

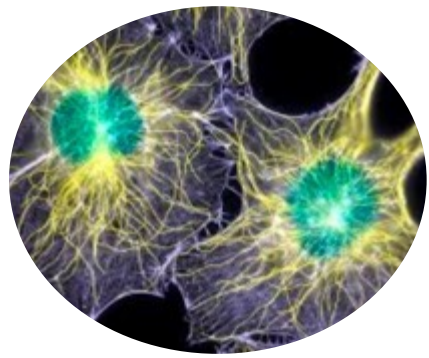
Fisiologia Celular e Molecular

Mestrado em Biologia Molecular e Genética

2º Semestre

6 ECTS

Aula T3 – 23 Fev 2023



Fisiologia Celular e Molecular

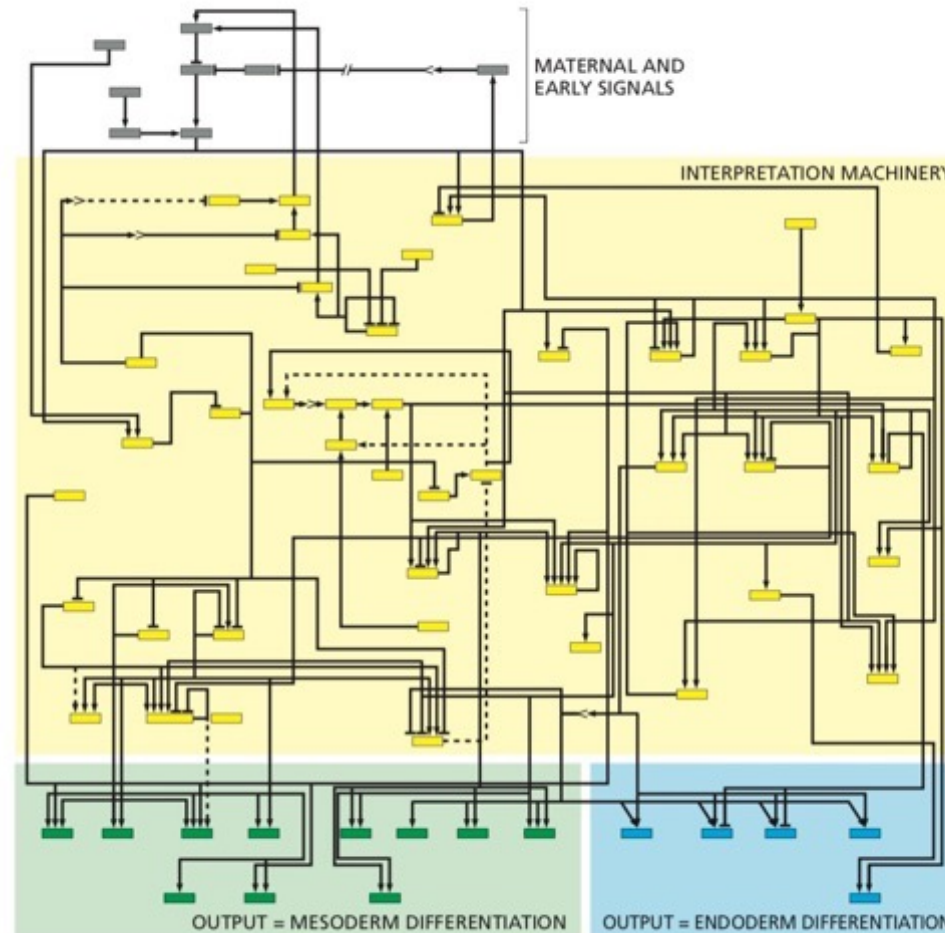
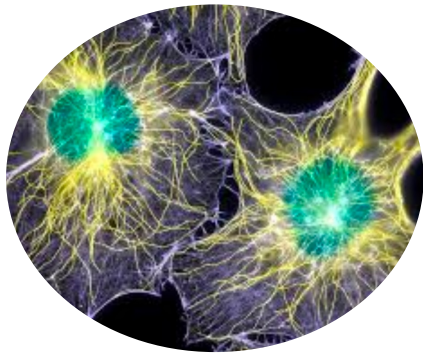


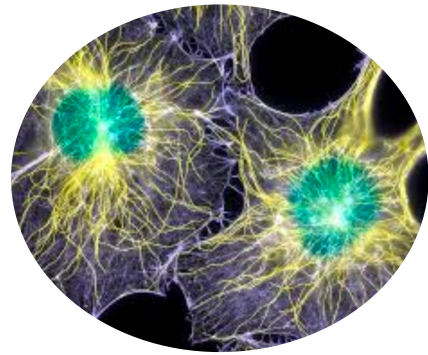
Figure 7-42 The exceedingly complex gene circuit that specifies a portion of the developing sea urchin embryo. Each colored small box represents a different gene. Those in yellow code for transcription regulators and those in green and blue code for proteins that give cells of the mesoderm and endoderm, respectively, their specialized characteristics. Genes depicted in gray are largely active in the mother and provide the egg with cues needed for proper development. As in Figure 7-40, arrows depict instances in which a transcription regulator activates the transcription of another gene. Lines ending in bars indicate examples of gene repression. (From I.S. Peter and E.H. Davidson, *Nature* 474:635-639, 2011. With permission from Macmillan Publishers Ltd.)



The enigma of environmental pH sensing in plants

Huei-Hsuan Tsai ¹ and Wolfgang Schmidt ^{1,2,3} 

Environmental pH is a critical parameter for innumerable chemical reactions, myriad biological processes and all forms of life. The mechanisms that underlie the perception of external pH (pH_e) have been elucidated in detail for bacteria, fungi and mammalian cells; however, little information is available on whether and, if so, how pH_e is perceived by plants. This is particularly surprising since hydrogen ion activity of the substrate is of paramount significance for plants, governing the availability of mineral nutrients, the structure of the soil microbiome and the composition of natural plant communities. Rapid changes in soil pH require constant readjustment of nutrient acquisition strategies, which is associated with dynamic alterations in gene expression. Referring to observations made in diverse experimental set-ups that unambiguously show that pH_e per se affects gene expression, we hypothesize that sensing of pH_e in plants is mandatory to prioritize responses to various simultaneously received environmental cues.



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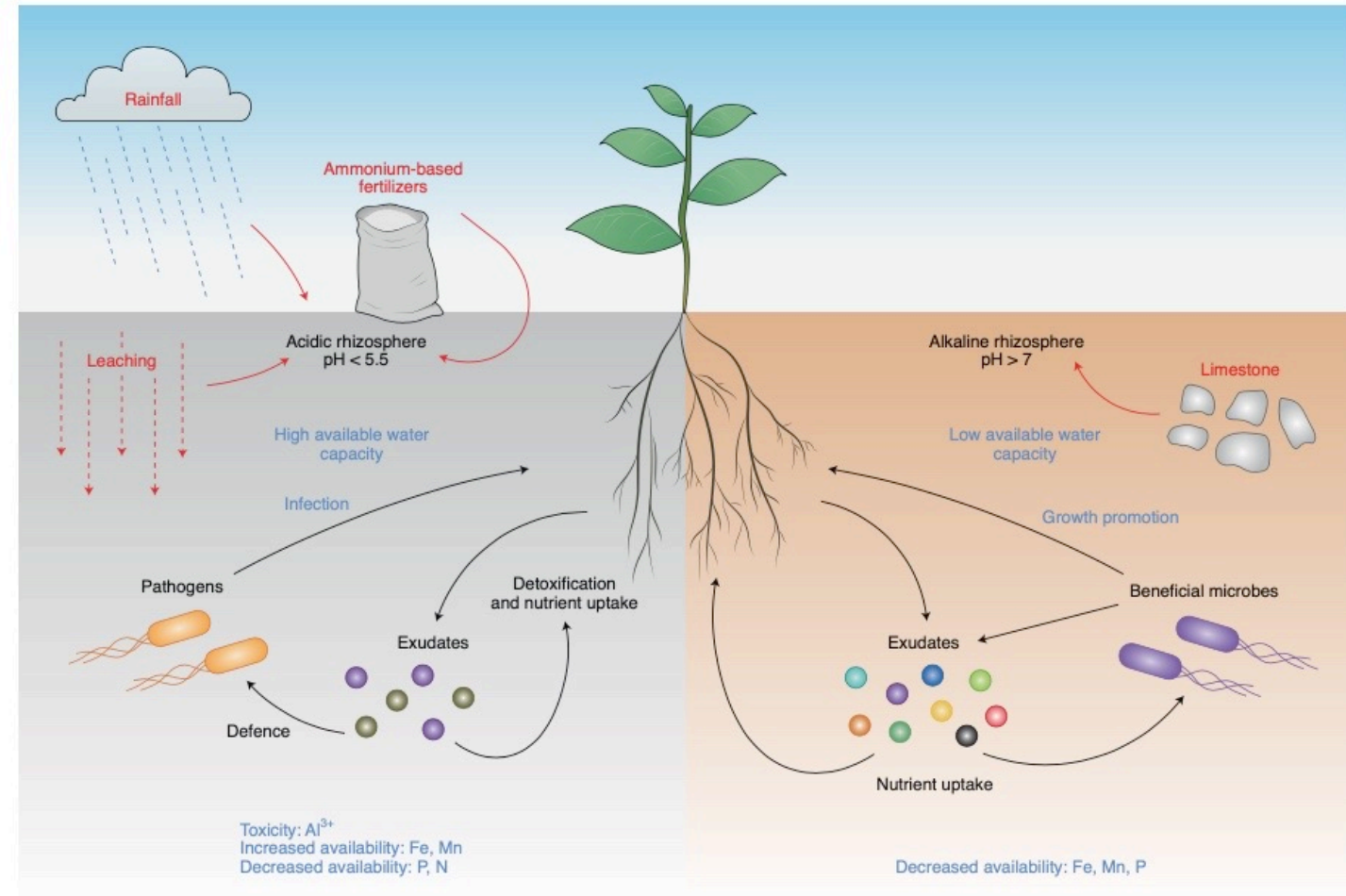
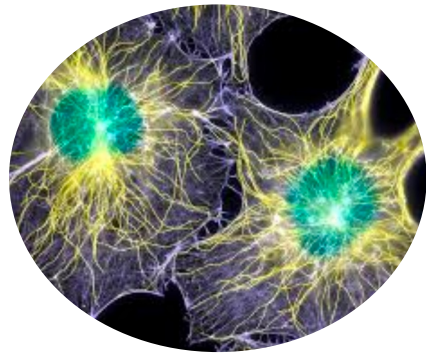


Fig. 1 | Factors that determine soil pH and effects of soil pH on plants. Soil pH-determining factors are indicated in red text, while effects of soil pH on plants are denoted in blue text. The main factors that cause soil acidity are leaching of alkaline elements by excessive rainfall and ammonium-based fertilizers. Major limitations of plant growth in acid soils are caused by pathogen infection, aluminium (Al^{3+}) toxicity, potentially toxic levels of Mn and Fe and decreased availability of P and N. Alkaline soils are mostly caused by limestone sediments and affect plant growth by low available water capacity and decreased availability of Fe, Mn and P. Changes in soil pH are accompanied by shifts in microbiome structure; beneficial soil microbes prefer neutral or mildly alkaline pH conditions, while lower soil pH alters the microbiome population to a more pathogen-dominated population. To cope with constraints associated with different soil pH environments, plants alter the compositions of root exudates to improve, tune or prioritize detoxification processes, and adapt nutrient-acquisition strategies and pathogen-defence responses.

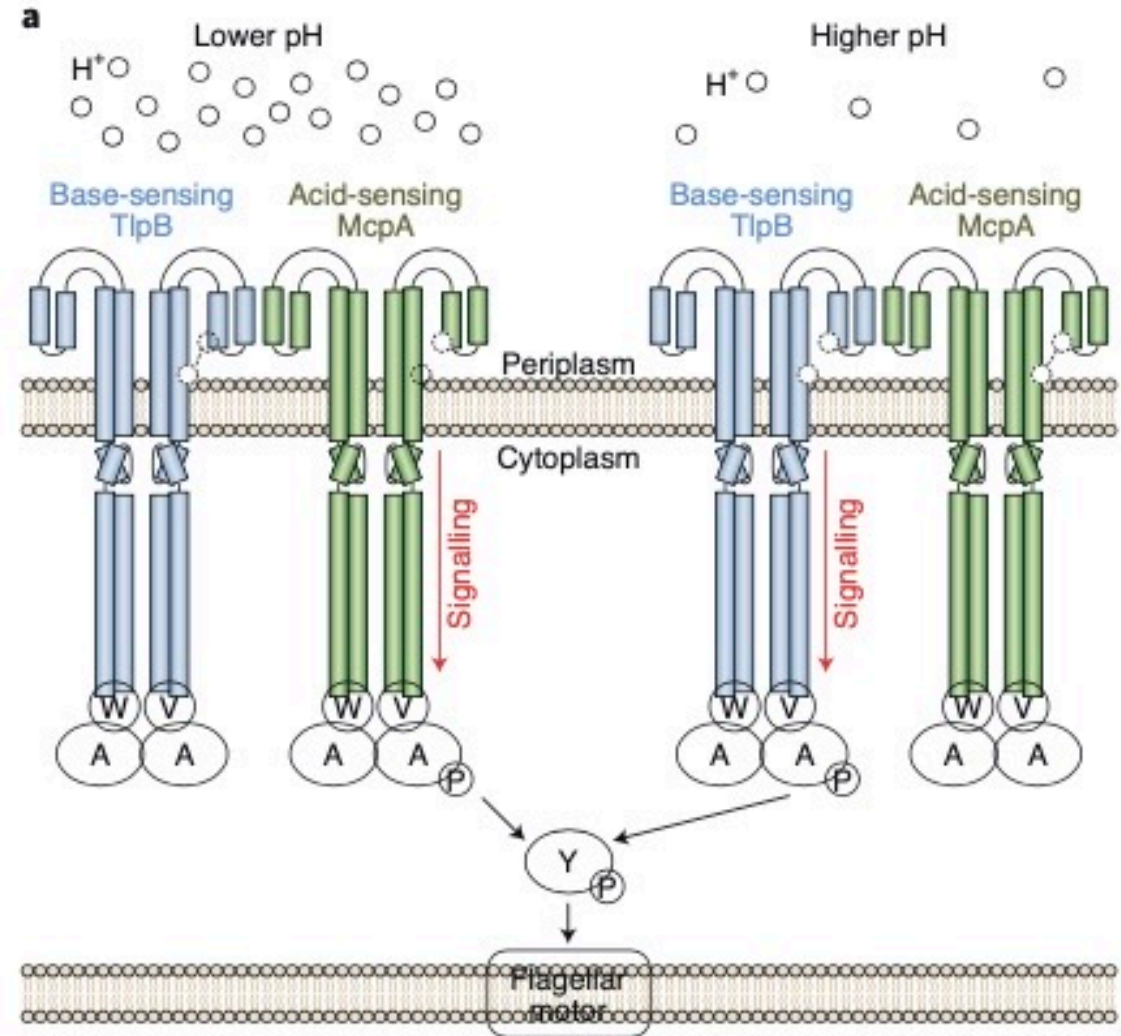


External pH sensing

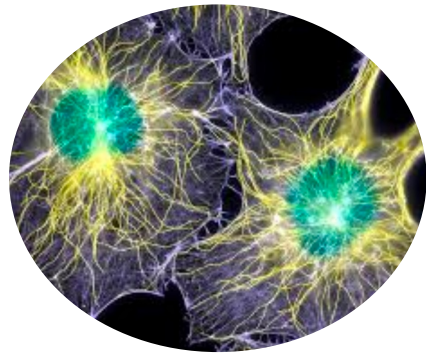
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Fig. 2 | pH sensing in bacteria. **a**, Proposed model for pH sensing and signalling in the soil bacterium *B. subtilis*. At low pH conditions, two ionizable residues on the base-sensing chemoreceptor TlpB are protonated (Lys and/or Glu; red circle) and form stable hydrogen bonds with two adjacent residues (Gln and/or Asp; white circle). As the pH increases, the residues become deprotonated, resulting in the loss of hydrogen bonding and destabilization of the local structure. At a higher pH, a key histidine residue on the acid-sensing receptor McpA is in a neutral, non-protonated state (lower white circle), forming hydrogen bonds with adjacent residues (Thr and/or Gln; upper white circle). As the pH decreases, the histidine residue becomes protonated (red circle), resulting in the loss of hydrogen bonding and destabilization of the local structure. For both TlpB and McpA, structural destabilization induces signalling through autophosphorylation of the histidine kinase CheA (A). Phosphate from CheA is subsequently transferred to CheY (Y), which then interacts with the flagellar motor to navigate *B. subtilis* towards more favourable pH conditions. P, phosphate group; V, CheV; W, CheW. **b**, Proposed push-pull pH taxis mechanism in *E. coli*. At low pH conditions, reduced methylation (indicated by dark grey circles) renders the serine receptor Tsr (blue) more sensitive to pH changes, navigating cells to more alkaline conditions. An increase in pH is sensed as an attractant stimulus by Tsr but as a repellent stimulus by the aspartate receptor Tar (green), leading to increased methylation of Tsr and decreased methylation of Tar. At high external pH, Tar is less methylated and navigates cells to more acidic conditions. **c**, The CadC-mediated signalling system of *E. coli* for sensing low external pH. After sensing of acidic pH, CadC undergoes a structural rearrangement that enables the cytoplasmic CadC DNA-binding domains to dimerize and activate the expression of the *cadBA* operon. *cadA* encodes a cytoplasmic lysine decarboxylase that increases cytoplasmic pH by producing the alkaline compound cadaverine and carbon dioxide at the expense of lysine and H⁺. *cadB* encodes a membrane-integrated lysine/cadaverine antiporter that is responsible for exporting cadaverine to the periplasmic space to increase the extracellular pH. Panel **a** adapted with permission from ref. ⁸, American Society for Microbiology. Panel **b** adapted with permission from ref. ⁹, Wiley.



Bacillus subtilis



External pH sensing in bacteria

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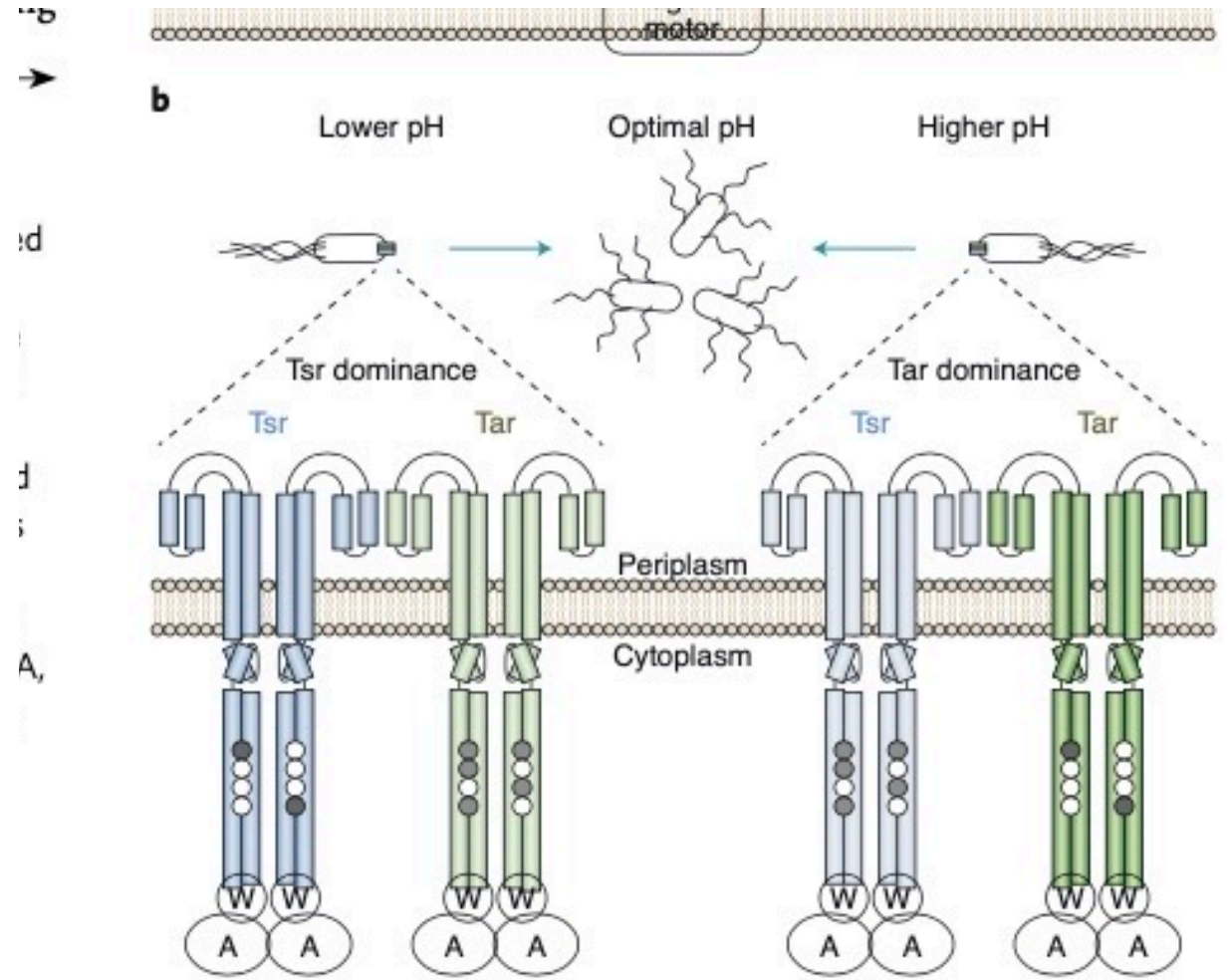
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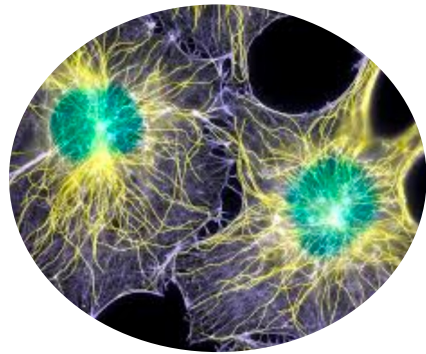
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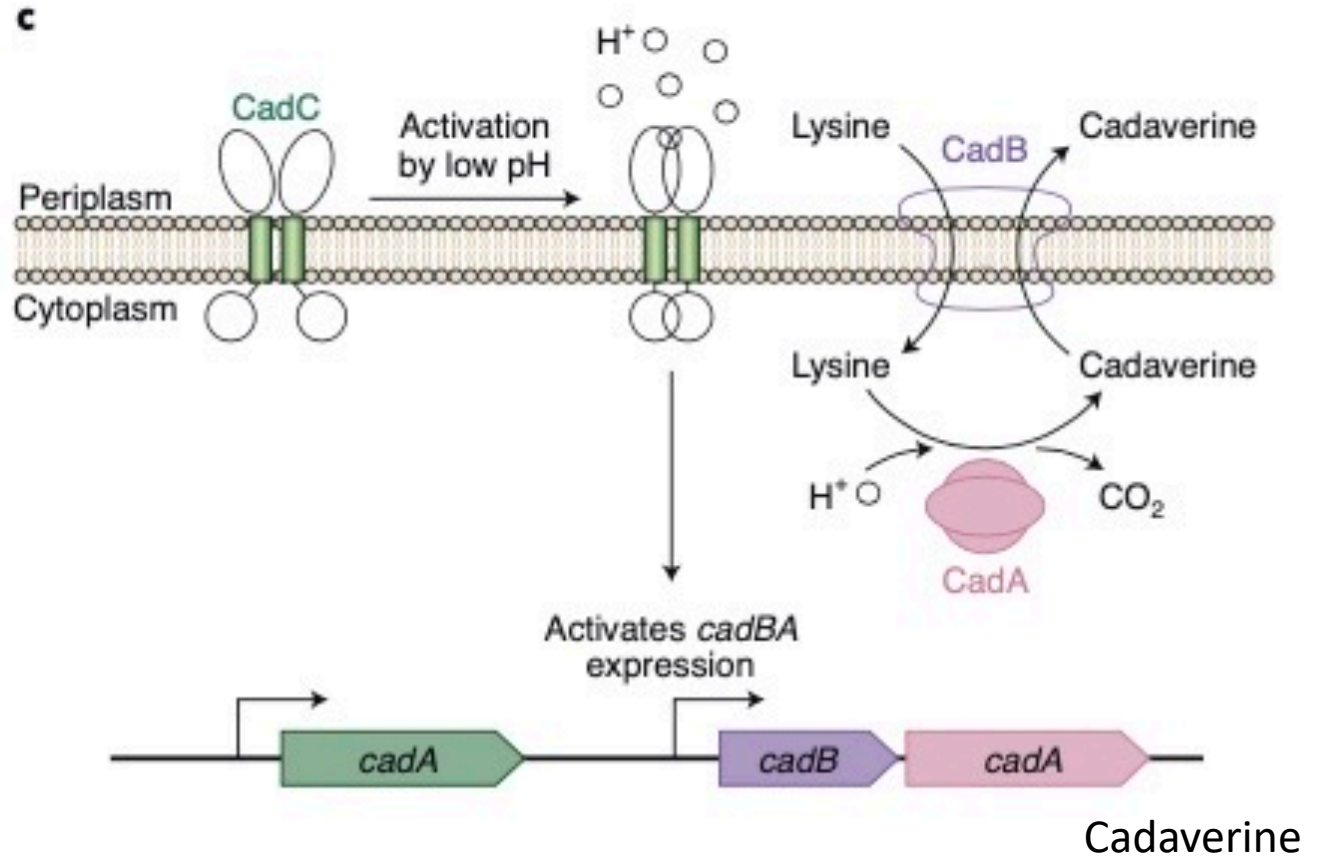


Escherichia coli

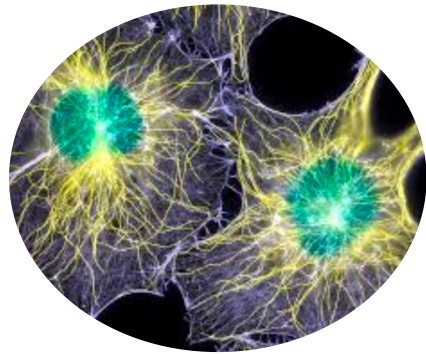


External pH sensing in bacteria

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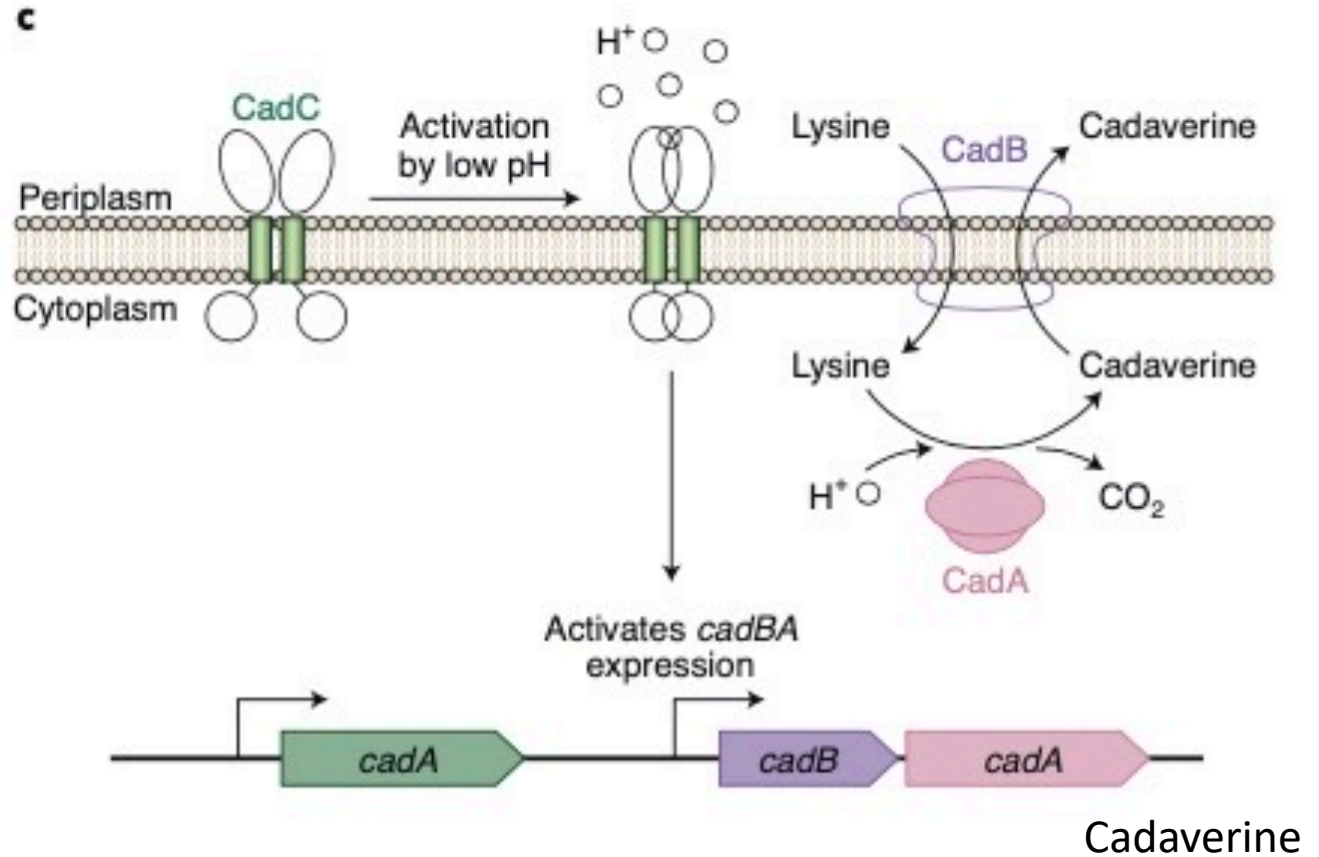


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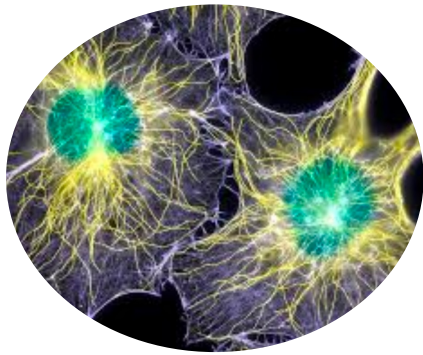


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External pH sensing in Eukaryotes – acid sensing ion channels

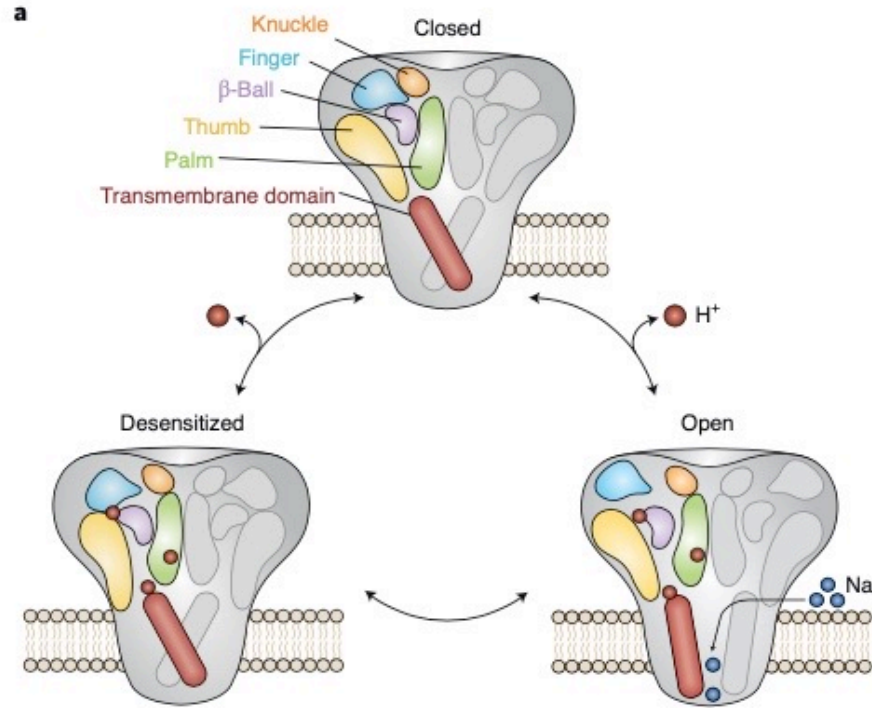


Fig. 3 | pH sensing in eukaryotes. **a**, ASIC gating in mammalian cells. Schematic view of an ASIC trimer showing the domains of one of the three subunits in the closed, open and desensitized states. In the closed state, the thumb is pushed away from the β -ball and finger domains, keeping the transmembrane domain in a constricted conformation that closes the pore. Following extracellular acidification, several domains become protonated (red circles), leading to structural rearrangements that open the pore to allow transient permeation of sodium ions (Na^+ ; blue circles). Desensitization of the channel occurs after continued exposure to protons, leading to a constricted conformation of the transmembrane domain that resembles the closed state. **b**, Rim-pathway-mediated pH sensing and signalling in *S. cerevisiae*. External pH is sensed by a PM complex consisting of the sensing components Rim21 and Dfg16, the chaperone Rim9 and the α -arrestin Rim8. In response to a neutral or alkaline pH signal, Rim8 binds to the cytoplasmic tail of Rim21 and Dfg16, which promotes the recruitment and assembly of ESCRTs to form a scaffolding platform for the Rim proteolysis complex (Rim23, Rim20 and the protease Rim13). Rim13 catalyses the pH-dependent cleavage and activation of the transcription factor Rim101. Once activated, cleaved Rim101 localizes to the nucleus to regulate gene expression required for the adaptation to alkaline pH. Panel **a** adapted with permission from ref. ³⁵, Elsevier.

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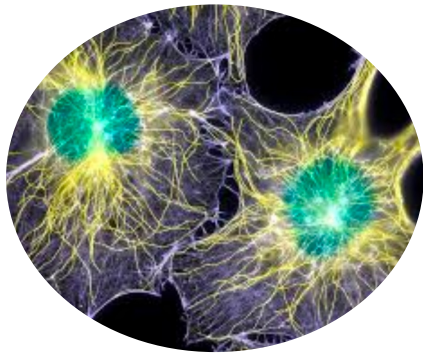
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The enigma of environmental pH sensing in plants

Huei-Hsuan Tsai¹ and Wolfgang Schmidt^{1,2,3}✉

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External pH sensing in Eukaryotes

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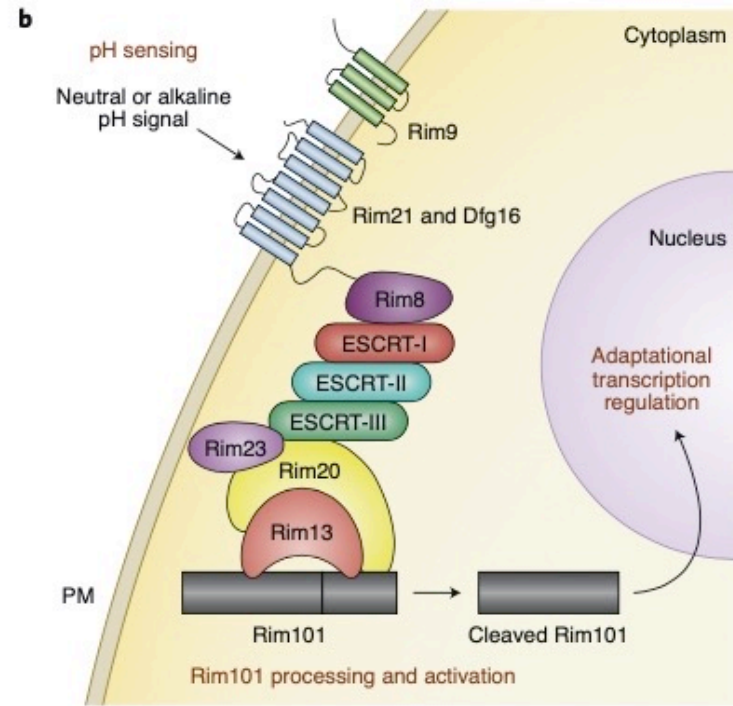
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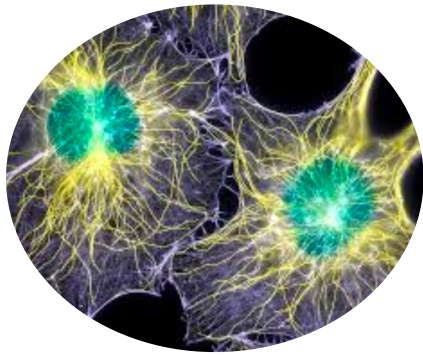
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Saccharomyces cerevisiae

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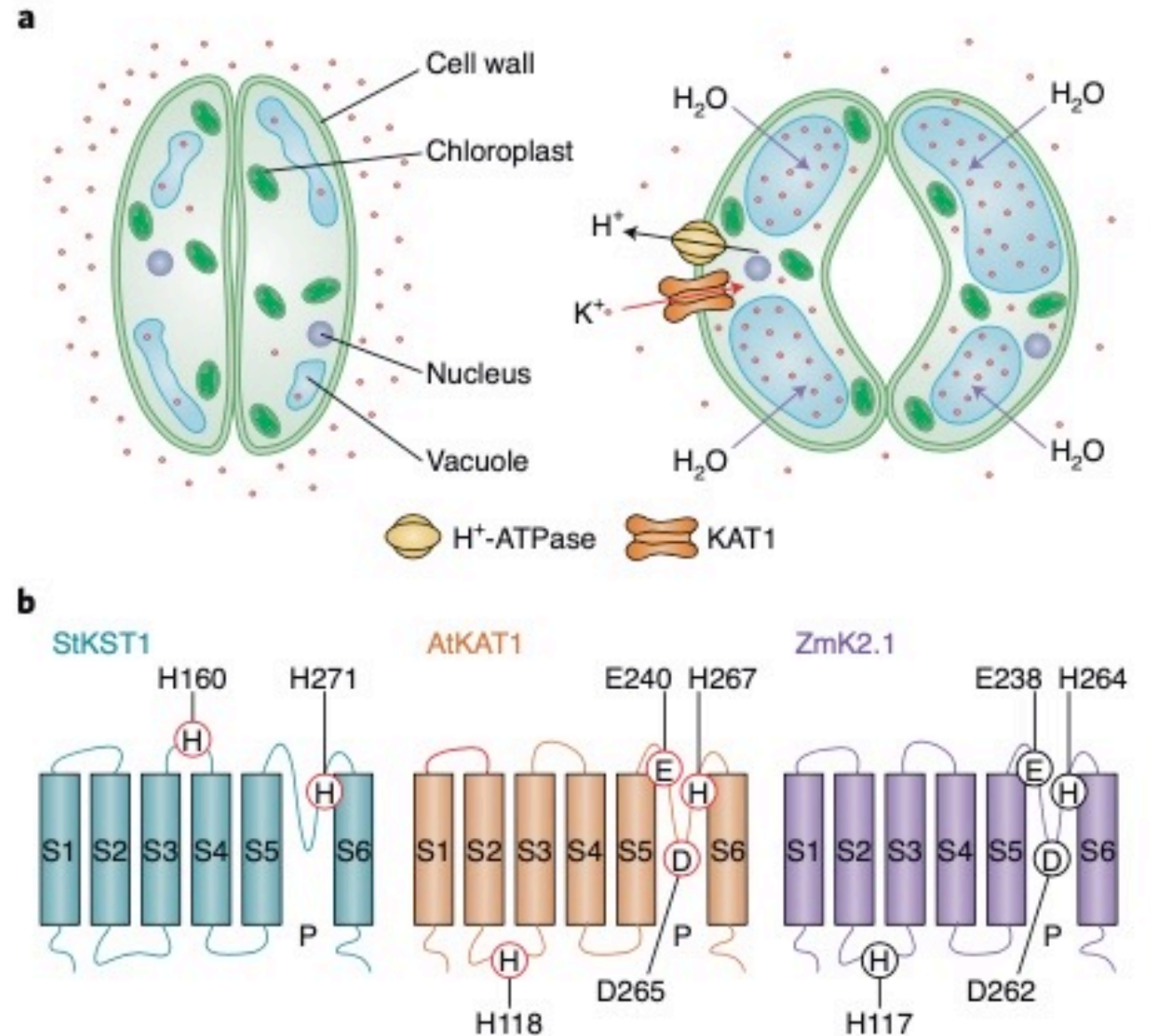
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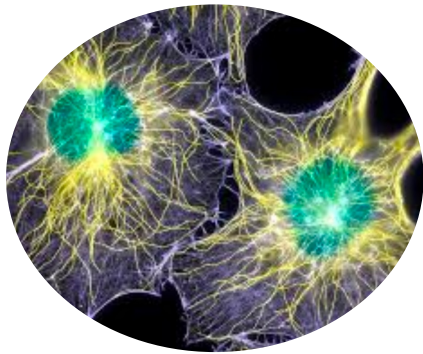
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Fig. 4 | Acid-activated K_{IR} channels in plants. **a**, Role of K_{IR} channels in stomatal guard cells. Stomatal opening is facilitated by H^+ -ATPase-mediated extracellular acidification and the subsequent activation of pH sensors on KAT-type K^+ channels that leads to K^+ influx, increase in turgor pressure and opening of the stomatal pore. KAT1 is shown as an example. **b**, General structure of the voltage-dependent K^+ channels StKST1, AtKAT1 and ZmK2.1. Transmembrane segments are denoted as S1–S6, P indicates the pore region. Amino acid residues and linkers that contribute to pH sensitivity are indicated by red circles/line for StKST1 and AtKAT1. Important residues in AtKAT1 are conserved in the maize homologue ZmK2.1, but despite sequence and structural homologies, ZmK2.1 is not activated by acidification. Instead, the S1–S2 linker appears to be the determining factor that distinguishes pH sensitivity between AtKAT1 and ZmK2.1.





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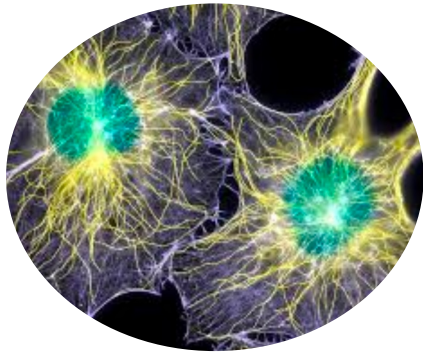
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The enigma of environmental pH sensing in plants

Huei-Hsuan Tsai ¹ and Wolfgang Schmidt ^{1,2,3} 

Here, we summarize evidence that conspicuously suggests that gene expression in plants is directly affected by pH_e , and put forward the hypothesis that plants possess means to perceive and effectively respond to alterations in pH_e .



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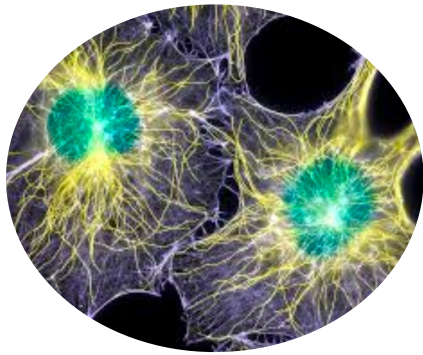
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Isto é o objectivo de todos os trabalhos
que vamos desenvolver em FCM