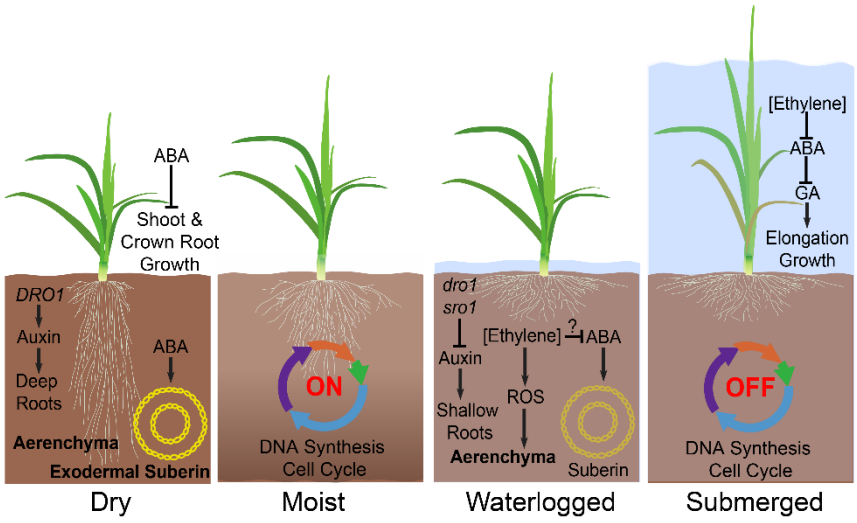

Figure 1. Characteristics of shoot and root growth plasticity associated with variation in soil moisture using semi-dwarf rice as the model. Roots in dry soils grow towards water reserves and limit water loss by deposition of a pronounced suberin barrier, particularly surrounding the exterior side of the exodermis. In moist soils, root architecture is more compact, crown root initiation and elongation is active and exodermal suberization is limited. Waterlogged roots containing aerenchyma proliferate near the soil surface and exodermal suberin prevents oxygen loss by radial diffusion. In fully submerged plants, cell division activity in root systems is rapidly switched off and leaf elongation is enhanced, promoting escape to the air. Key phytohormones, their relationships and genes mentioned in the main text are shown.

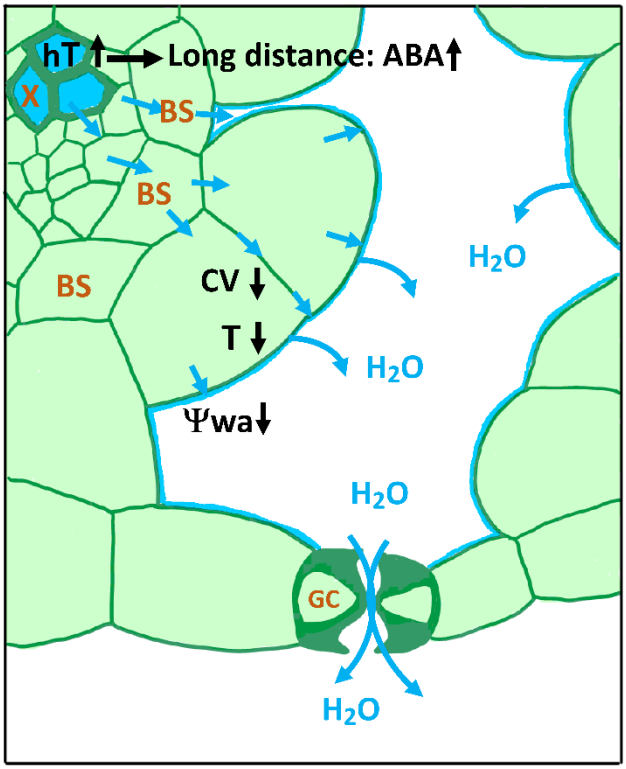

Figure 2. Water flux from the xylem vessel to the evaporation site in a schematic leaf cross section. Water transported through the xylem (X) of a small leaf vein passes through bundle sheath cells (BS) to mesophyll and epidermal cells, which surround guard cells (GC), the sites of transpiration. GC are symplastically isolated (have no plasmodesmata) and water supply occurs from the apoplast. Transpiration exceeding water replenishing at the evaporation sites results in a reduced water potential of the apoplastic fluid (ψ_{wa}). This key event triggers a number of changes: a reduction of turgor (T) and cell volume (CV) within the mesophyll, and an increase in the hydraulic tension (hT) within the vasculature. The change in hT serves as fast long-distance signal and there might be parallels with mechanical sensing and relay of touch and wounding. The hydraulic signal is converted into the chemical signal ABA to adjust water conductance and stomatal aperture. Mechano-sensitive components are likely involved in all cell types but have not been unequivocally identified.

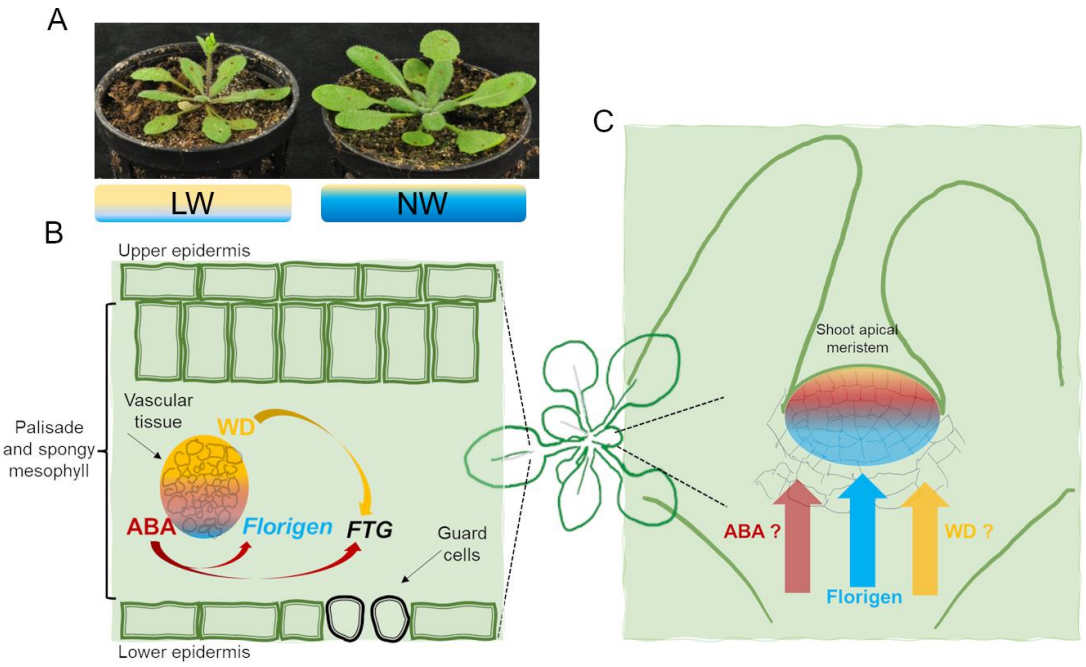

Figure 3. Known and uncharacterised signals causing DE and their sites of action. A, wild-type *Arabidopsis* photographed at approximately 3 weeks from germination. Plants were grown under normal or low-watering (NW and LW, respectively) regimes. LW plants exhibit DE. B, Cartoon outlining a leaf cross-section. Water deficit signals (WD, yellow) are transmitted through the vasculature to stimulate ABA production (dark red). ABA positively contributes to florigen (cyan) transcriptional activation in leaves in response to water deficit. ABA and WD promote expression of many other flowering time genes (FTG, black) regulating guard cells activity and drought tolerance traits. C, besides florigen proteins, different signals converge at the shoot apical meristem to relay water deficit information and regulate floral transition accordingly. Despite the ability of the shoot meristem cells to integrate these cues, it is still unclear how water deficit signals (WD) can reach the apex and if significant ABA translocation occurs.

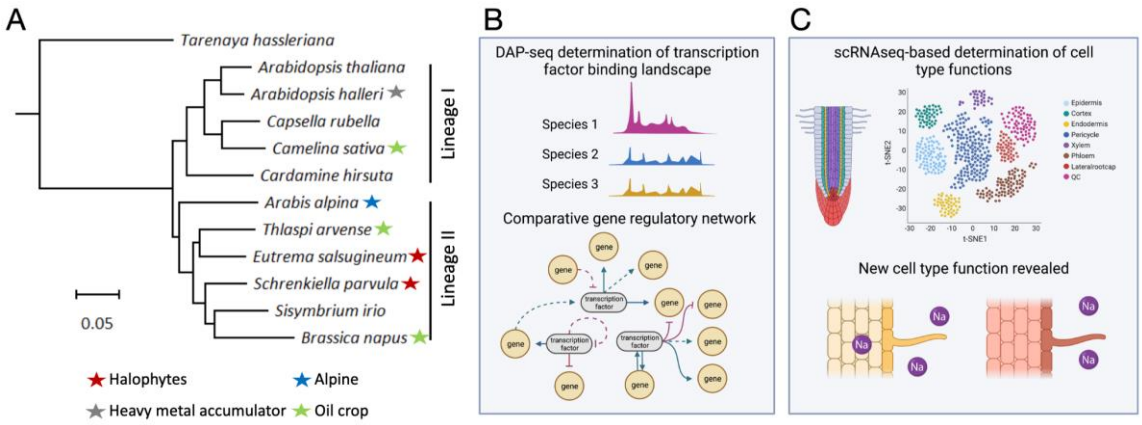

Figure 4: The Brassicaceae family contains species with diverse physiological traits and agronomic value.

A. Phylogeny of species in the Brassicaceae family with sequenced genomes. Stars indicate different physiological or agronomic properties of interest. Figure courtesy of Dong-Ha Oh.

B. DAP-seq facilitates the rapid determination of transcription factor-genome interaction landscapes. Gene regulatory network architecture can be compared between species to identify rewiring events.

C. scRNAseq enables cell-type functions to be explored between species and for new functions to be uncovered.

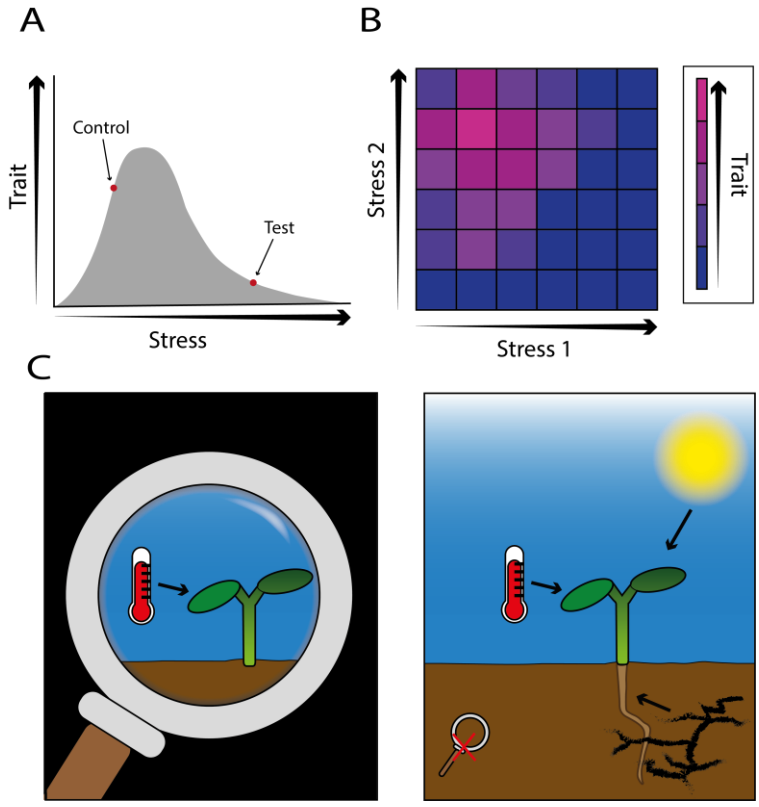

Figure 5. How do plants integrate climate signals?
A) Most environmental cues do not have linear effects on plant development. When investigating the molecular basis for a particular cue, it is important to consider that different intensities of the same stress likely act through different molecular pathways. B) Representing gradients of two stresses as a heat map could be a potential way to conceptualize complex interactions. Note that the intensity of Stress 1 affects the way plants respond to the intensity of Stress 2. C) Through taking a step back and studying the interactions between signaling pathways in their relevant environmental context, we will come closer to understanding the molecular basis for plant development in natural environments.

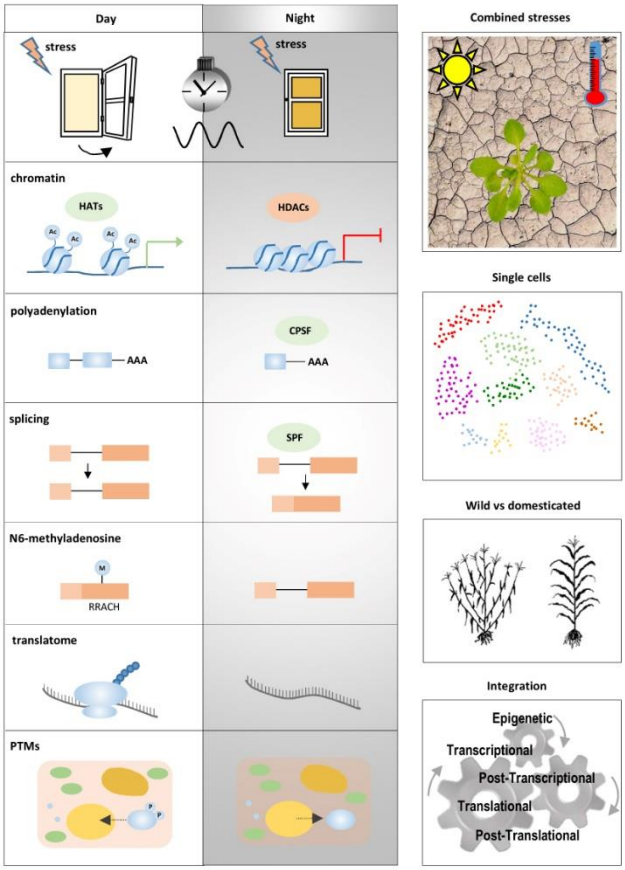

Figure 6. Circadian gating by the clock differentially regulates plant responses to stress depending on the time-of-day. Figurative examples of open questions related to the circadian gating via changes in chromatin, post-transcriptional, translational and post-translational (PTMs) mechanisms of regulation that ultimately define gene expression and protein function under stress conditions. Single-cell approaches with plants grown under conditions that closely follow the natural environment, including combined stresses, and comparisons of domesticated crops and their wild relatives are circadian research areas of increasing interest. Understanding the coupling among the different gating mechanisms will pave new ways for using the circadian clock to tackle the effects of abiotic stress on plants.

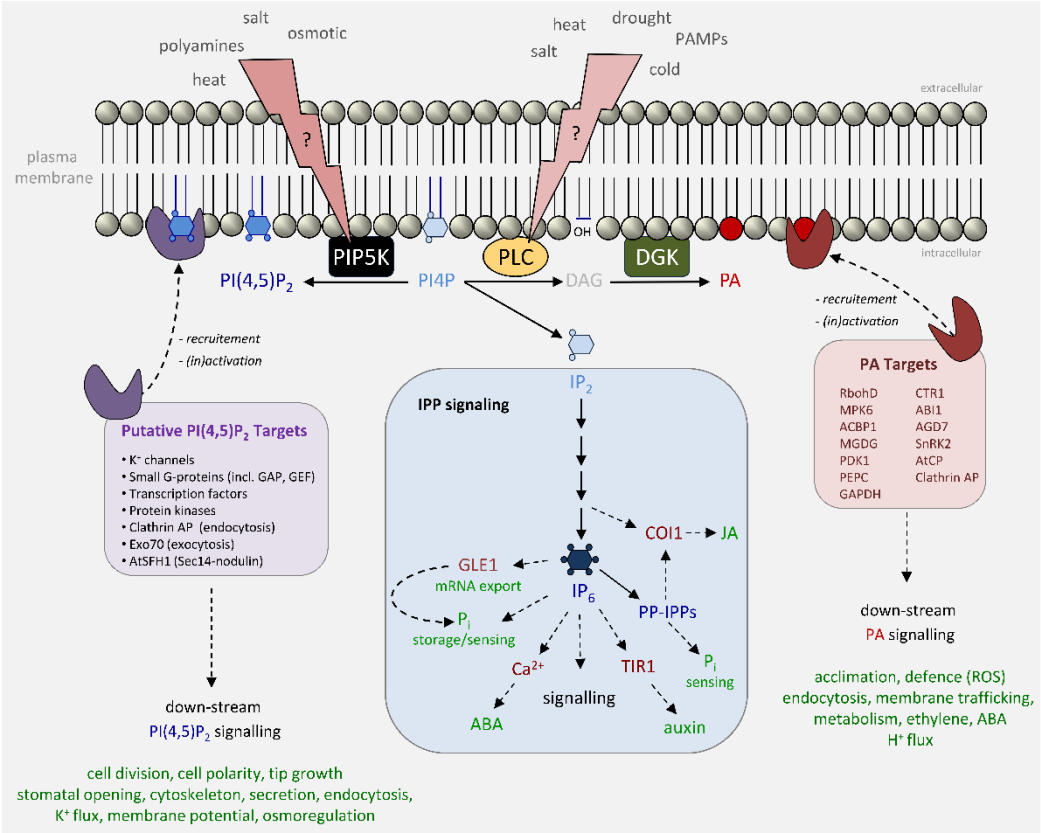

Figure 7: Possible signaling mechanisms of PI(4,5)P_2, PA and PLC in plant abiotic-stress signaling. PI(4,5)P_2 concentrations in plants are extremely low but synthesis is rapidly triggered by activation of one or more of the 11 PIP5Ks of Arabidopsis via unknown receptors or activation mechanisms (indicated by "?"). As such, PI(4,5)P_2 is triggered by heat, salinity, osmotic stress and polyamines, but also during polar growth, predominantly accumulating at the plasma membrane. This likely causes recruitment of target proteins to the membrane or (in)activation of targets that are already present at the membrane. Putative plant targets are based on the mammalian literature and correlations with plant literature.

Plant PLCs likely hydrolyse PI4P and generate membrane-bound DAG and the water-soluble headgroup, IP_2. Again, receptors and activation mechanisms remain unknown (indicated by "?"). The newly formed DAG is rapidly phosphorylated by DGK to form the signaling lipid PA for which several protein targets have been identified. IP_2 can be stepwise phosphorylated by IPP multi-kinases to produce IP_5 and IP_6, and IPP-pyrophosphates IP_7 and IP_8, for which several signaling functions and targets are emerging. Protein targets are indicated in purple [PI(4,5)P_2] or dark red [PA]. Lipid (-derived) signals are indicated in blue. Solid lines indicate metabolic conversions, dashed lines represent 'activation' or 'cause'. Effects are indicated in green.

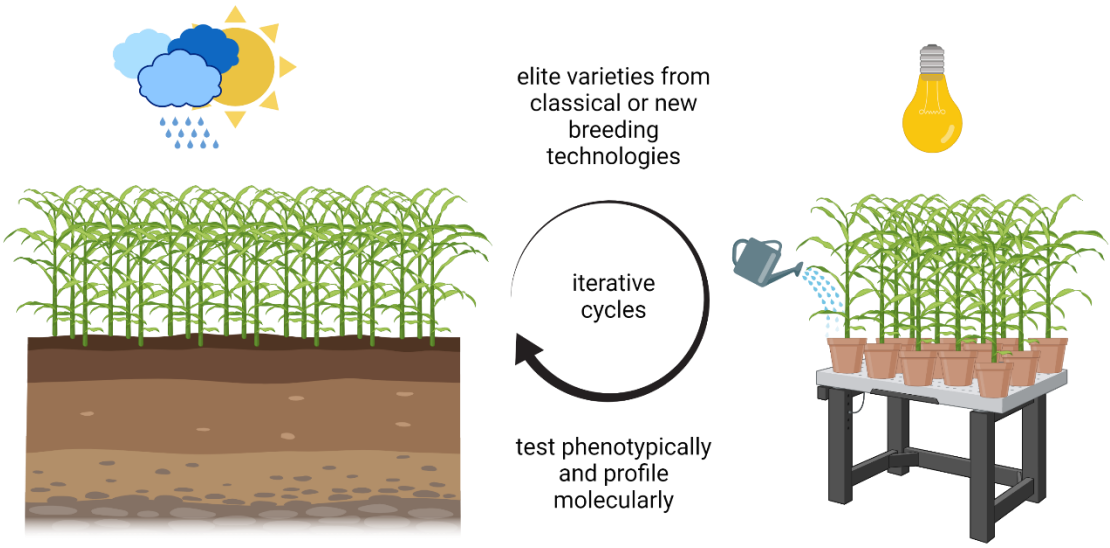

Figure 8: Schematic representation of the differences in management, light, and irrigation schemes between plants grown in the field (left) and in controlled conditions. Iterative cycles of field testing and molecular profiling of all lines with putatively improved traits will be necessary to increase our understanding how the stresses that plants experience in the field relate to the controlled abiotic stress conditions. Created with BioRender.com.

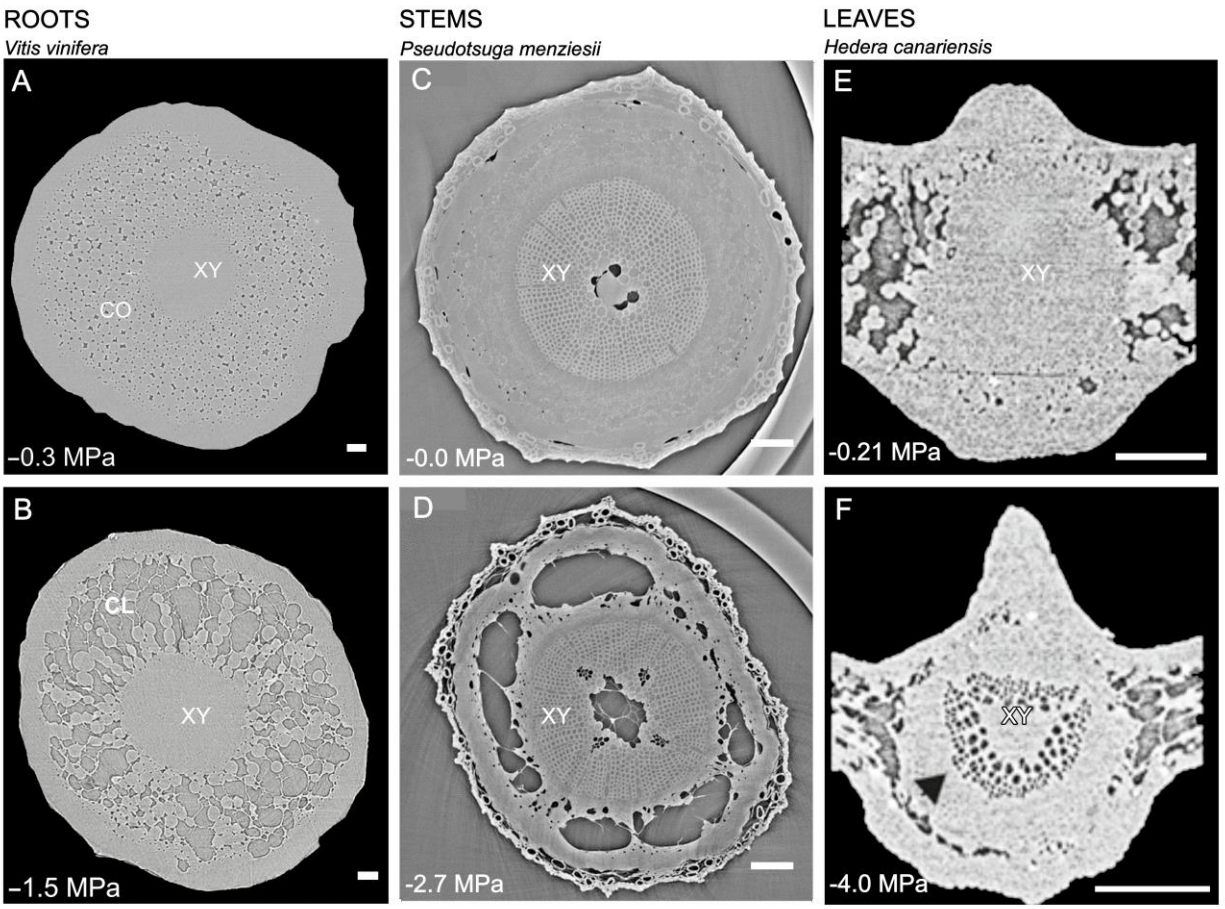

Figure 9: The effects of drought on tissues across plant organs.
Representative transverse X-ray micro-computed tomography images show the *in vivo* functional status of the vascular system and the surrounding tissues under well-watered (A, C and E) and drought (B, D and F) conditions in roots, stems, and leaves. In A and B, the roots of grapevine (*Vitis vinifera*) the xylem network (XY) remains functional under a range of water potentials (ψ_w) but mild drought (-1.5 MPa) leads to massive cellular damage and formation of cortical lacunae (CL in panel B) that physically decouple the vascular system from the surrounding soil (Cuneo et al., 2021). In C and D, the vascular systems of the immature stems of conifer seedlings (*Pseudotsuga menziesii*) are highly drought tolerant (XY), but deformation of the cortex under strong drought in these resembles that in roots (Miller et al., 2020). In E and F, leaves of ivy (*Hedera canariensis*), subjected to very strong dehydration (-4.0 MPa) show xylem cavitation in the midrib (XY) and the strong mesophyll tissue deformation that occurs during drought exceeding the turgor loss point (Scoffoni et al., 2017). Bars = 200 µm in A and B; 100 µm in C and D; 250 µm in E and F. Values in the lower left corner of each panel indicate the ψ_w of the plant during the experiment.

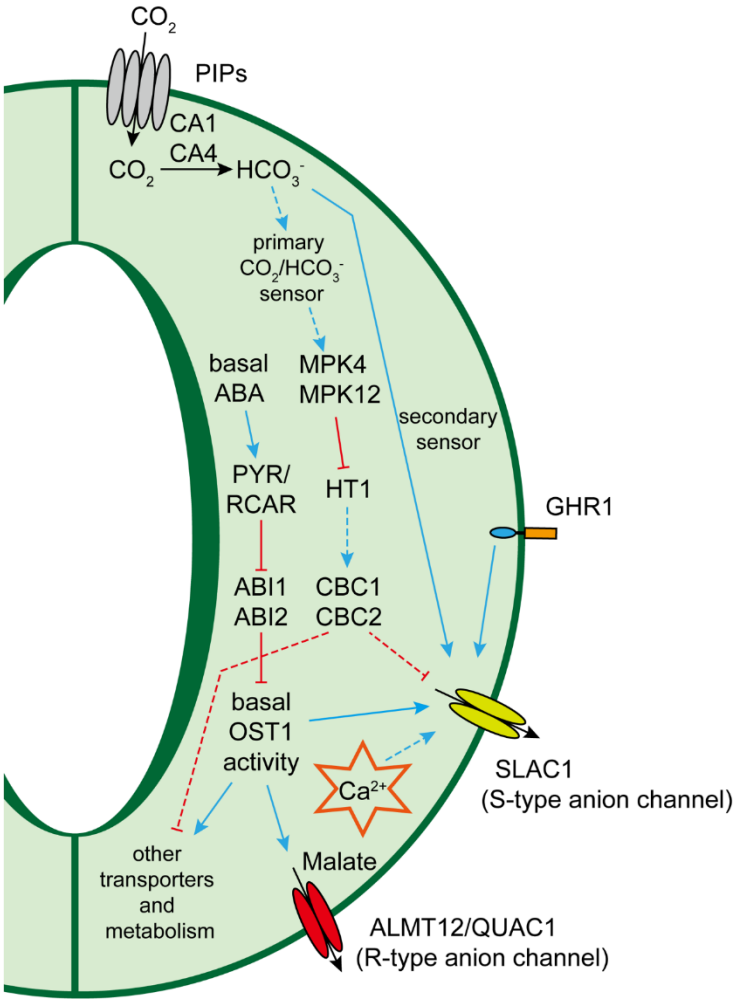

Figure 10: Simplified model for CO_2 sensing and signaling in stomatal closure.
Schematic diagram of signaling components and pathways in CO_2-induced stomatal closure. Black arrows indicate the directions of transport or enzymatic reaction. Blue arrows and red blocks represent positive and negative regulation of high CO_2-triggered stomatal closure respectively. PIP2 Aquaporins (PIPs) and β-carbonic anhydrases (CA1/CA4) facilitate CO_2 influx and HCO_3^- and proton production in guard cells. Protein kinases including MPK4/MPK12, HT1, and CBC1/CBC2 are known early signaling components involved in regulating ion transporters, including the S-type anion channel SLAC1 in response to CO_2/HCO_3^- elevation. However, many detailed mechanisms in CO_2 sensing and signal transduction remain to be investigated as indicated by dash arrows and red dashed blocks. Basal ABA signaling and basal OST1/SnRK2 kinase activities facilitate high CO_2-induced stomatal closure. A receptor-like pseudokinase GHR1 is required for SLAC1 activation under CO_2 elevation and several other environmental stimuli.

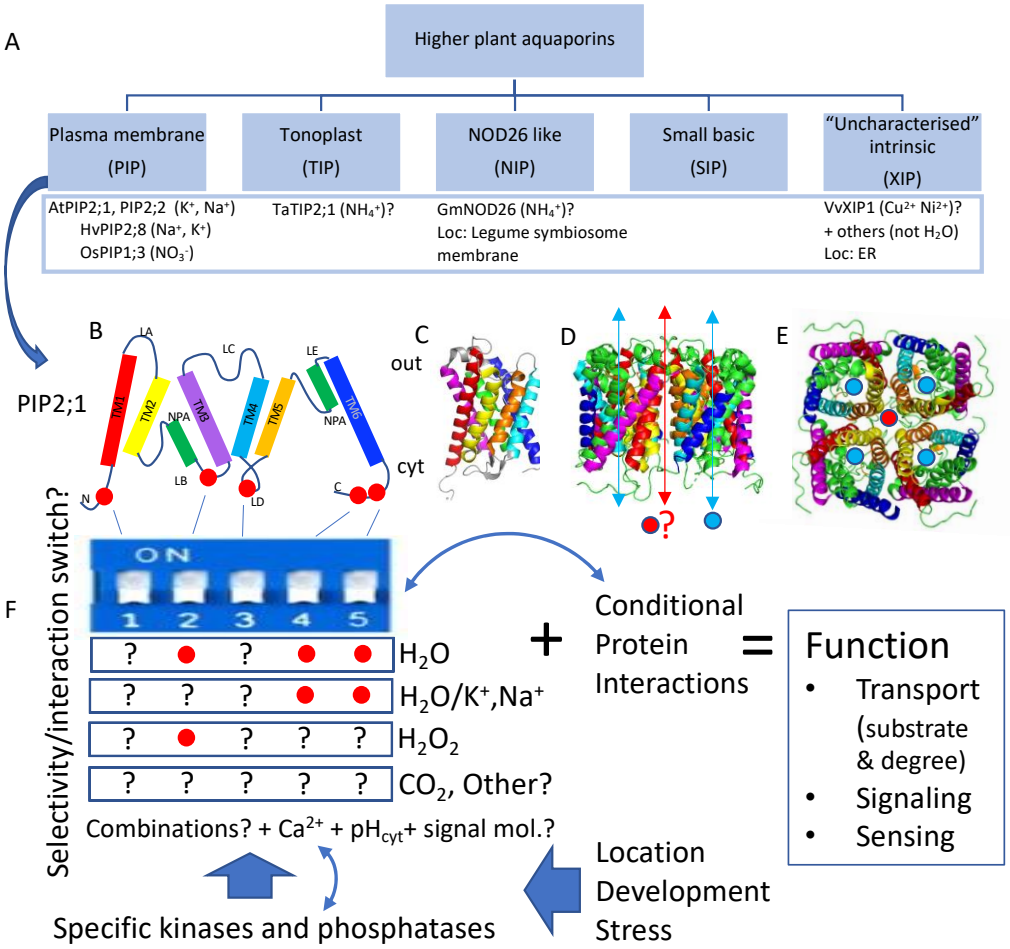

Figure 11. How an aquaporin can be multifunctional. A. Aquaporin family with those indicated to transport ions electrogenically (under each subfamily, ?=maybe, Loc= membrane location for specific isoform, note many NIPs are located on the plasma membrane) (Tyerman et al., 2021). B. Features of PIP2;1 showing transmembrane helices (TM1-6), loops (A-E) and half helices forming the central selectivity NPA filter. Red dots = phosphorylation sites. C. Folded monomer structure (SoPIP2;1 PDB 1z98)(Törnroth-Horsefield et al., 2006). D Homotetramer as the functional unit with 4 monomeric pores that transport water (blue arrows, dots) and probably H2O2, also NH3 in TIP2;1 (Kirscht et al., 2016) and the central pore (red arrow, dot) implicated in ion transport for mammalian AQP1 (Henderson et al., 2022). AQPs also likely form heterotetramers (e.g. ZmPIP1;2 + ZmPIP2;1) (Chaumont and Tyerman, 2014). E. View normal to the membrane plane on the cytoplasmic face. F Hypothesis for multifunctionality of PIP2;1. Phosphorylation by specific kinases occurs at one or more of the sites on cytoplasmic loops like a digital dipswitch that determine protein interactions and/or substrate selectivity, e.g. ion water reciprocity (Qiu et al., 2020) or H2O2 permeation (Rodrigues et al., 2017), or protein interactions (Prado et al., 2019). Selectivity is fixed to a certain degree in the monomeric pores by the structure of the ar/R selectivity filter and other residues. The hydrophobic central pore may still allow ion permeation and is proposed to be gated at a low probability dependent on monomer gating (Tyerman et al., 2021). There are likely feedbacks (small blue arrows) due to some substrates and protein interactions also affecting the signalling that determines the kinase/phosphatase activity.

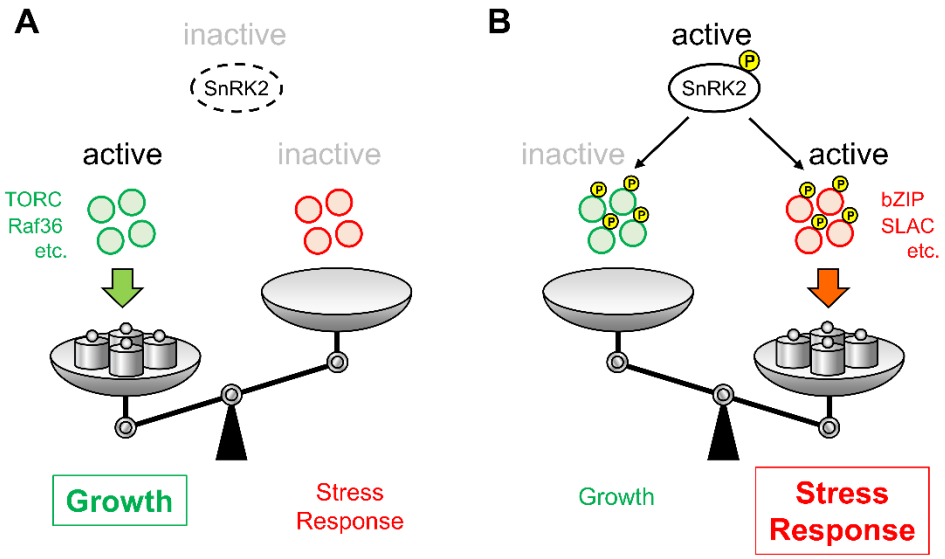

Figure 12. SnRK2s regulate growth and stress response under drought.

A. Under normal conditions, plants use energy to grow, and TORC or Raf36 are involved in this process.

B. Under drought conditions, SnRK2s are activated and phosphorylate substrates, e.g. basic leucine-zipper (bZIP) transcription factors or slow anion channels (SLAC), to induce stress responses. SnRK2s also phosphorylates TORC or Raf36 to inhibit plant growth.

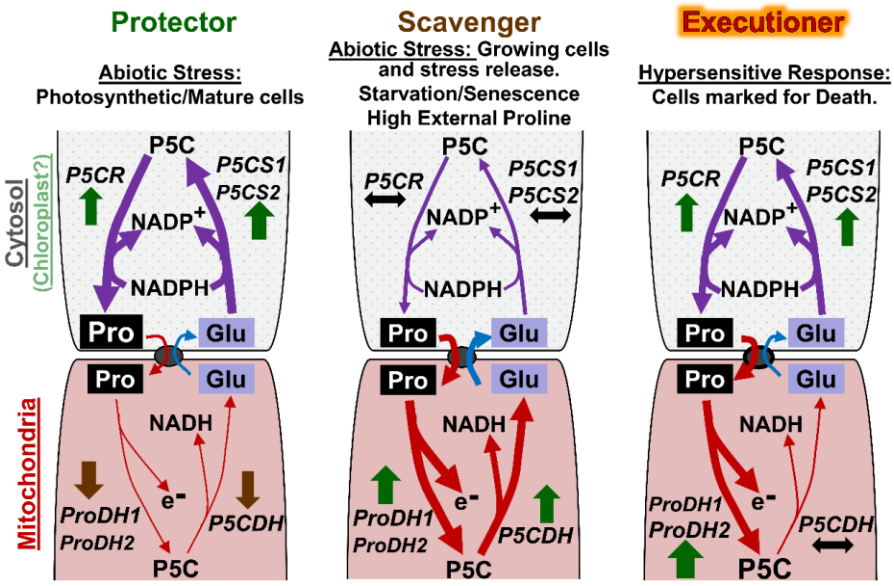

Figure 13. The protector, scavenger and executioner modes of proline metabolism and the conditions or tissue in which they are observed.

Up and down arrows (Red, brown or grey) indicate changes in gene expression compared to unstressed control. In most cases where data is available, protein levels of the corresponding enzyme change in the same direction. Thickness of the red or purple arrows represents predicted relative metabolic flux through different steps in proline metabolism. Abiotic stress refers to drought, freezing or high salt treatments where sustained accumulation of free proline is observed in many plant species.

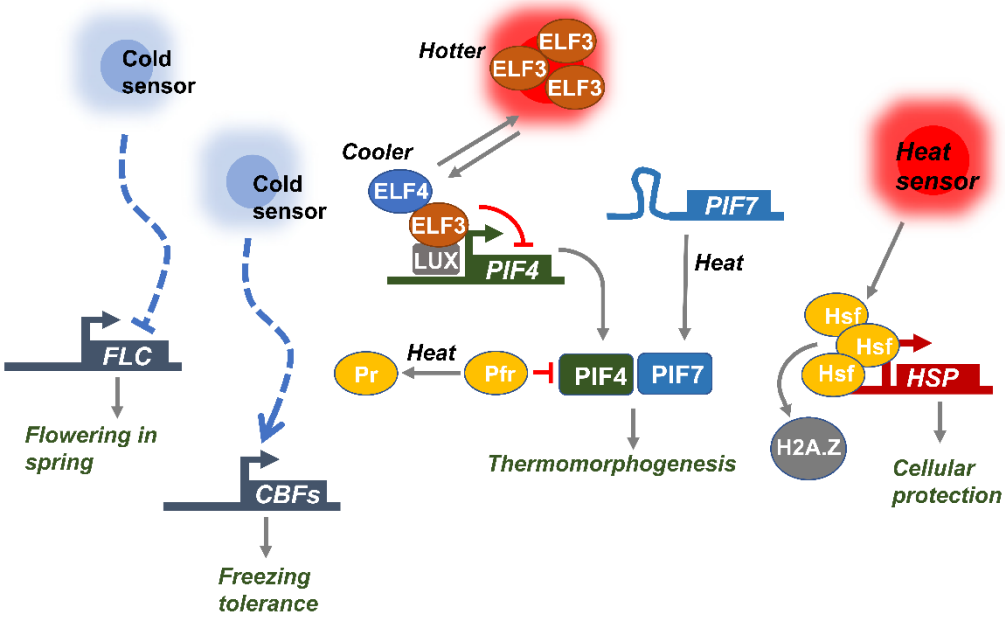

Figure 14: Temperature response mechanisms.
Temperature controls many key adaptive traits in plants such as freezing tolerance, growth, flowering and heat stress protection. Vernalization results in the stable repression of expression of the floral repressor *FLC*. Thermomorphogenesis integrates temperature information from at least three distinct biophysical responses: protein phase separation of ELF3, thermal reversion of phyB (Pfr to Pr) and enhanced translation of *PIF7* in warm conditions. Heat stress also induces the induction of protective heat shock proteins (HSP) via the activation of heat shock factors (Hsf) via an unknown mechanism.