



Drought stress and desiccation tolerant plants

**Dorothea Bartels
Institute of Molecular Plant Physiology
and Biotechnology of Plants
University of Bonn
Germany**

Outline

Introduction: Desiccation tolerant resurrection plants

Desiccation tolerance in seeds versus vegetative tissues

LEA genes: expression profiles and promoter *cis*-elements

ELIP genes: expression and protective functions

Possible model for stress perception

Oropetium thomaeum



Fully hydrated



desiccated



rehydrated

Plants were originally collected in Jodhpur, Rajasthan, India close to a riverbed

Metabolic arrest in vegetative tissues of the desiccation tolerant resurrection plant *Craterostigma plantagineum*



Fully turgid plant



Desiccated plant



Rehydrated plant

Not only the induction of anhydrobiosis demands a specific genetic programme but also the recovery/rehydration

Severe (C.p.) and intermediate phenotype L.b. also reflected in physiology



C. plantagineum
Savannah



L. subracemosa
Marshy areas



L. brevidens
Rainforest



tolerant

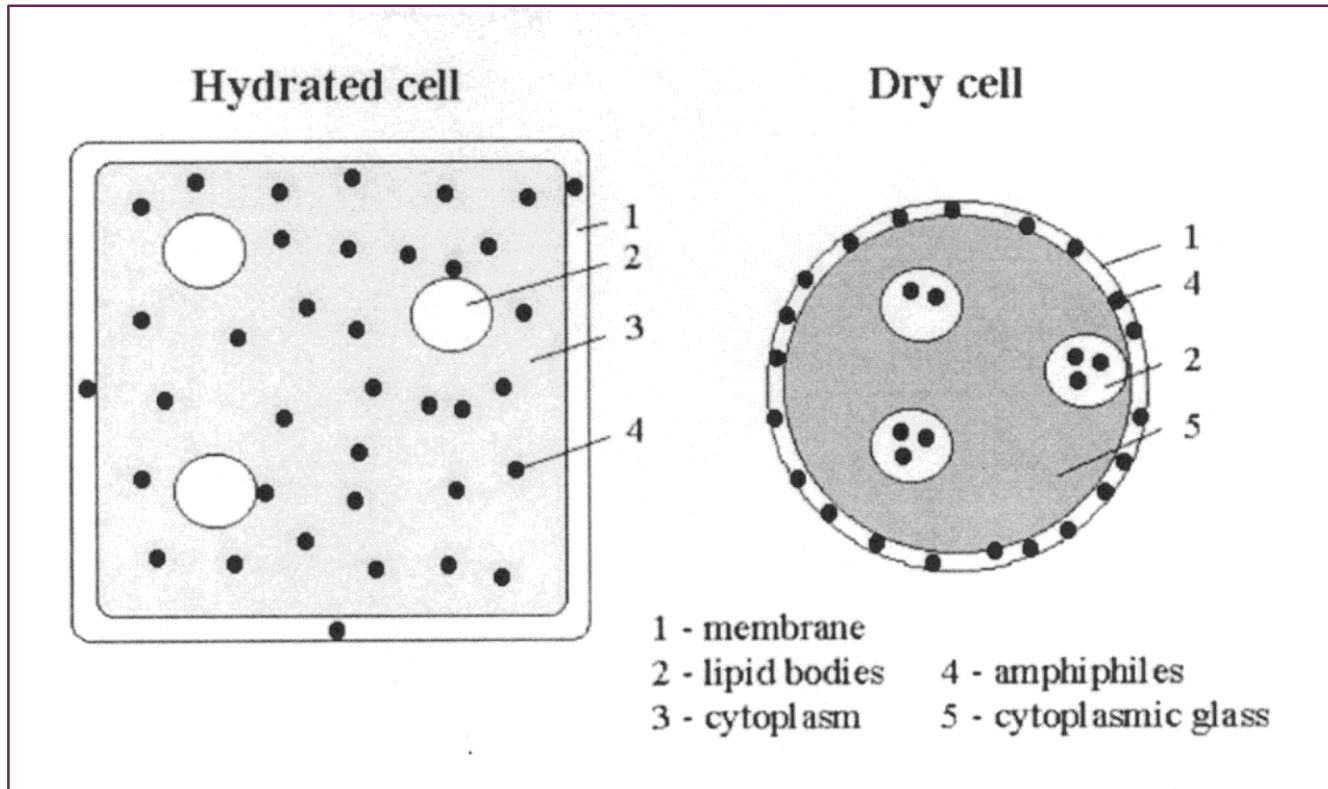


sensitive



tolerant

Comparison of a hydrated and a dry cell



Cell shrinkage causes crowding of cytoplasmic components and the cell contents become increasingly viscous, enhancing the probability for molecular interactions that can cause protein denaturation and fusion of apposed membranes

***Craterostigma plantagineum* a desiccation tolerant homiochlorophyllous resurrection plant**

Changes during desiccation indicate complexity of the phenomenon



Carbohydrate metabolism

Octulose /Sucrose low

Sucrose (very high)

Protein metabolism

Stress Proteins(LEA proteins, photoprotective proteins (very high)

Cellular structures

**Protoplast shrinkage, mechanical tension
cell wall, cellular surface**

Many data available which describe the changes which take place during dehydration/rehydration

Acquisitions of seed specific pathways are major determinants of desiccation tolerance in vegetative tissues of resurrection plants

Questions:

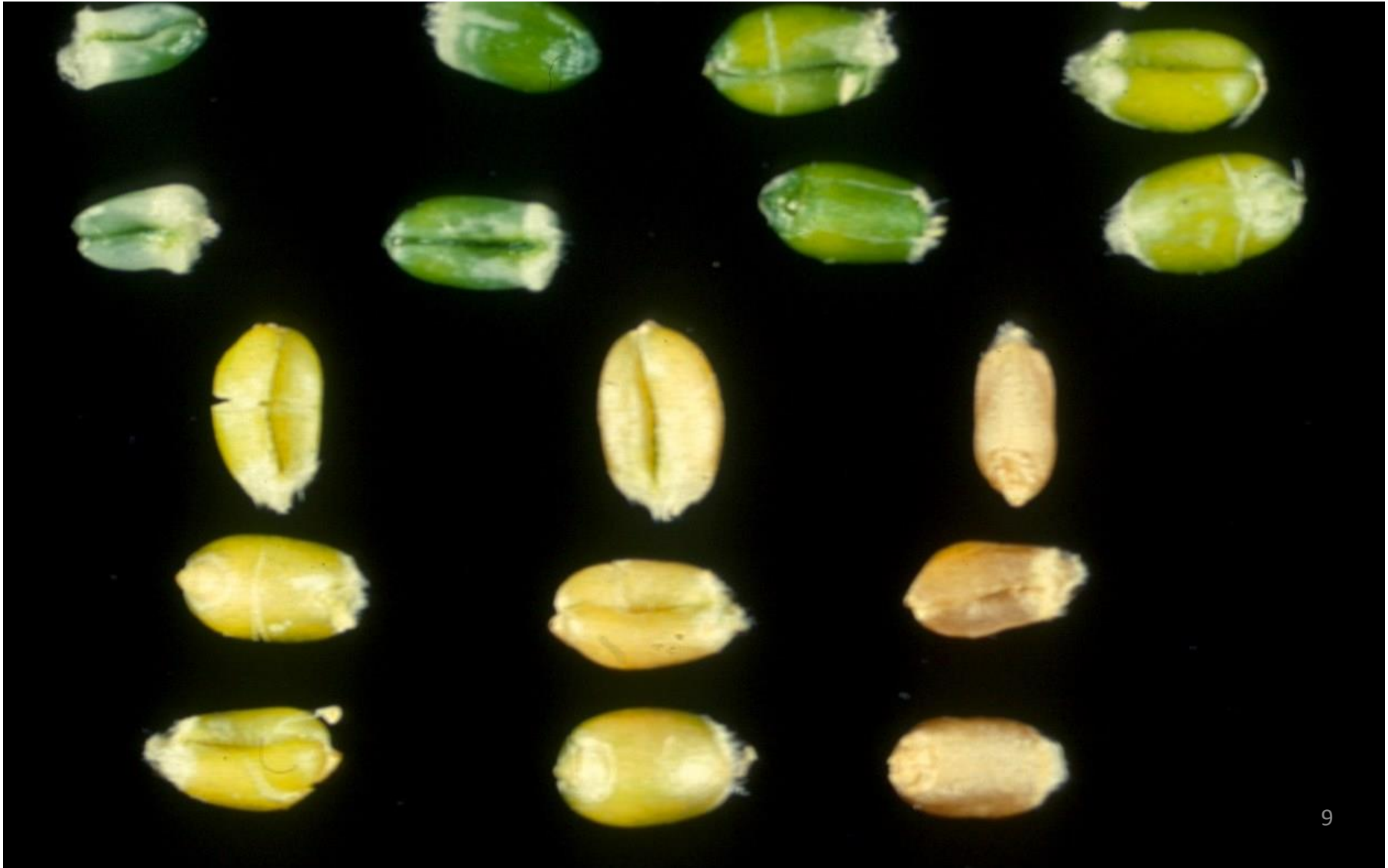
Which are the essential evolutionary adaptations which allow the targeted expression of stress pathways?

Which are the triggers?

Most higher plants exhibit desiccation tolerance when they form seeds

How do seeds prepare for metabolic arrest?

The phytohormone ABA regulates seed maturation: Synthesis of „Late embryogenesis abundant“ (LEA) proteins, induction of dormancy



Comparison of desiccation tolerance in seeds and resurrection plants

Many pathways, responses on the cellular Metabolites and regulatory features in common seeds and vegetative tissues

Differences ?

Photosynthesis

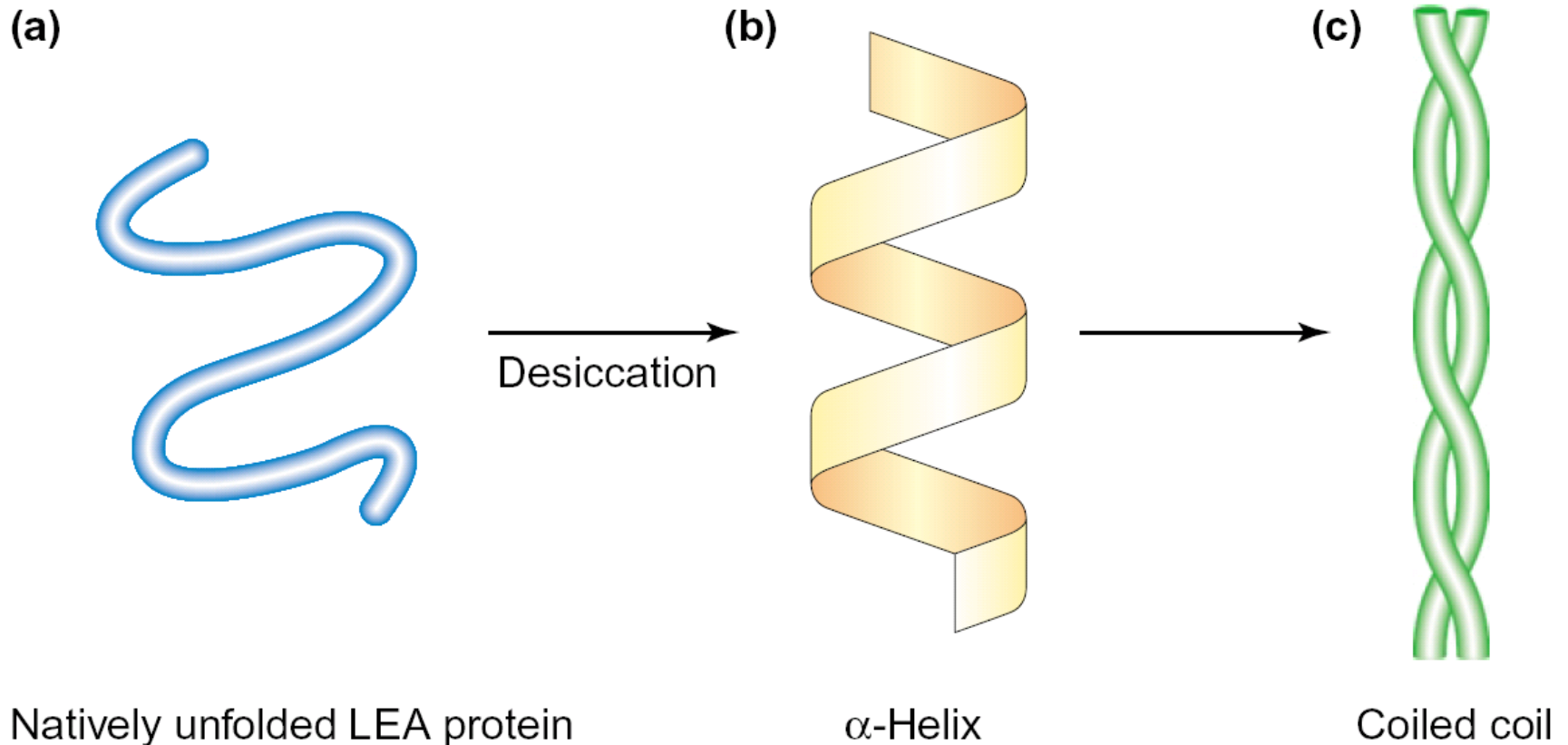
**Two types of resurrection plants homiochlorophyllous
(retain chlorophyll and
poikilochlorophyllous (degrade chlorophyll))**

LEA Proteins

Major components of seeds and desiccated vegetative tissues tissues of resurrection plants

**LEA Proteins change their structure according to water availability
They are unstructured in the presence of water, very hydrophilic**

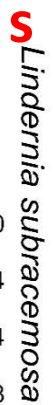
What is the role of the structural changes?



Michael J. Wise and Alan Tunnacliffe (2004). *Trends Plant Sci.* 9, 13-17

Comparison of *LEA* genes in *L. brevidens* and *L. subracemosa*

- Classify *LEA* genes in both species
- Identify homologous/corresponding genes via sequence comparisons in both species
- Organization within the genomes
- Abundancies of expression in 7 stages during dehydration/rehydration using qPCR



Hints from comparative transcriptomics and genomics

C. plantagineum, *L. brevidens* - *L. subracemosa*

Selective LEA genes are more abundantly up-regulated in

C. plantagineum, *L. brevidens* than in *L. subracemosa*

Subsets are induced by dehydration in all three species

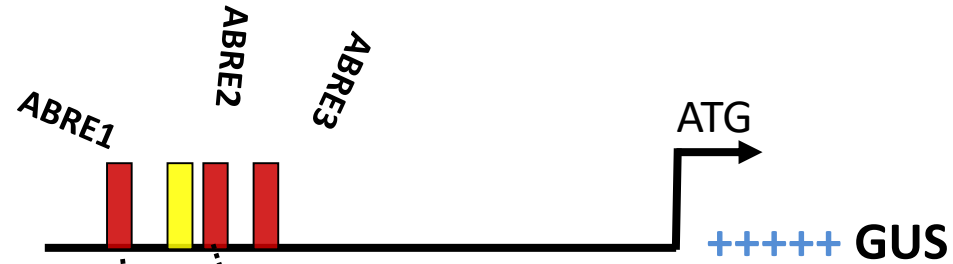
**A way to obtain selective gene expression
modulation/selection of promoter *cis* elements**

**This seems to be realized in promoters of some
LEA genes**

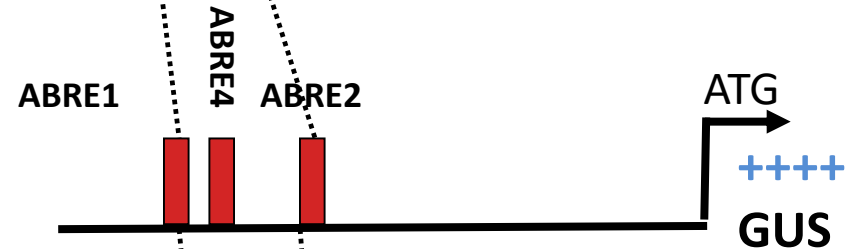
Promoter architecture of 11-24 genes in Lindernia species

ABRE and DRE elements influence promoter strength in response to stress

Cp LEA-like 11-24 promoter fragment
Craterostigma plantagineum



Lb LEA-like 11-24 promoter fragment
Lindernia brevidens



Ls LEA-like 11-24 promoter fragment
Lindernia subracemosa



■ = ACGT-box ■ = DRE

100 bp

Conclusions from comparative promoter analysis

Conserved promoter elements (ABRE, DRE) determine promoter activity

Plant (*C.plantagineum*) which experiences desiccation tolerance in its life cycle has the strongest promoter: relevance of DRE element

L.brevidens has elements to transcriptionally activate the gene but lower activity

L. subracemosa (non- desiccation tolerant) lacks some promoter elements

Hypothesis:

Mutations are not random, selection driven by environment)

Monroe et al Nature (2022):Mutation bias reflects natural selection in *Arabidopsis thaliana*

Livnat et al: Sick Cell Mutation Driven by Pressure, not Random Chance

Protection of photosynthetic structures essential in homiochlorophyllous species

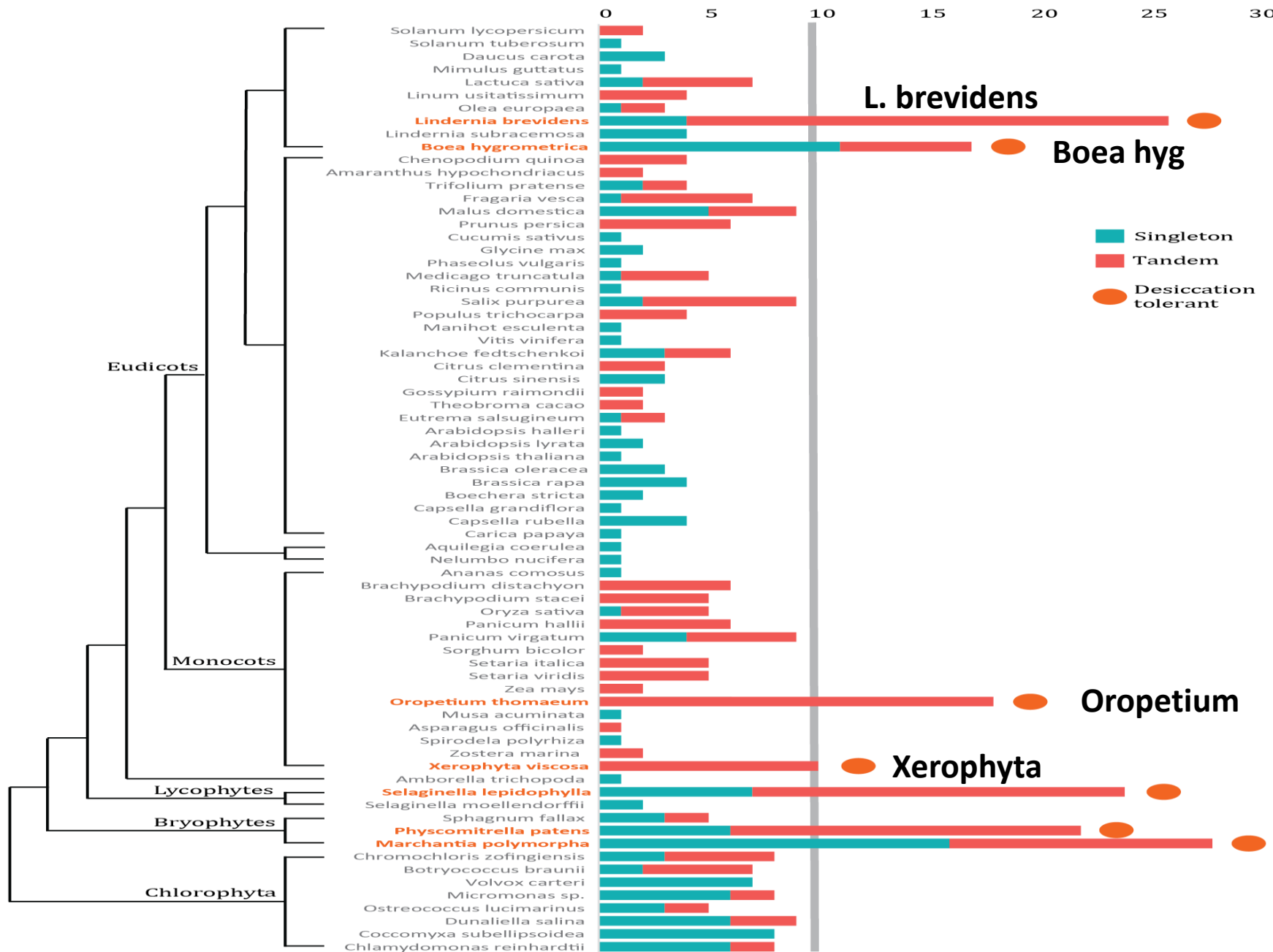
ELIP = Early-Light Induced
members of the light-harvesting complex protein superfamily
first time associated with dehydration in *C. plantagineum*

***ELIP* gene copies and expression patterns**

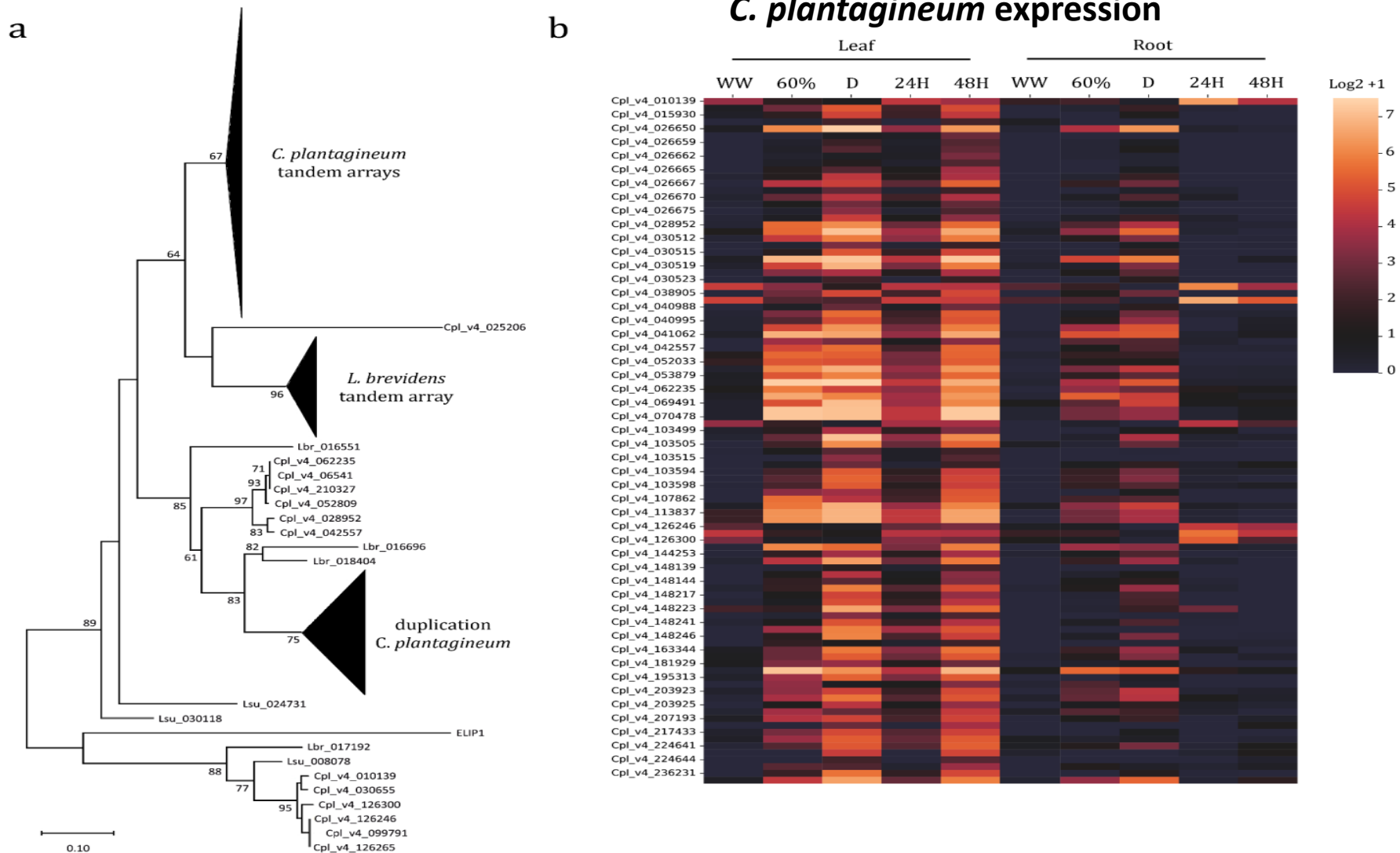
**Overrepresentation of ELIP transcripts in leaf tissues of
desiccation tolerant homiochlorophyllous plants**
(VanBuren et al. Plant Physiol 2019)

ELIP genes in desiccation sensitive and tolerant plants

Number of ELIPs



Independent tandem duplication of ELIPs in *L. brevidens* and *C. plantagineum*

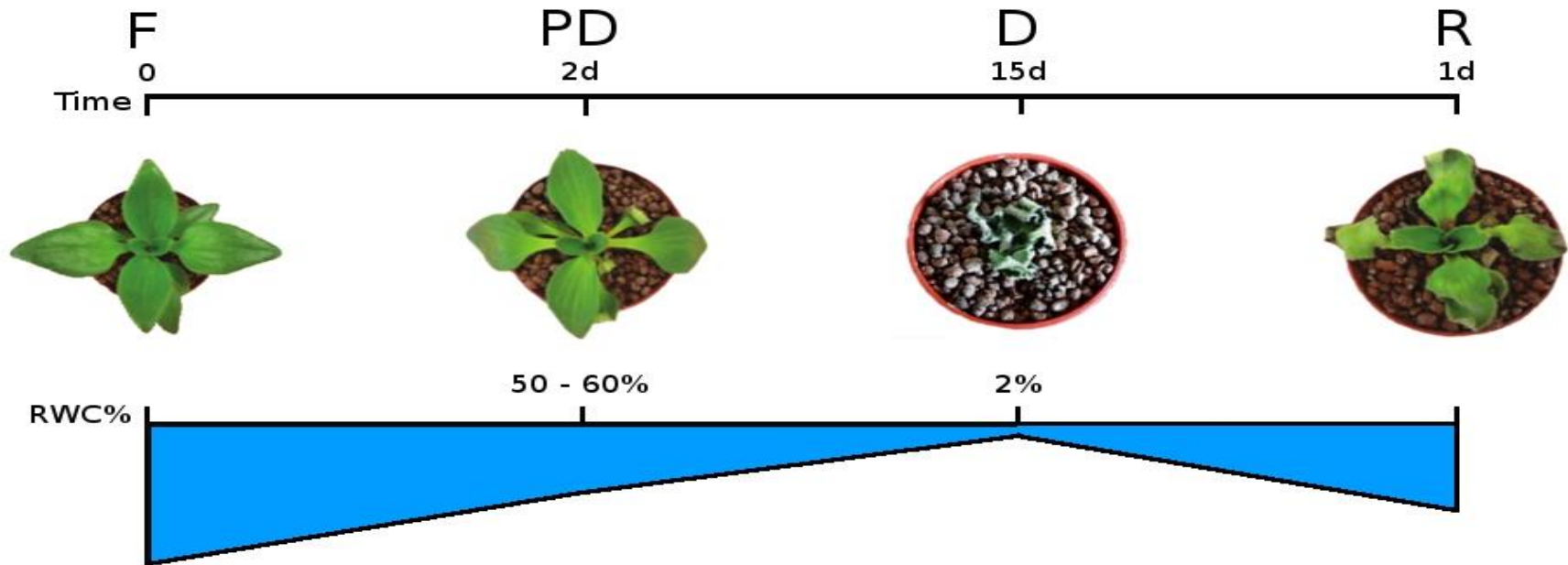


C. plantagineum: nearly 200 copies, *L. brevidens*: 186 copies, *L. subracemosa*: 4 copies

**Overrepresentation of ELIP transcripts in leaf tissues of
C. plantagineum, *L.brevidens* and other desiccation
tolerant species**

**Gene duplications contribute to higher abundancies in
desiccation tolerant species**

Deep transcriptome sequencing



Core network of desiccation-responsive genes across tissues
(protective functions and maintenance of structural requirements)

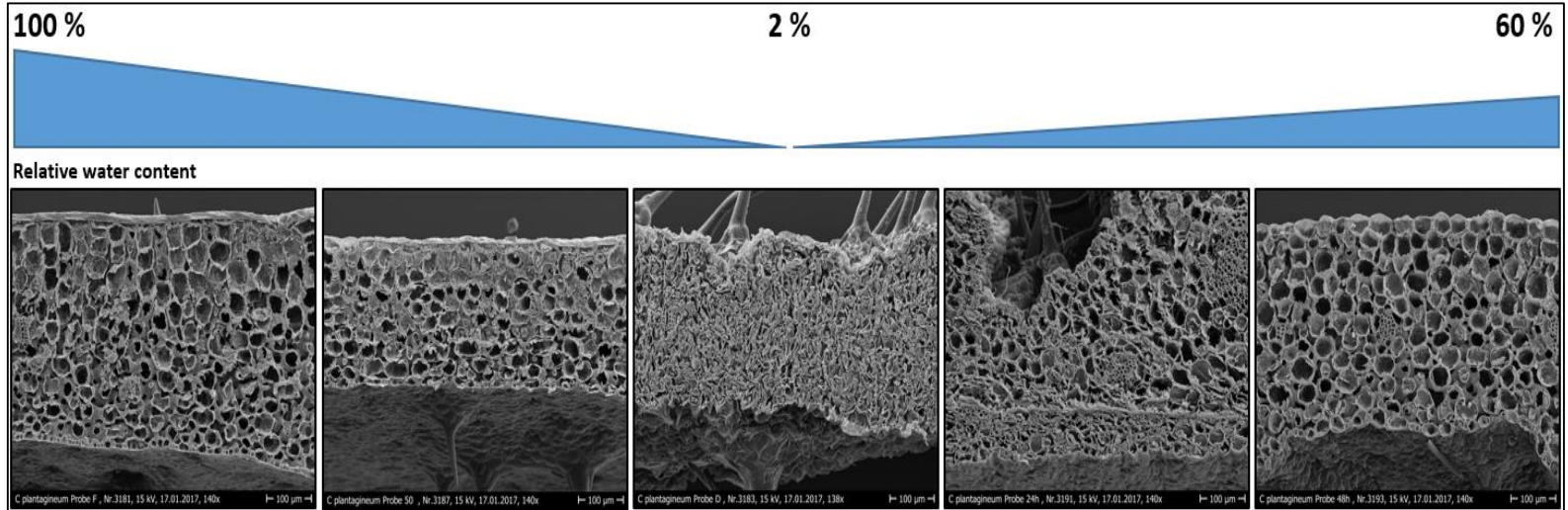
Secondary adaptations related to tissue functions and possibly habitat:
(facilitated by polyploidy)

Differences between roots and leaves: transport processes / photosynthesis
ABA signalling / photooxidation

VanBuren and colleagues

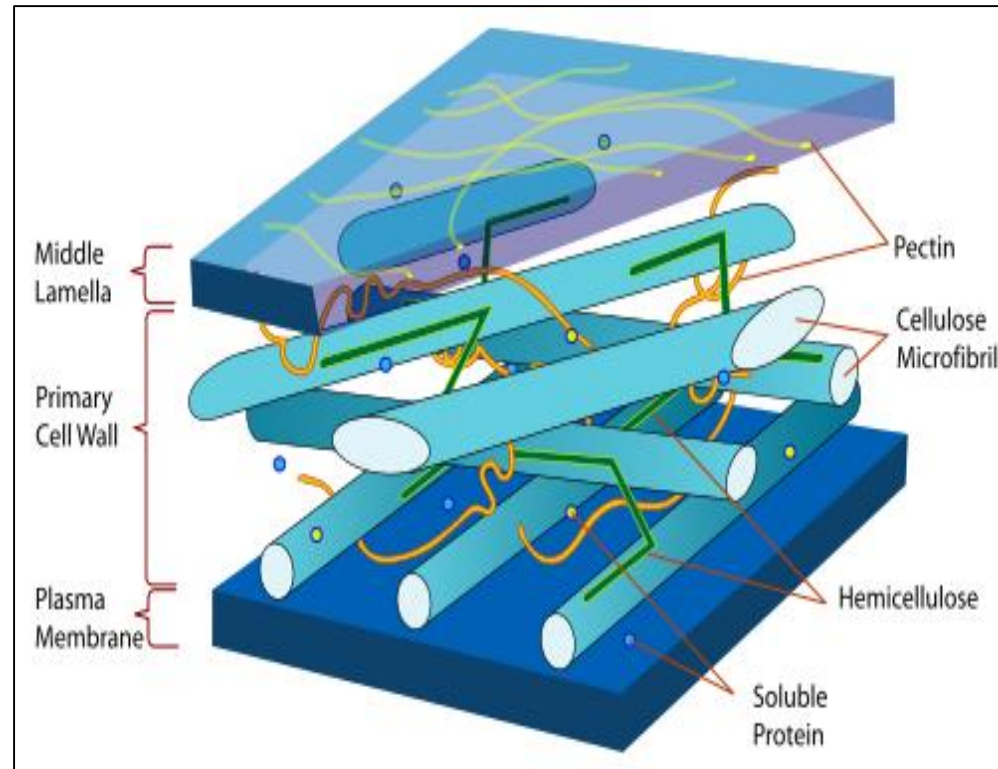
**In leaves cell wall modifications may be a trigger to prepare for
Desiccation tolerance**

Transverse sections



**Shrinkage of cells and curling of cell walls was observed.
48 h of rehydration are needed to restore the starting point**

Cell wall



Primary cell wall: cellulose, hemicellulose and pectin

Secondary cell wall: cellulose (35-50%), xylan (20-35%), lignin (10-25%)

Structural proteins (5%): hydroxyproline-rich glycoproteins(HRGP), arabinogalactan proteins (AGP), glycine-rich proteins (GRPs), and proline-rich proteins (PRPs)

Specific changes in pectin composition were found during dehydration and rehydration

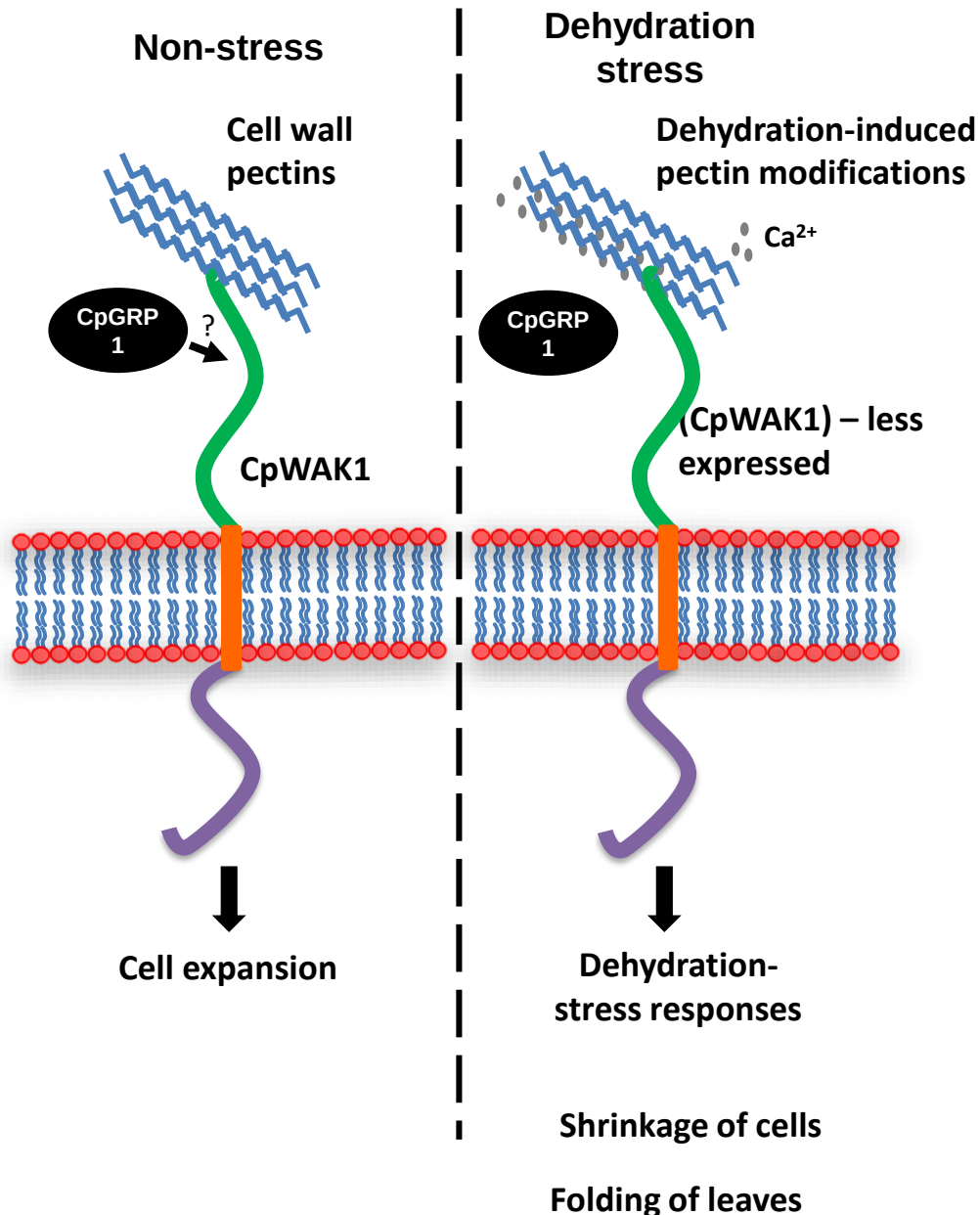
Do pectins interact with proteins?

Several cell wall-localized proteins were identified in *C. plantagineum* : Cell-wall associated kinase (CpWAK)

Glycine-rich protein (GRP1)

The expression of these proteins is modulated by dehydration/rehydration

CpGRP1/CpWAK1 model of stress perception



Non-stress

- CpWAK1 is bound to cell wall pectins and regulates cell expansion
- CpGRP1 binds to CpWAK1 and participates in growth regulation

Dehydration stress

- CpGRP1 together with pectin might form a complex which binds to CpWAK1
- Activation of dehydration-specific signalling pathways
- Reduction of CpWAK1 levels and leaf folding

Many data available which describe the changes which take place during dehydration/rehydration

Questions:

Which are the essential evolutionary adaptations which allow the targeted expression of stress pathways?

Selected genome duplications e.g. ELIPs, LEAs

Selection of promoter *cis* elements

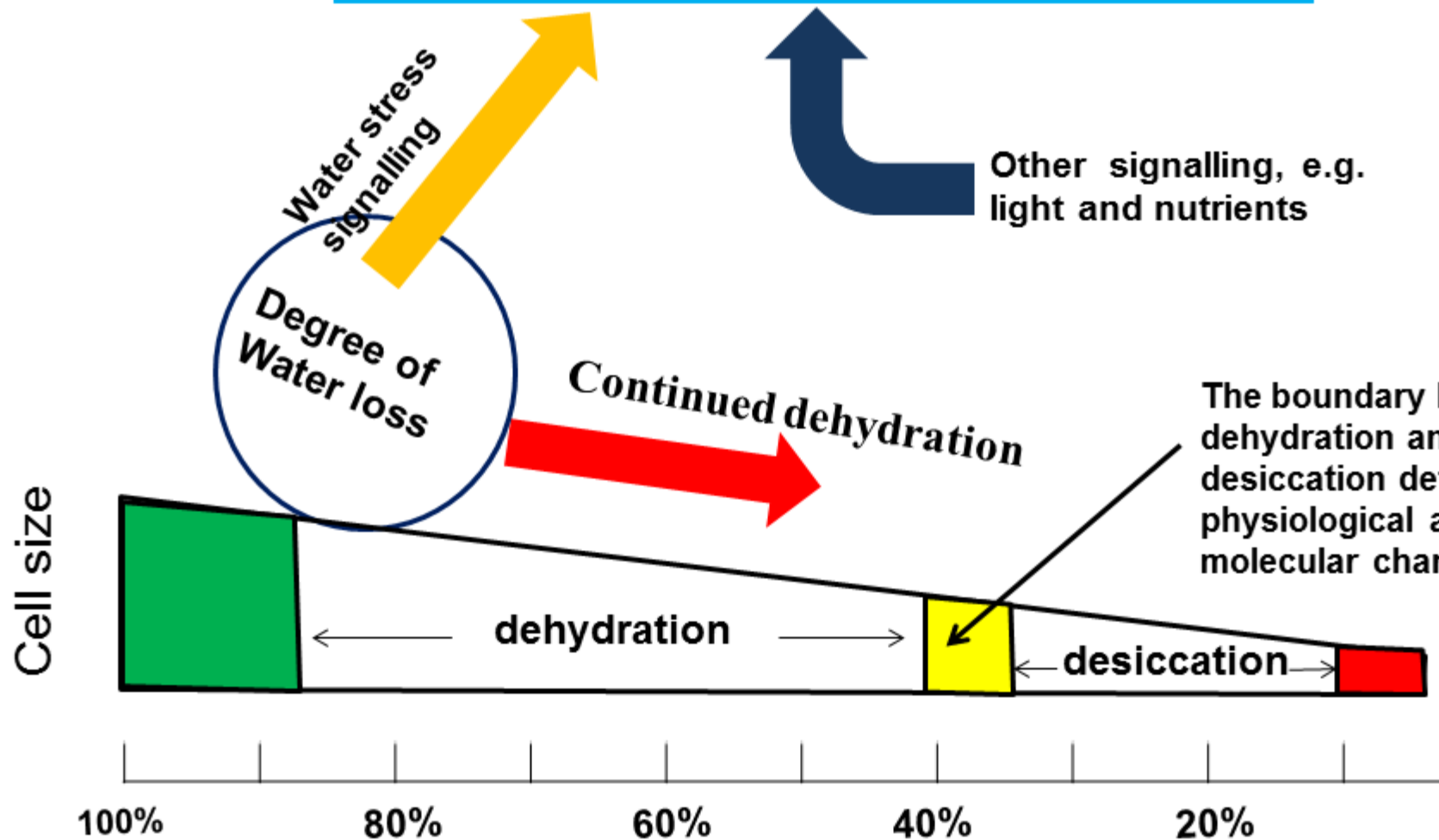
(hypothesis: mutations are not random, selection driven by environment)

Which are the triggers? Cell wall modifications

Biochemical function of stress-responsive proteins mainly unknown

Response strategy:

Regulation of photosynthesis and respiration;
changes of metabolite levels, gene expression and
protein accumulation; cell structure changes, etc.



IMBIO, BONN

Valentino Gariola

Aishwarya Singh

Niklas Jung, Peilei Chen



The Arabidopsis Functional
Genomics Network

Collaborations:
Rob VanBuren and colleagues
Michigan State
Paul Knox, Leeds

Eberhard Fischer, Koblenz

