in intracellular ROS levels triggers massive K<sup>+</sup> loss from the cytosol by activating non-selective cation channels (NSCCs) (Shabala *et al.*, 2016). At the same time, when plants deal with energy crises imposed by hypoxia, the relative amount of ATP allocated for (defence-related) protein synthesis is increased from 38% to 73%, while energy allocation to net K<sup>+</sup> transport drops from 85 nmol ATP g<sup>-1</sup> FW min<sup>-1</sup> to almost zero (Atwell *et al.*, 2015). The failure to maintain high cytosolic K<sup>+</sup> levels can induce cell elimination via PCD by unblocking activities of caspase-like proteases and endonucleases (Shabala *et al.*, 2007; Demidchik *et al.*, 2010). Thus, we propose that this mechanism may operate in formation of lysigenous aerenchyma under low-oxygen conditions (Box 3). The supporting evidence for this model comes from recent electrophysiological and fluorescence imaging experiments on barley conducted in our laboratory. By screening a large number (>300) of barley accessions with contrasting waterlogging stress tolerance, we have revealed that tolerant genotypes are capable of forming aerenchyma after 4–5 d from hypoxia onset, while this process is started 3 d later in the sensitive varieties (Zhang *et al.*, 2015). When contrasting genotypes were selected and examined, it was found that most tolerant genotypes show early accumulation of ROS species (Box 3; panel B). These genotypes also possess increased sensitivity of K<sup>+</sup> channels to H<sub>2</sub>O<sub>2</sub> (Box 3; panel C). These observations are fully consistent with the above model (Box 3) that low-oxygen sensing in tolerant varieties leads to early accumulation of ROS, following their activation of K<sup>+</sup> efflux channels and a consequent decrease in the cytosolic K<sup>+</sup> levels that then leads to execution of PCD by caspase-like proteases whose activity is suppressed by the elevated K<sup>+</sup> level. The full validation of this model should come from experiments combining time-dependent tissue-specific ROS accumulation in root cortical cells of contrasting genotypes/species, changes in transporter activity and transcript levels of key genes involved in ROS and ethylene signalling, and K<sup>+</sup> and Ca<sup>2+</sup> homeostasis and monitoring of PCD events. The molecular identity of NSCCs mediating ROS-induced K<sup>+</sup> loss has also to be revealed.

*Membrane transporters and formation of adventitious roots*

Findings from Sauter's lab (Steffens *et al.*, 2012) suggested that reprogramming of epidermal cell fate by ethylene requires mechanical and ROS signals and involves mechano-sensing channels. While the exact molecular nature of the candidate mechano-sensitive channel remains unknown, earlier Nakagawa *et al.* (2007) showed that Arabidopsis MCA1 channels possess mechano-sensitivity and mediate Ca<sup>2+</sup> uptake. At

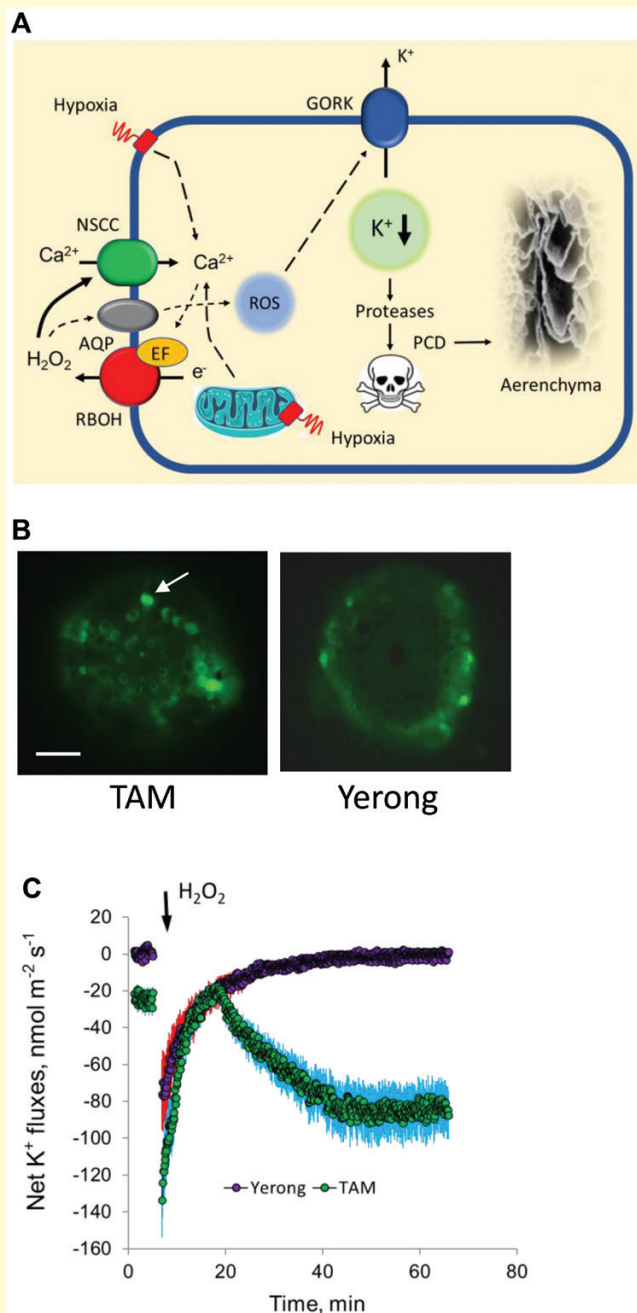
the same time, plants employ the so-called 'Ca-ROS Hub' that operates in amplification of ROS and Ca<sup>2+</sup> signals (Demidchik *et al.*, 2018) by forming a positive feedback loop involving Ca<sup>2+</sup>-permeable channels and NADPH oxidase. NADPH oxidase is a plasma membrane-bound enzyme complex. After binding Ca<sup>2+</sup> to one of its ER hands, NADPH oxidase produces the extracellular superoxide anion, O<sub>2</sub><sup>-</sup>, that is then converted to H<sub>2</sub>O<sub>2</sub> in the apoplast. The Arabidopsis genome contains 10 NOX (NADPH oxidase) genes, and earlier we showed that in Arabidopsis roots, RBOHD shapes hypoxia-specific Ca<sup>2+</sup> signatures via the modulation of apoplastic H<sub>2</sub>O<sub>2</sub> production (Wang *et al.*, 2017a). Thus, it is suggested that MCA1 may operate in tandem with RBOH, forming the 'ROS-Ca Hub' and conferring PCD of epidermal cells about root primordia (Box 4). Previous studies have reported very rapid (within 1–2 min) hypoxia-induced cytosolic Ca<sup>2+</sup> elevation in Arabidopsis plants (Sedbrook *et al.*, 1996) that was consistent with observed depolarization of membrane potential in the same time frame (Box 4). Future experiments should use accessions with contrasting abilities to develop adventitious roots when exposed to hypoxia, followed by their electrophysiological (MIFE and patch-clamp), biochemical, molecular, and microscopy assays. Another interesting possibility would be to compare the rice (*Oryza sativa*) *arl1* (*adventitious rootless 1*) mutant with its wild type. By matching the electrophysiological data to transcriptomics analysis, one will be able to understand the causal link between cell type-specific changes in the cytosolic Ca<sup>2+</sup> and K<sup>+</sup> levels, downstream signalling pathways, and membrane effectors (mechano-sensitive channels) that participate in cell fate determination and elimination of epidermal root cells.

*Implications and prospects*

Until now, the major focus of researchers working in the field was on understanding the role of ethylene and ROS signalling in plant adaptation to soil flooding (Voeselek and Bailey-Serres, 2015; Pucciariello and Perata, 2017; Hartman *et al.*, 2021). The role of ion transporters operating either downstream of, or in parallel to, the above signalling pathways remains largely unexplored. In the Arabidopsis genome, 43% of all protein sequences have at least one transmembrane-spanning (TMS) domain, with 18% proteins having at least two TMS domains and thus are associated with cellular membranes (Ward, 2001). The Arabidopsis genome also confers membrane transport proteins from 46 unique families containing ~880 members (Mäser *et al.*, 2001). Importantly, many of them can be directly regulated by ROS and low oxygen (Ward *et al.*, 2009; Wang *et al.*, 2017b; Demidchik *et al.*, 2018). Thus, revealing the roles and tissue-specific regulation of key transporters and understanding their roles as downstream targets for low-oxygen signalling and their interaction with ethylene- and ROS-mediated signalling pathways will fill the fundamental gap in our knowledge and reveal the fine print of one of the

### Box 3. Membrane transporters and aerenchyma development

The proposed model for formation of lysigenous aerenchyma under low-oxygen conditions (A). Hypoxia stress is sensed by mitochondria and/or some plasma membrane (PM)-based sensor and results in a rapid (1–2 min; Sedbrook *et al.*, 1996) elevation in cytosolic  $\text{Ca}^{2+}$ , leading to stimulation of NADPH oxidase (encoded by RBOH). The  $\text{H}_2\text{O}_2$  produced in the apoplast is transported across the PM via aquaporins (AQP). Elevated cytosolic  $\text{H}_2\text{O}_2$  activates  $\text{K}^+$ -permeable efflux channels (GORK in the model), increasing activity of caspase-like proteases and triggering PCD that results in elimination of the cortical cell and formation of aerenchyma.



The model is built upon findings that low oxygen stress induces accumulation of ROS in root cortical cells, and this accumulation occurs faster in the genotype (TAM) with a greater proportion of aerenchyma

**Box 3. Continued**

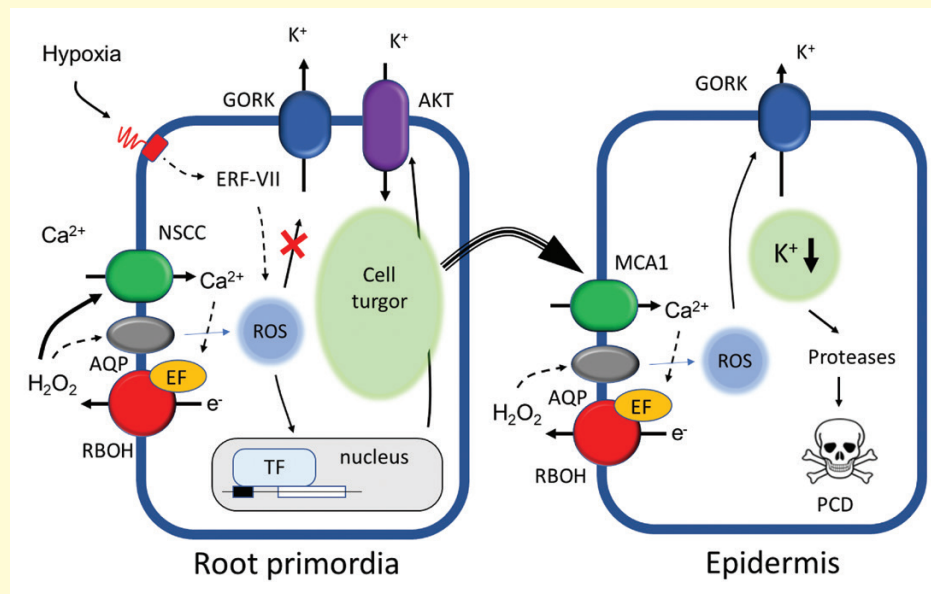
than the one (Yerong) with a relatively lower proportion of aerenchyma under waterlogging conditions (B). The sensitivity of  $K^+$  efflux channels to ROS is also increased in the genotype with ability for faster aerenchyma formation (C).

(B) Quantification of  $H_2O_2$  accumulation in roots of two barley genotypes (TAM, left panel; Yerong, right panel) after 3 d of waterlogging stress. Scale bar=200  $\mu$ m. ROS accumulation (marked by an arrow) was visualized by fluorescence imaging after staining with H<sub>2</sub>DCF-DA (see [Niu \*et al.\*, 2018](#) for details) and was much stronger in TAM compared with Yerong. These patterns matched faster formation of aerenchyma and increase sensitization of  $K^+$  efflux channels in TAM.

(C) Kinetics of net  $H_2O_2$  (10 mM)-induced  $K^+$  fluxes measured from epidermal root cells of TAM and Yerong barley genotypes after 3 d of hypoxia treatment measured by the non-invasive microelectrode ion flux measuring (MIFE) technique. For all details of MIFE operation and treatments, please refer to [Gill \*et al.\* \(2018\)](#). Data are means  $\pm$ SE ( $n=6$ ). The sign convention is 'efflux negative'.

**Box 4. Membrane transporters and adventitious root development.**

The proposed model for involvement of membrane transporters in adventitious root development. Hypoxia conditions in root primordia are sensed by a PM-based receptor and then signalled by ERF-VII to elevate intracellular ROS content ([Sasidharan and Voesenek, 2015](#)). ROS signal is then amplified by the ROS- $Ca^{2+}$  'hub' composed of RBOH and a ROS-inducible  $Ca^{2+}$ -permeable non-selective cation channel (NSCC) ([Demidchik \*et al.\* 2018](#)). ROS and  $Ca^{2+}$  signals induce appropriate transcription factors (TFs) in the nucleus to increase cell turgor pressure and drive expansion growth (e.g. by up-regulating expression and/or activity of AKT  $K^+$  uptake channels in the model). ROS activation of  $K^+$  efflux GORK channels is prevented by their desensitization in this tissue.



In the epidermal cell,  $Ca^{2+}$  channels making up the above 'hub' are not ROS inducible but instead possess mechano-sensitivity (e.g. MCA1 channel in the model; [Nakagawa \*et al.\*, 2007](#)), so the RBOH- $Ca^{2+}$  hub will operate only in the presence of a mechanical pressure exerted on the cell by expanding primordia. At the same time, the GORK channels in root epidermis possess high ROS sensitivity (e.g. [Shabala \*et al.\*, 2016](#)) and could be rapidly activated by ROS accumulation in the cytosol transported from the apoplast via aquaporins (AQP). As a result, cytosolic  $K^+$  drops to levels which allow activation of proteases, and the epidermal cell undergoes PCD, thus allowing developing adventitious roots to penetrate.

very important mechanisms evolved by plants to deal with flooded conditions. This fundamental knowledge may then be used by plant breeders to improve waterlogging tolerance in major staple crops. It would be naive to expect that there will be one single gene that can be a ‘silver bullet’ that will fix all the problems; however, the chances that some of these new traits may be gamechangers are very high. The sooner plants are able to embrace these anatomical adaptations the smaller will be the detrimental effect of hypoxia stress on their overall long-term performance during flooding stress.

The importance of understanding the role of membrane transporters in low-oxygen conditions goes well beyond improving plant adaptive responses to flooding. Low-oxygen conditions are often found in some plant tissues under non-stressful conditions (e.g. meristems, seeds, and tubers), indicating that oxygen gradients play a role in the control of stem cell activity in shoot meristems (Meitha *et al.*, 2018; Weits *et al.*, 2019). Thus, understanding mechanisms by which plants sense and signal low-oxygen conditions, and the role of specific ion transporters in this process, is of a fundamental importance to understanding a broad array of processes related to dormancy and plant organogenesis.

## Author contributions

SS: conceptualization and drafting the manuscript; LS, XH, and XZ: conducting experiments supporting key concepts; LACJV, SH, YU, and MZ: commenting on the draft and improving appropriate sections.

## Conflict of interest

No conflict of interest declared.

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