Cell Growth

How to grow in a cage?

(;,;)

Adelin Barbacci adelin.barbacci@inra.fr

Why studying growth is still central in plant biology?

Introduction

Plant growth is central to

- Survival

  adaptation to physical environment (gravity, wind, light...)

- Morphogenesis

  a shape can be described only by growth speed and growth direction

And by consequence is central to

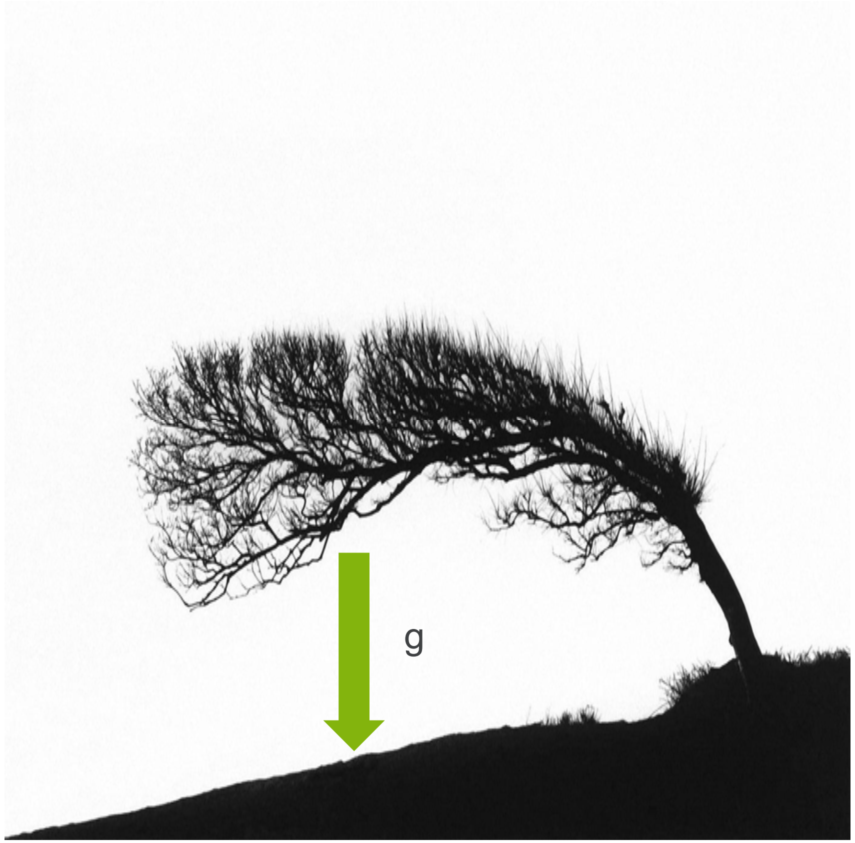
- plant quality

  wood properties (tension wood),

  fruit quality and textures (crunchiness, mealiness, firmness)

- Yield increase

A complex multiscale mechanism



How can tree can stand during long time?

- @ cell level: Gravisensing (statolithes)
- @ tissue level: Proprioception (internal deformation field)
- @ tissue level: Reaction wood formation (cambial growth) -> generation of anisotropic growth stresses
- @ tree level: Bending of the trunk

Moulia, B. and Fournier, M. (2009) The power and control of gravitropic movements in plants: A biomechanical and systems biology view. J. Exp. Bot., 60, 461–486.
Bastien, R., Bohr, T., Moulia, B. and Douady, S. (2013) Unifying model of shoot gravitropism reveals proprioception as a central feature of posture control in plants. Proc. Natl. Acad. Sci. U. S. A., 110, 755–60. Available at: http://www.pnas.org/content/110/2/755.full.
Barbacci, A., Constant, T. and Nepveu, G. (2009) Theoretical and experimental study of a mechanical model describing the trunk behaviour of mature beech trees (Fagus sylvatica L.) under the static loading of the crown. Trees - Struct. Funct., 23, 1137–1147.

But today,

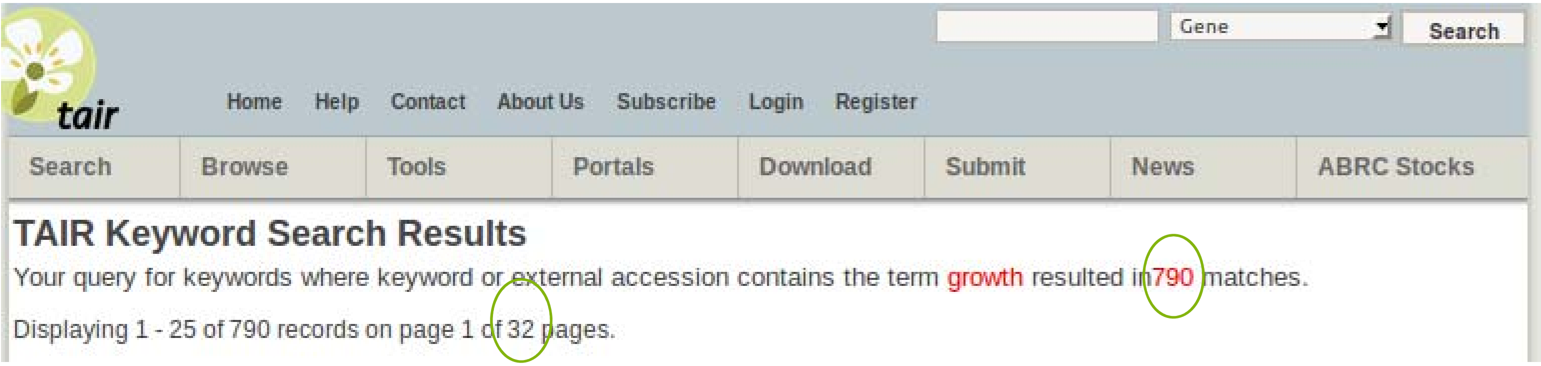
We focus on the elementary mechanisms occurring @ cell level

Interaction 1/1

(If you want more interaction please interupt me with questions, comments ...)

- What is growth for you?
- What are the keywords associated to growth?

Genes involved in growth



Simplifications are needed:

what are the core mechanisms of growth?

Quand il y a plus que 3 flèches dans un graph il devient impossible de comprendre les causes

Etienne Klein (physicien théorique)

Modeling (=simplification)

Is life following laws?

No

Biology ~ Stamp collection

Yes

There is an invariant

Energy

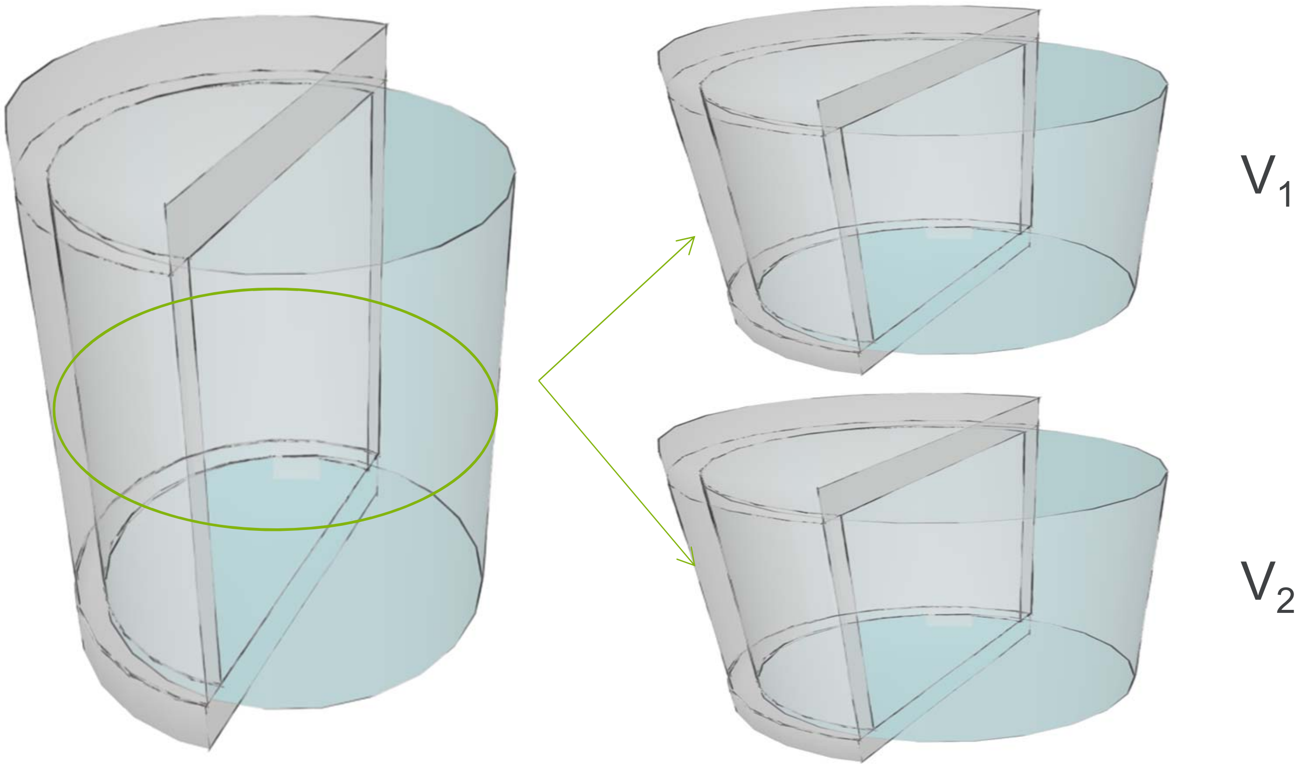
(Bio)physics

Barbacci, A., Magnenet, V. and Lahaye, M. (2015) Thermodynamical journey in plant biology. Front. Plant Sci., 6, 481.
Available at: http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=4485339&tool=pmcentrez&rendertype=abstract.
Zwieniecki, M. a. and Dumais, J. (2011) Quantifying Green Life: Grand Challenges in Plant Biophysics and Modeling. Front. Plant Sci., 2, 1–4.

Cell growth

Some clarifications

Growth is a variation of volume



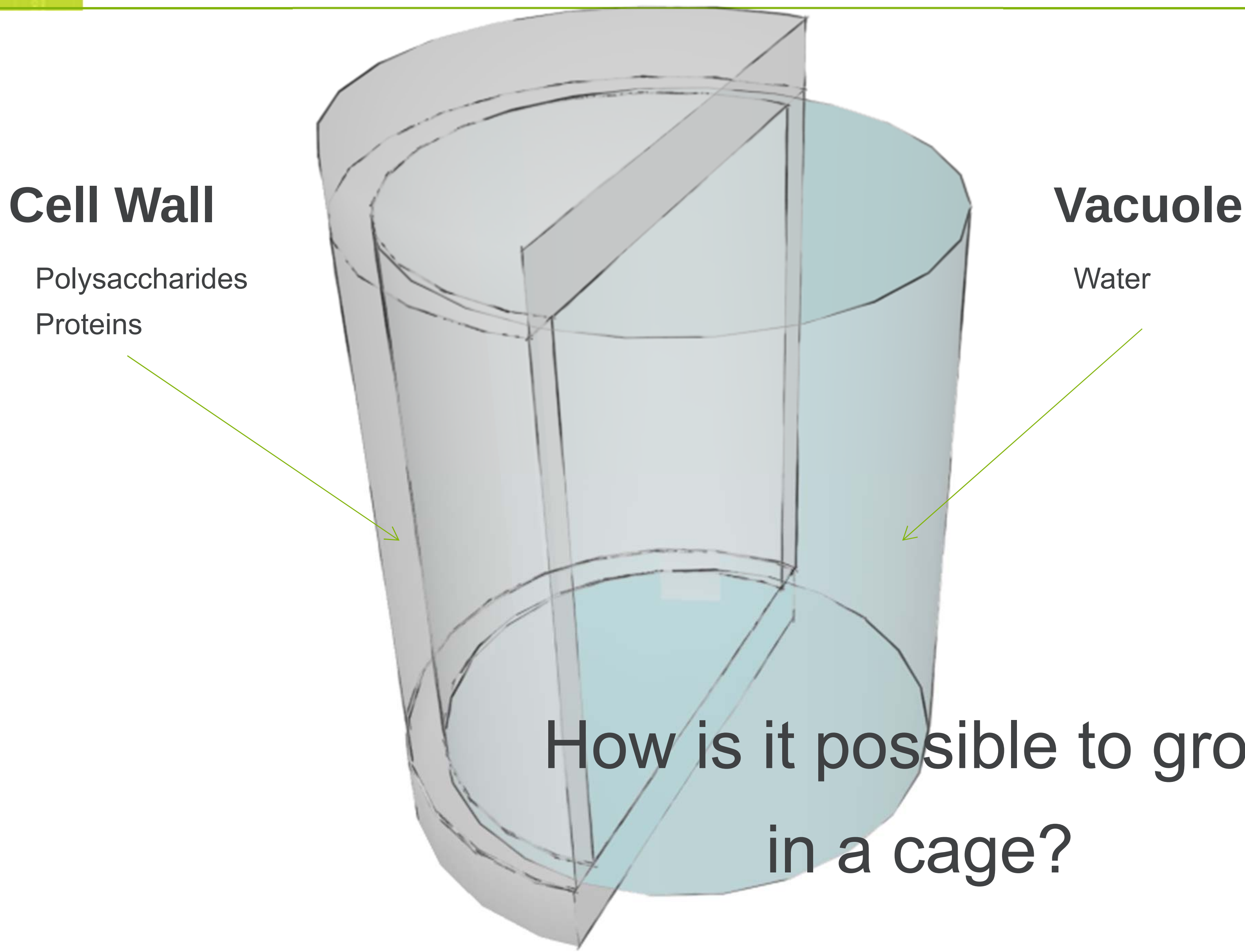
$V_1 = V_2 = 1/2\ V$

$V = V_1 + V_2$

$dV = 0$

Growth involves variation of volume

(so expressed as a power law of length (m^2,m^3) not in g or g/g or g/g/s ...)

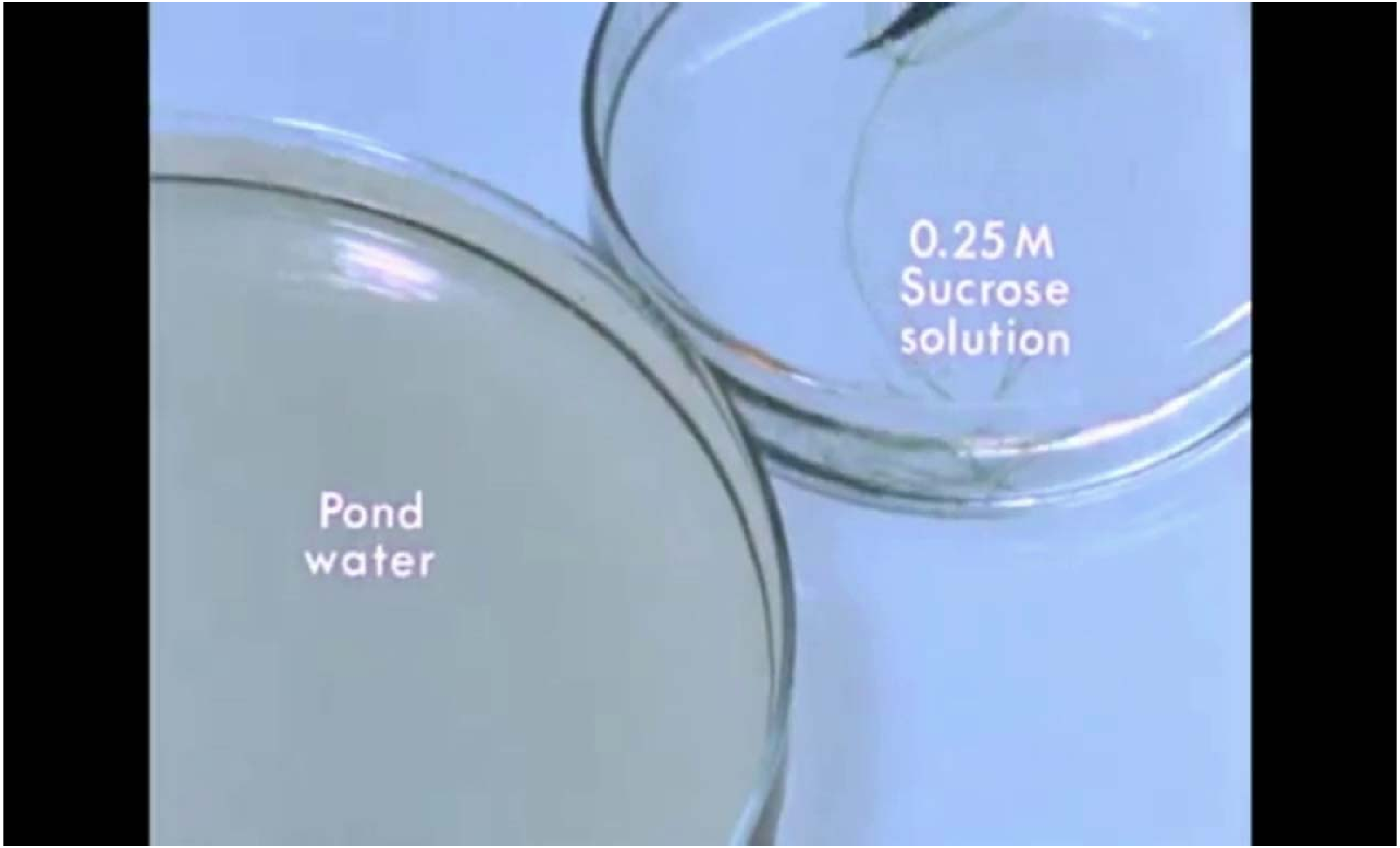


How is it possible to grow in a cage?

Cell growth

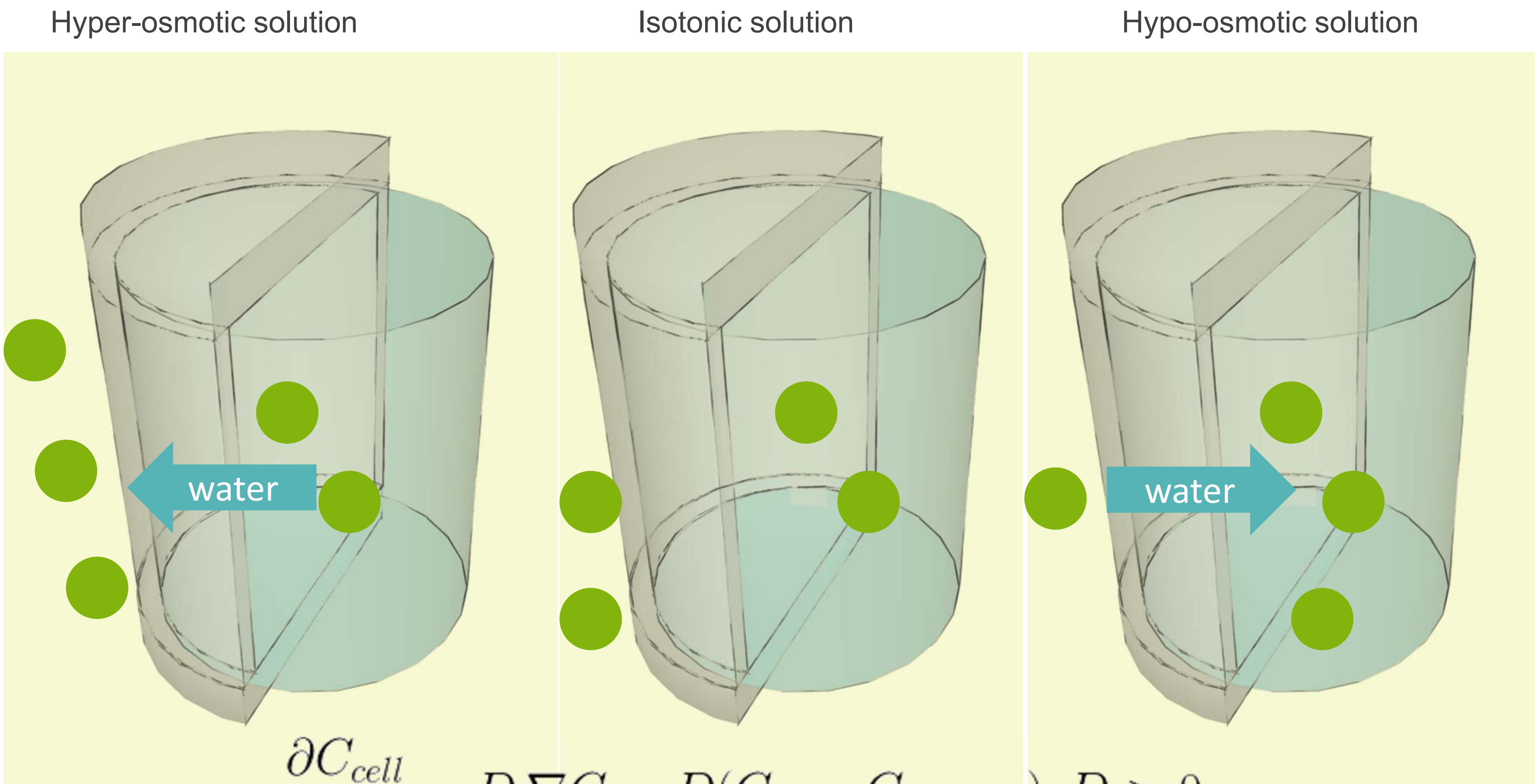
Core mechanisms

Experiment 1: importance of water



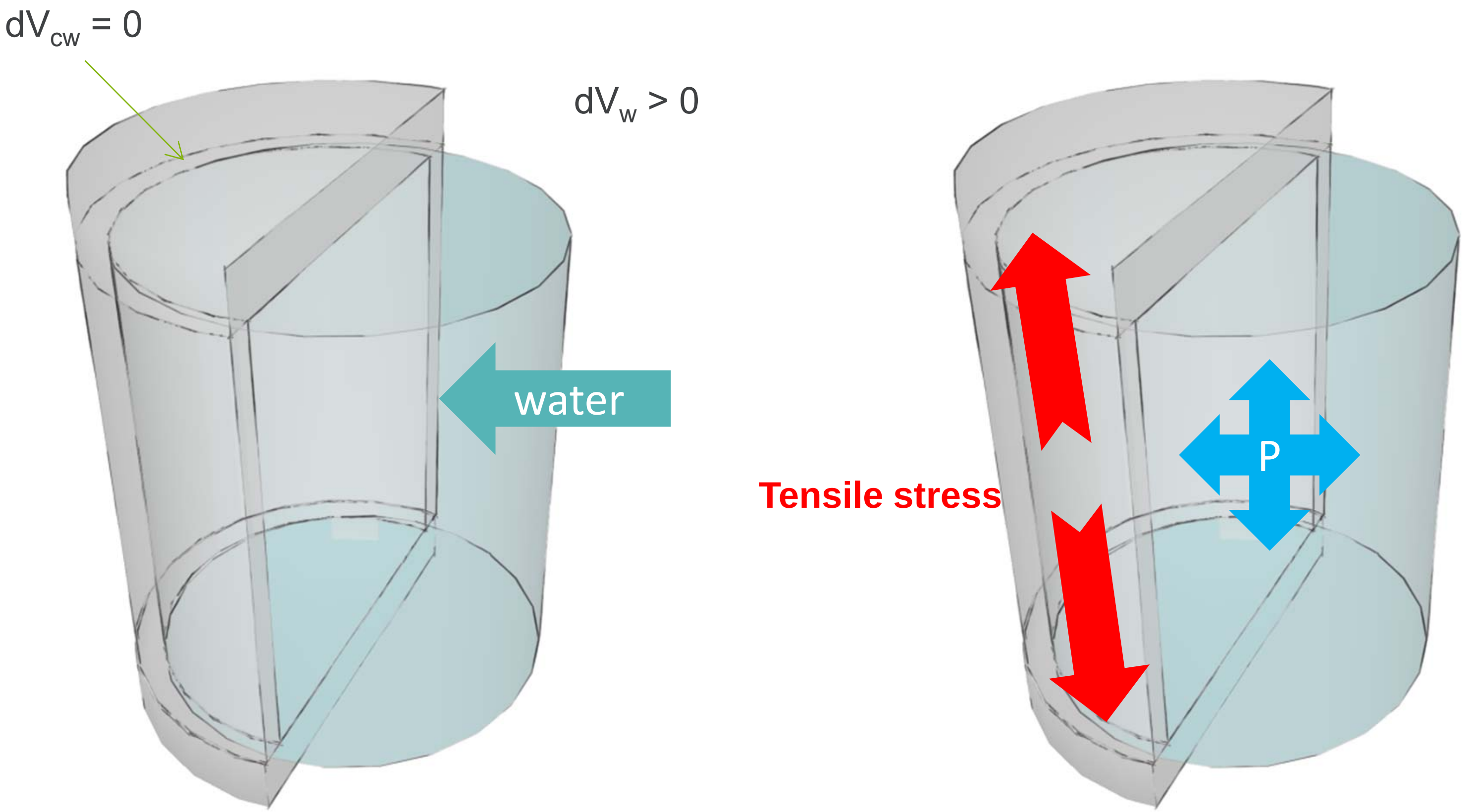
Relation between water and turgor pressure

Water flux is driven by gradient of concentration



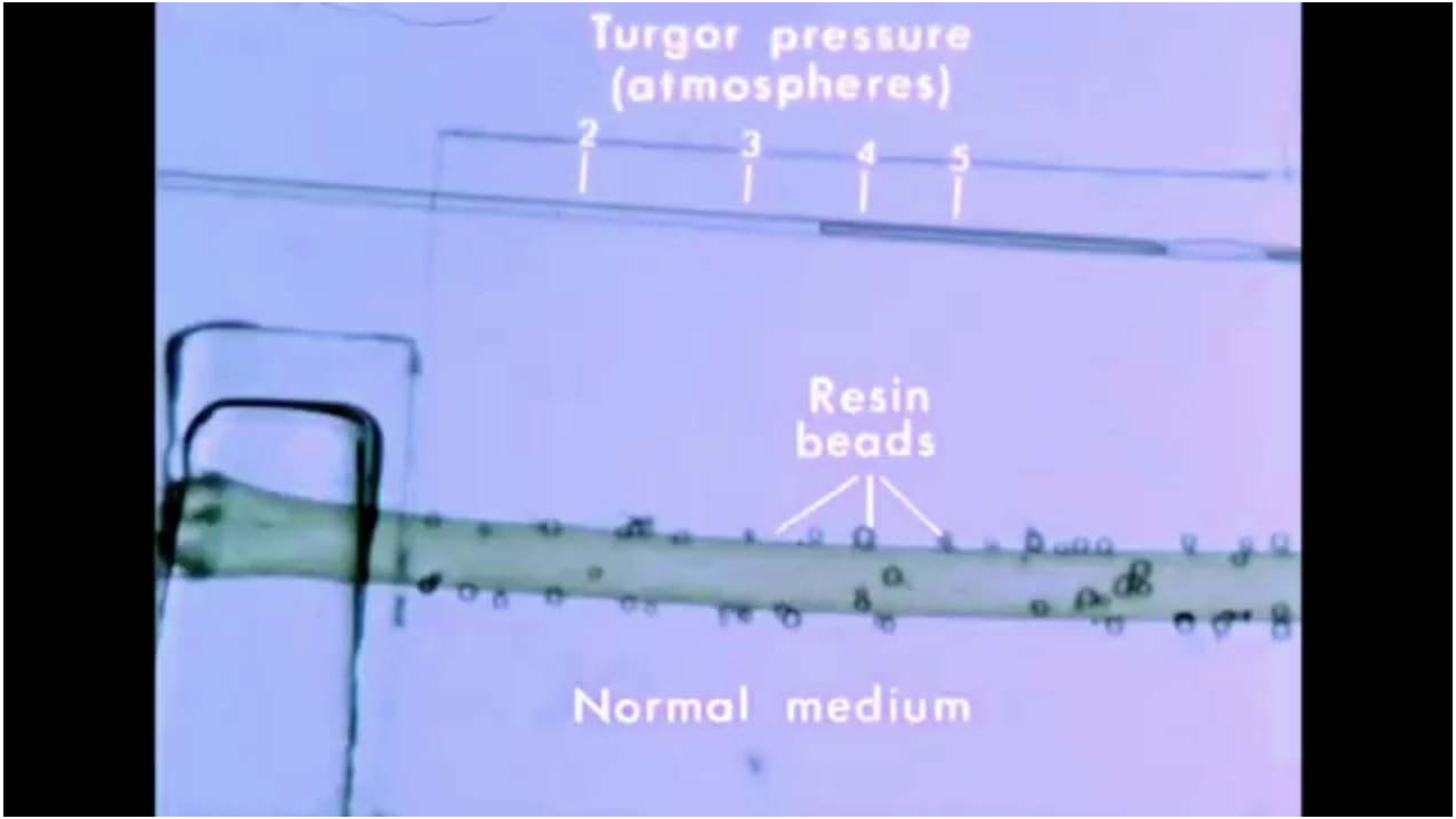
$$\frac{\partial C_{cell}}{\partial t} = D \nabla C = D(C_{cell} - C_{solution}), D > 0$$

Water flux is driven by gradient of concentration



Magnitude of turgor pressure ~ 5 bar

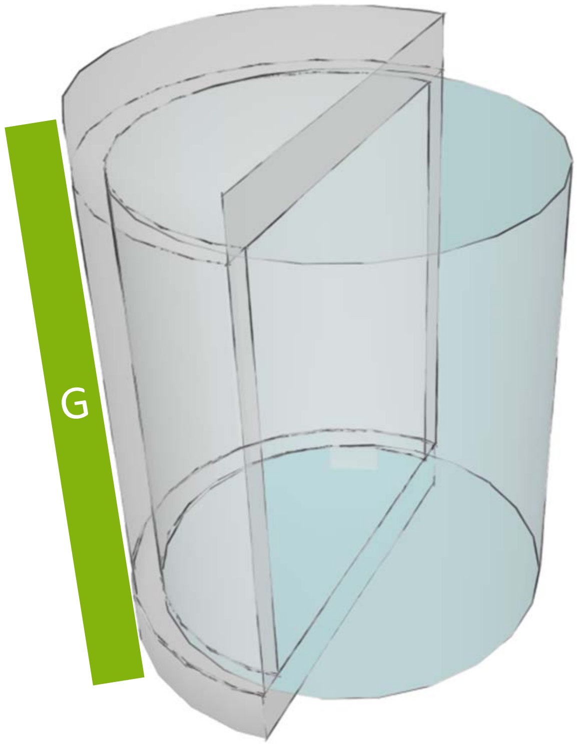
Growth (can) occurs everywhere



Growth (can) occurs in restricted area

2 different growth patterns exist

Diffuse growth (everywhere)



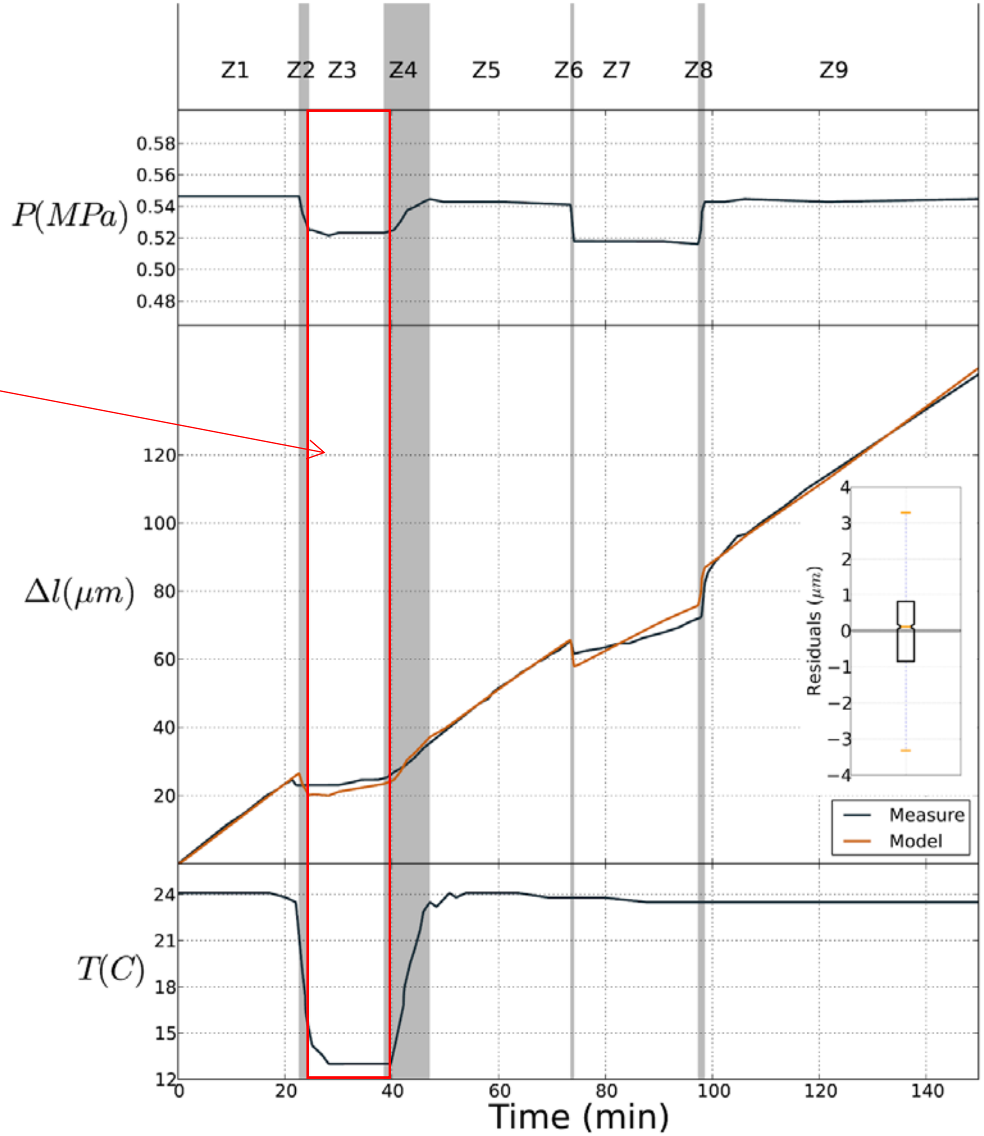
Parenchyma's cells
Internodal cells of characae

Apical growth (restricted area)

Pollen tube
Fungus hyphae

Growth is irreversible

When internal pressure is down, growth stopped but is not negative



Take home messages

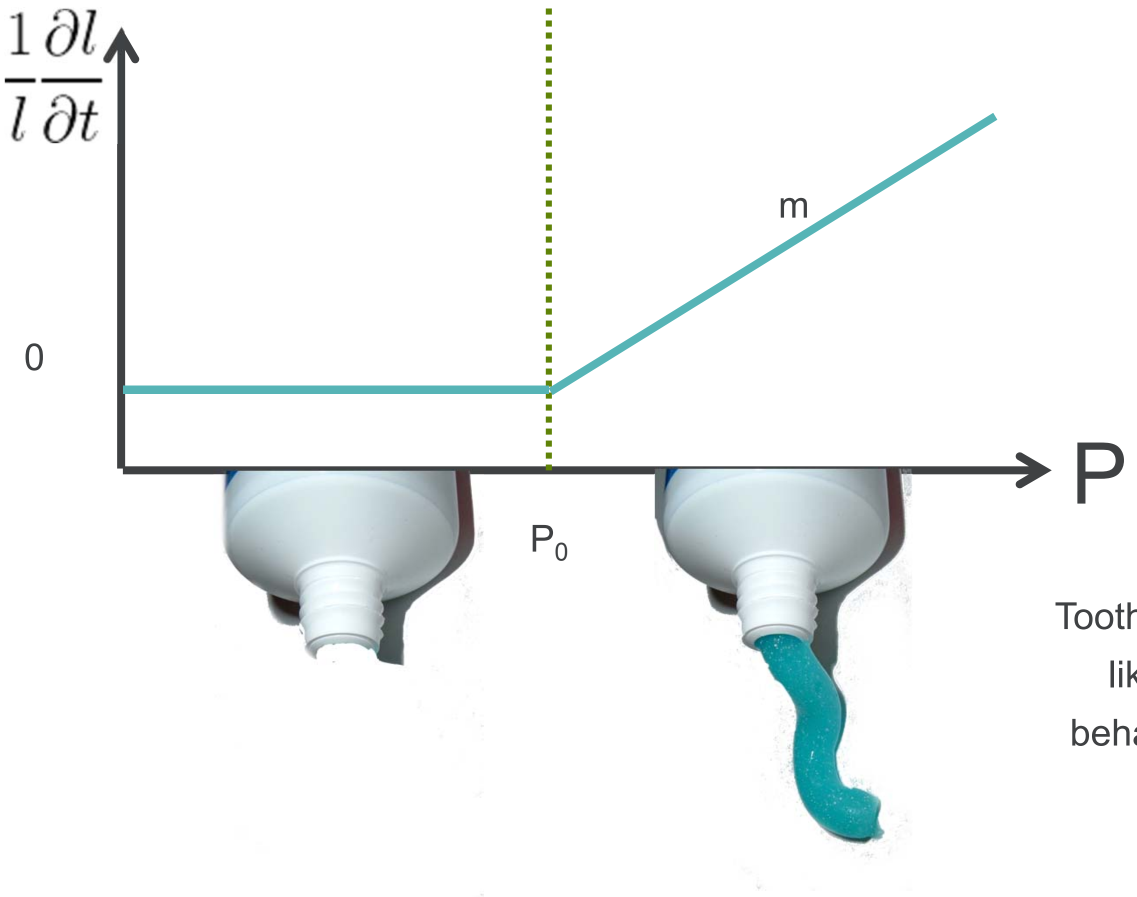
- Irreversible cell expansion is an essential aspect of plant growth and morphogenesis.
- Growth may be highly localised, as in tip-growing cells, or more diffuse growth
- Expansive growth of plant cells requires simultaneous uptake of water into the cell and irreversible expansion of the cell wall.
- Cell growth begins with cell wall loosening which leads to ‘stress relaxation’ of the cell wall which in turn creates the water potential difference needed for water uptake by the cell, resulting in the physical enlargement of the cell.
- The growing cell wall is composed of a network of cellulose microfibrils embedded in pectins and hemicelluloses that make up the wall matrix; these materials combine to form a load-bearing structure that controls cell mechanics and physically limits cell growth.
- Deposition of new polymers to the wall is usually coordinated with surface expansion, but these are separable processes.
- Plant cell walls enlarge more rapidly at low pH (‘acid growth’), a process that is mediated by nonenzymatic proteins named α-expansins.
- Cessation of cell enlargement likely involves multiple processes, including tightening of the matrix-cellulose network and reduced expression of wall-loosening proteins.

Is it enough?

Put everything in a model

Lockhart's model (1965)

$$\frac{1}{l}\frac{\partial l}{\partial t} = m\left(P - P_0\right) \qquad P > 0$$
$$= 0$$



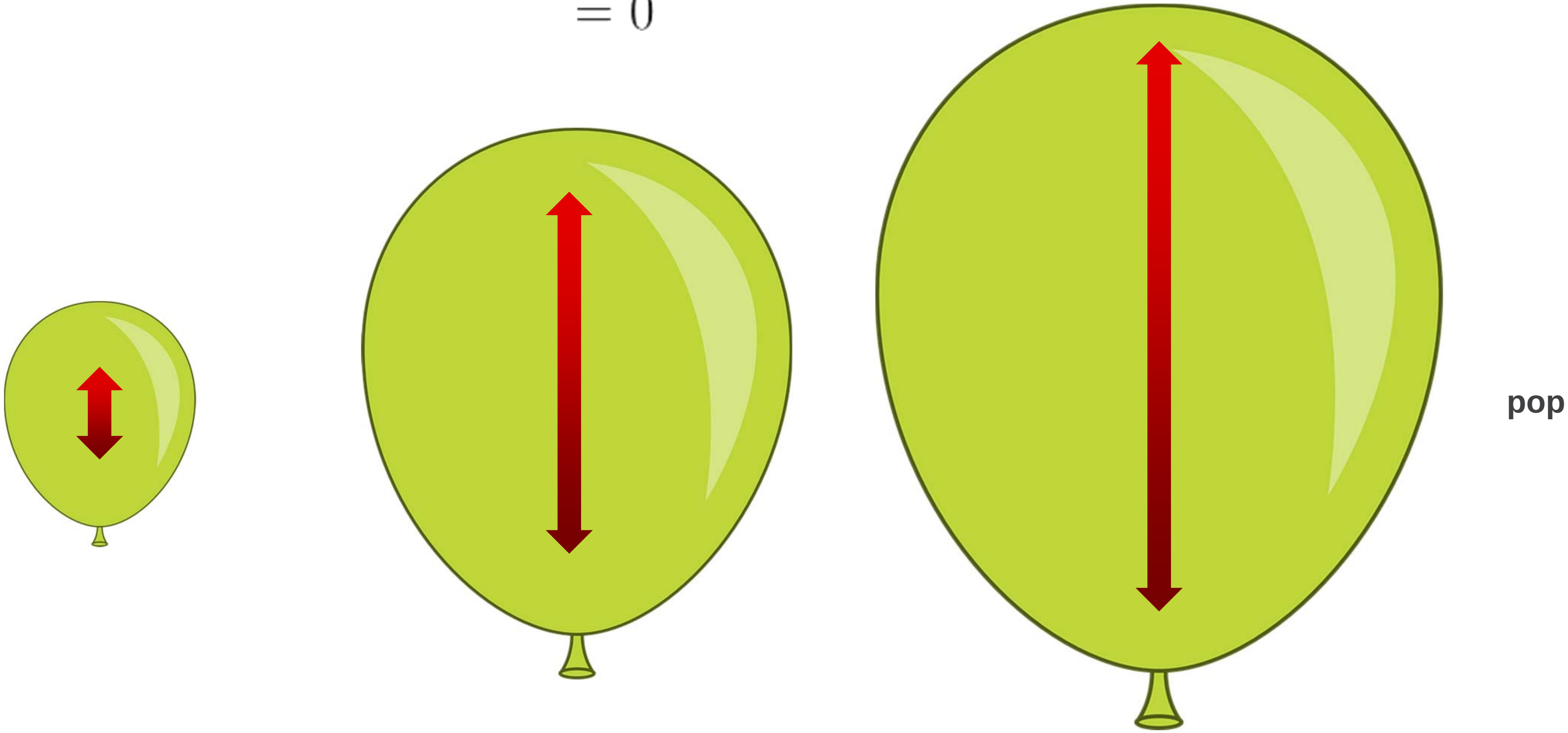
Toothpath

like

behavior

Lockhart's model (1965)

$$\frac{1}{l}\frac{\partial l}{\partial t} = m\left(P - P_0\right) \qquad P > 0$$
$$= 0$$

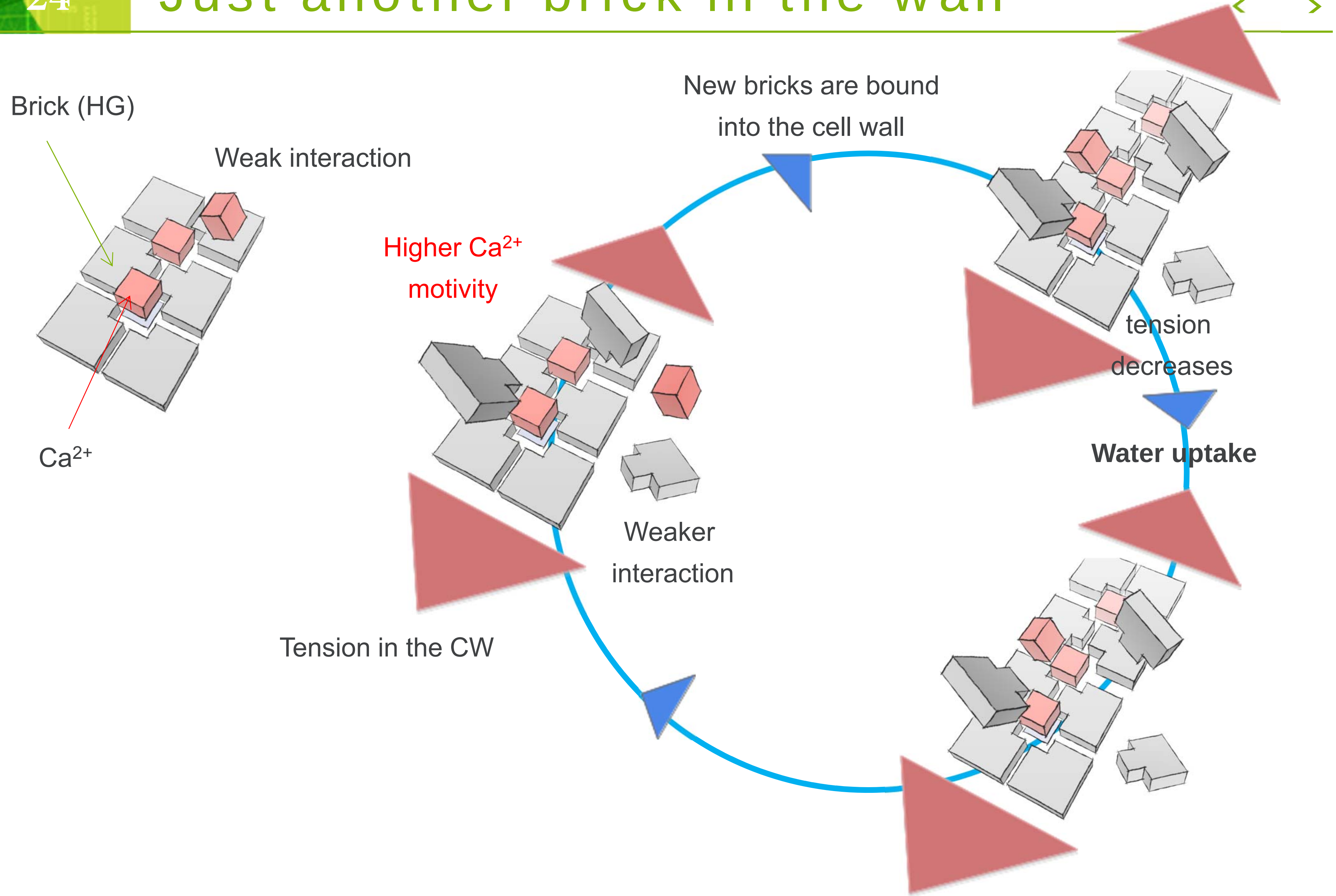


A mechanisms is missing

Cell growth

Irreversible extension of the cell wall

Just another brick in the wall



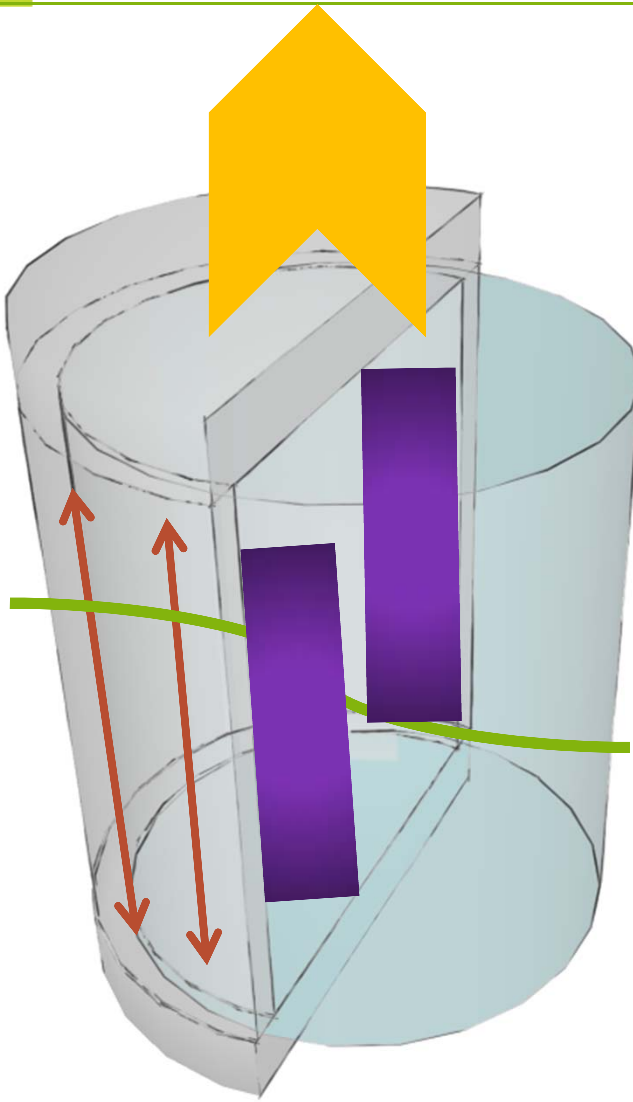
Proseus, T.E. and Boyer, J.S. (2008) Calcium pectate chemistry causes growth to be stored in Chara corallina: A test of the pectate cycle. Plant, Cell Environ., 31, 1147–1155.

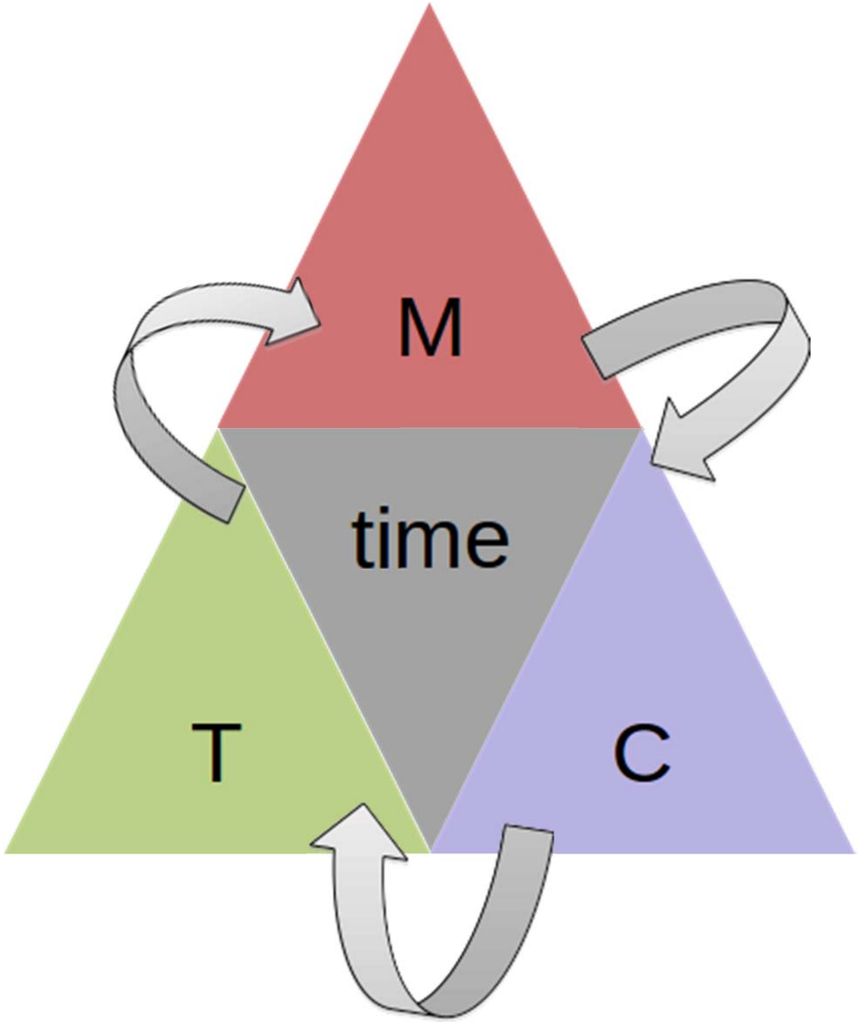
Take home messages

- Irreversible cell expansion is an essential aspect of plant growth and morphogenesis.
- Growth may be highly localised, as in tip-growing cells, or more diffuse growth
- Expansive growth of plant cells requires simultaneous uptake of water into the cell and irreversible expansion of the cell wall.
- Cell growth begins with cell wall loosening which leads to 'stress relaxation' of the cell wall which in turn creates the water potential difference needed for water uptake by the cell, resulting in the physical enlargement of the cell.
- Deposition of new polymers to the wall is usually coordinated with surface expansion, but these are separable processes.
- The growing cell wall is composed of a network of cellulose microfibrils embedded in pectins and hemicelluloses that make up the wall matrix; these materials combine to form a load-bearing structure that controls cell mechanics and physically limits cell growth.
- Plant cell walls enlarge more rapidly at low pH ('acid growth'), a process that is mediated by nonenzymatic proteins named α-expansins.
- Cessation of cell enlargement likely involves multiple processes, including tightening of the matrix-cellulose network and reduced expression of wall-loosening proteins.

Is it enough?

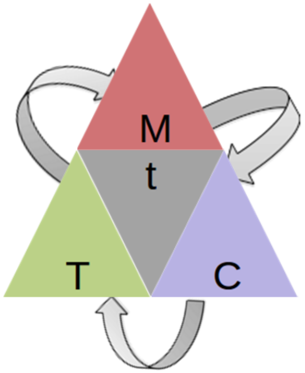
New model (more complex)





Barbacci A, Lahaye M, Magnenet V (2013) Another Brick in the Cell Wall: Biosynthesis Dependent Growth Model. PLoS ONE 8(9): e74400. https://doi.org/10.1371/journal.pone.0074400

Principle of model



Internal energy $\Psi(V, S, \bar{N})$

Constitutive equations

$$\begin{bmatrix} -dP \\ dT \\ d\bar{\mu} \end{bmatrix} = \begin{bmatrix} \gamma & \alpha & B \\ \alpha & \beta & D \\ B & D & \bar{\bar{G}} \end{bmatrix} . \begin{bmatrix} dV \\ dS \\ d\bar{N} \end{bmatrix}$$

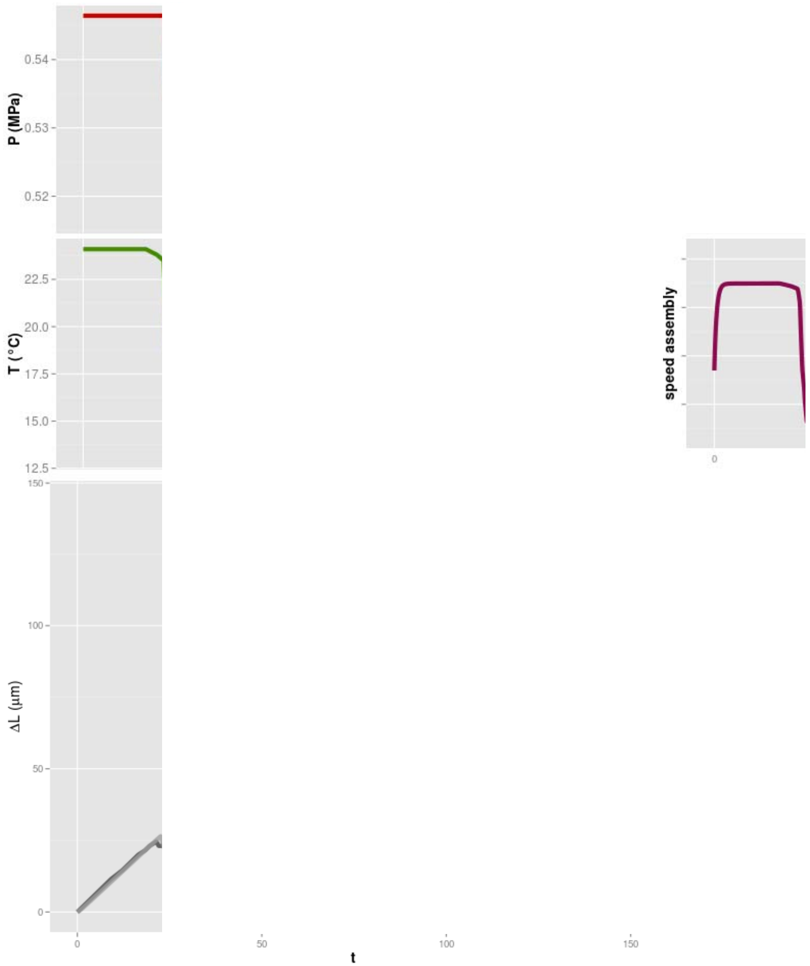
Gibbs-Duhem

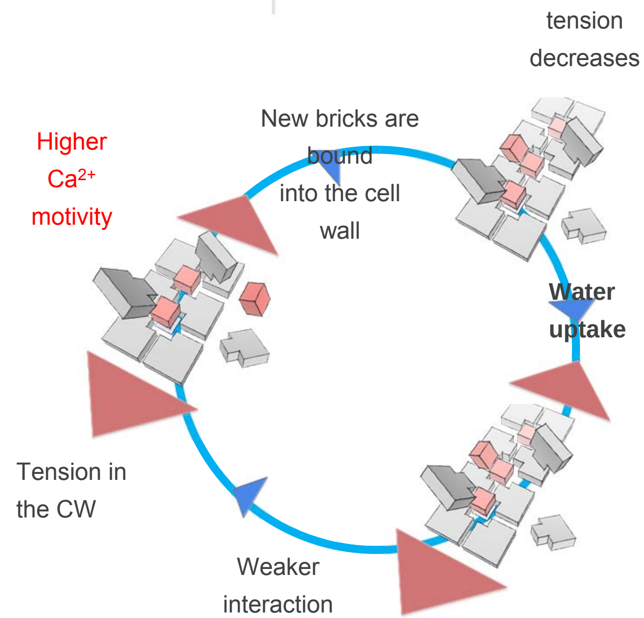
$$\begin{bmatrix} \gamma & \alpha & B \\ \alpha & \beta & D \\ B & D & \bar{\bar{G}} \end{bmatrix} . \begin{bmatrix} V \\ S \\ \bar{N} \end{bmatrix} = 0$$

Grouping chemical species by reactions & time

$$\begin{bmatrix} -\dot{P} \\ \dot{T} \\ -\dot{\bar{A}} \end{bmatrix} = \begin{bmatrix} \gamma & \alpha & B \\ \alpha & \beta & D \\ B & D & \bar{\bar{G}} \end{bmatrix} . \begin{bmatrix} \dot{V} \\ \dot{S} \\ \dot{\bar{\xi}} \end{bmatrix}, \dot{\bar{\xi}} \equiv \bar{L}.\bar{A}$$

Results of the model





Take home messages

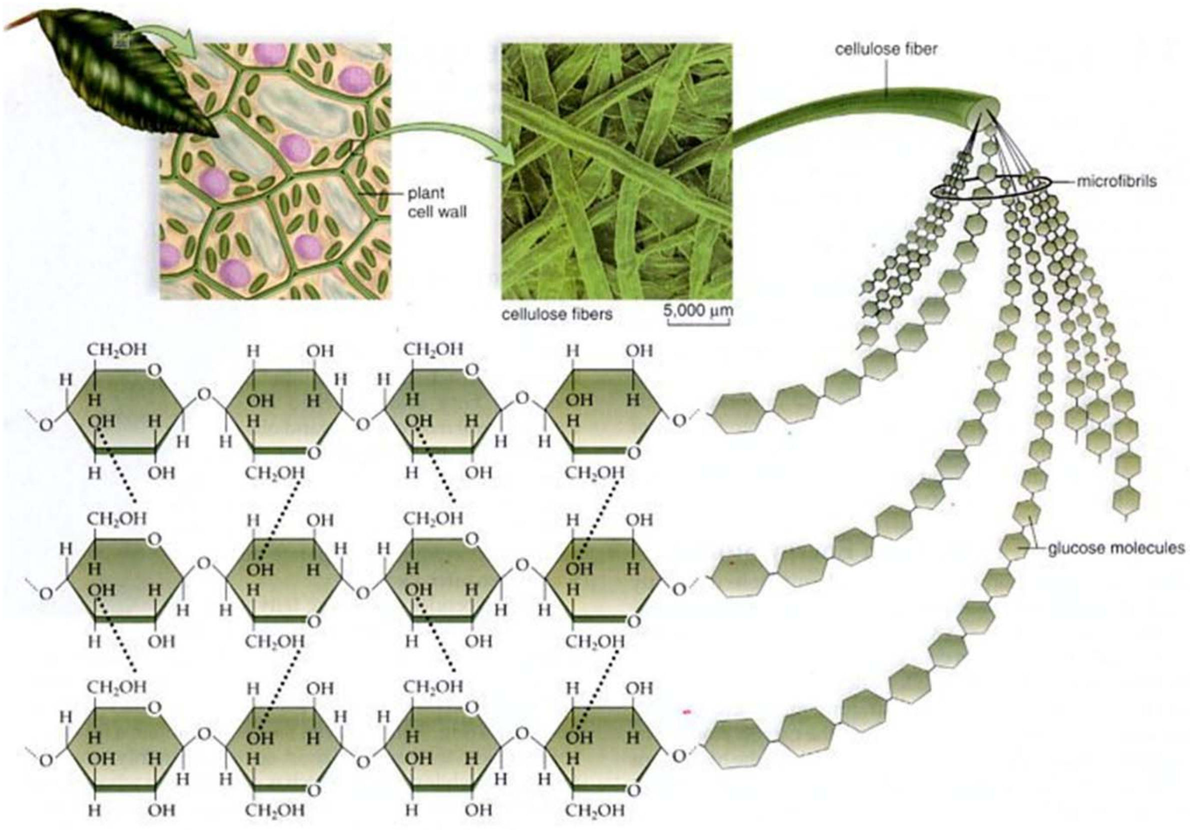
- Irreversible cell expansion is an essential aspect of plant growth and morphogenesis.
- Growth may be highly localised, as in tip-growing cells, or more diffuse growth
- Expansive growth of plant cells requires simultaneous uptake of water into the cell and irreversible expansion of the cell wall.
- Cell growth begins with cell wall loosening which leads to 'stress relaxation' of the cell wall which in turn creates the water potential difference needed for water uptake by the cell, resulting in the physical enlargement of the cell.
- Deposition of new polymers to the wall is usually coordinated with surface expansion, but these are separable processes.
- The growing cell wall is composed of a network of cellulose microfibrils embedded in pectins and hemicelluloses that make up the wall matrix; these materials combine to form a load-bearing structure that controls cell mechanics and physically limits cell growth.
- Plant cell walls enlarge more rapidly at low pH ('acid growth'), a process that is mediated by nonenzymatic proteins named α-expansins.
- Cessation of cell enlargement likely involves multiple processes, including tightening of the matrix-cellulose network and reduced expression of wall-loosening proteins.

Not so bad

Plant cell wall

Composition

Plant cell wall is composed mainly by polysaccharides

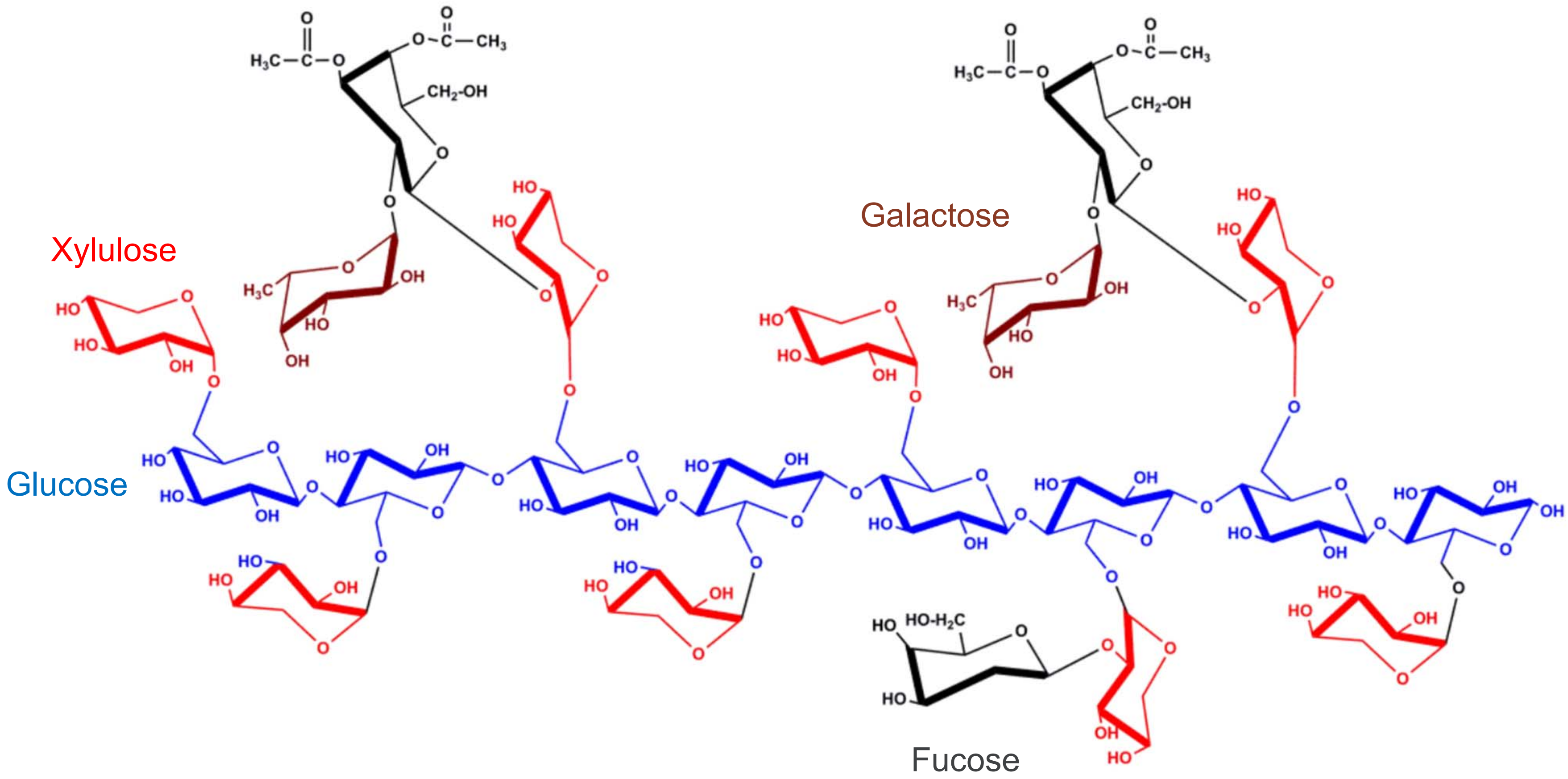


linear polymer of glucose

Plant cell wall is composed mainly by polysaccharides

xyloglucan,
glucuronoarabinoxylan,
mannans

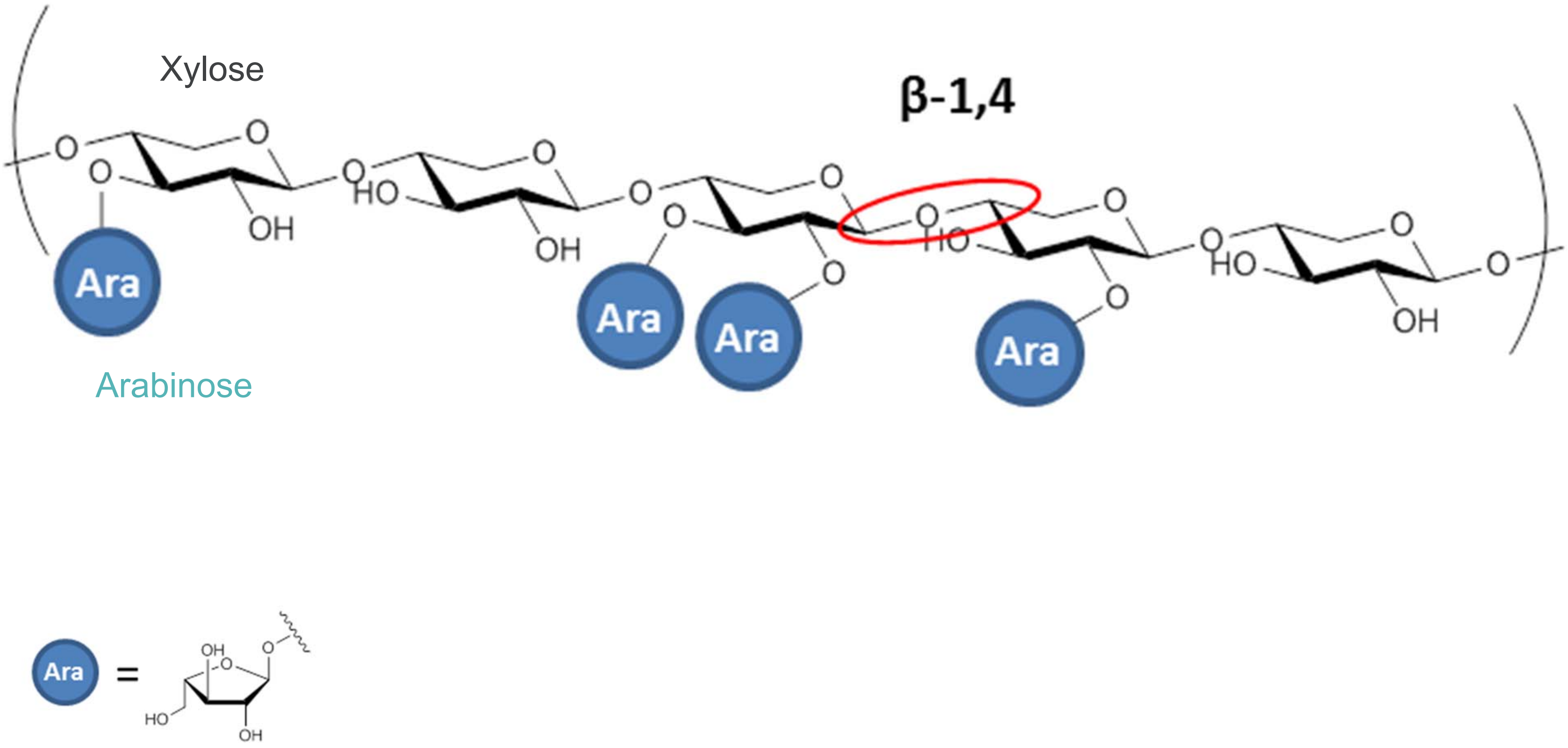
Hemicelluloses



Cosgrove, D.J. (2016) Plant cell wall extensibility: Connecting plant cell growth with cell wall structure, mechanics, and the action of wall-modifying enzymes. J. Exp. Bot., 67, 463–476.

Plant cell wall is composed mainly by polysaccharides

xyloglucan,
glucurono**arabinoxylan**, **Hemicelluloses**
mannans

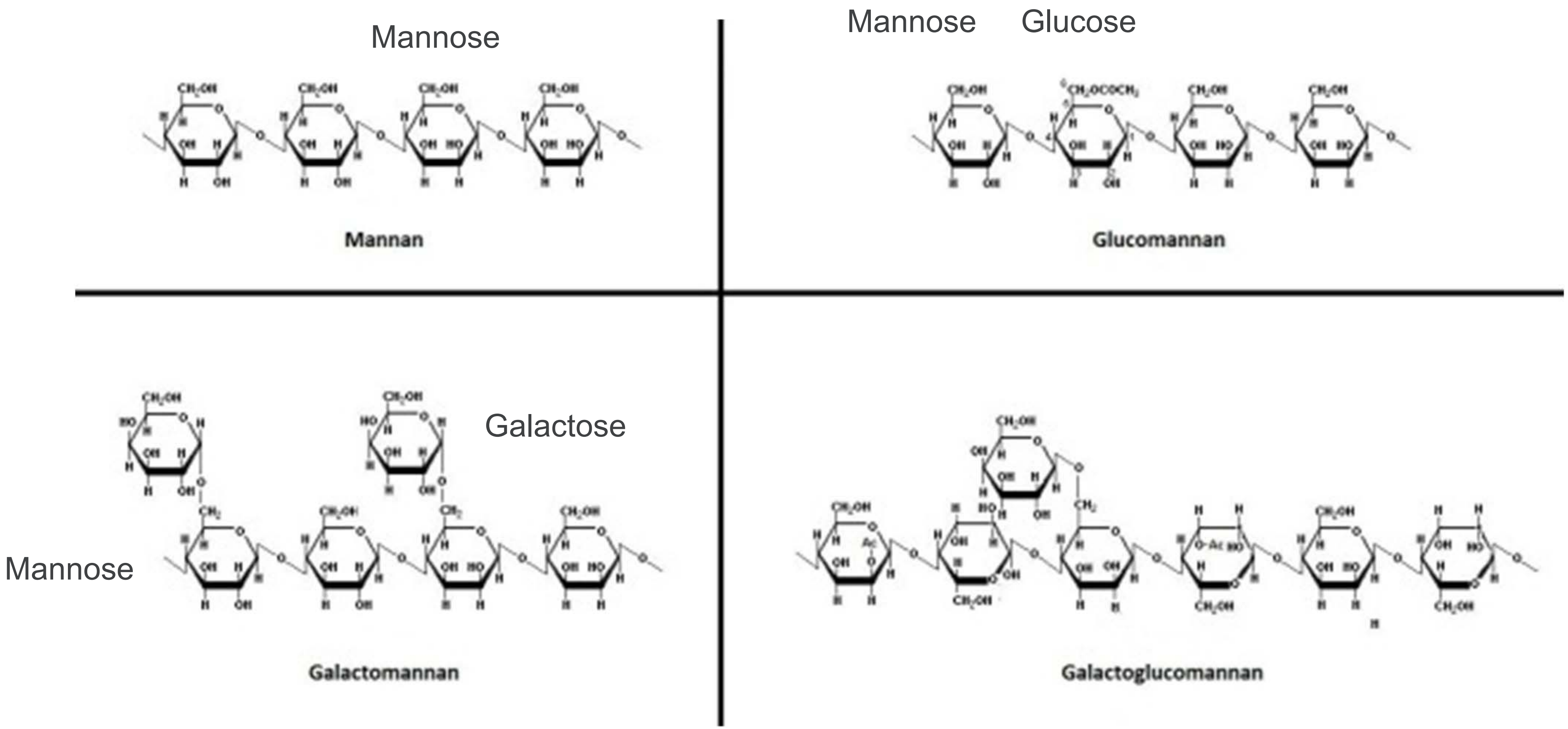


Plant cell wall is composed mainly by polysaccharides

xyloglucan,

glucuronoarabinoxylan, **Hemicelluloses**

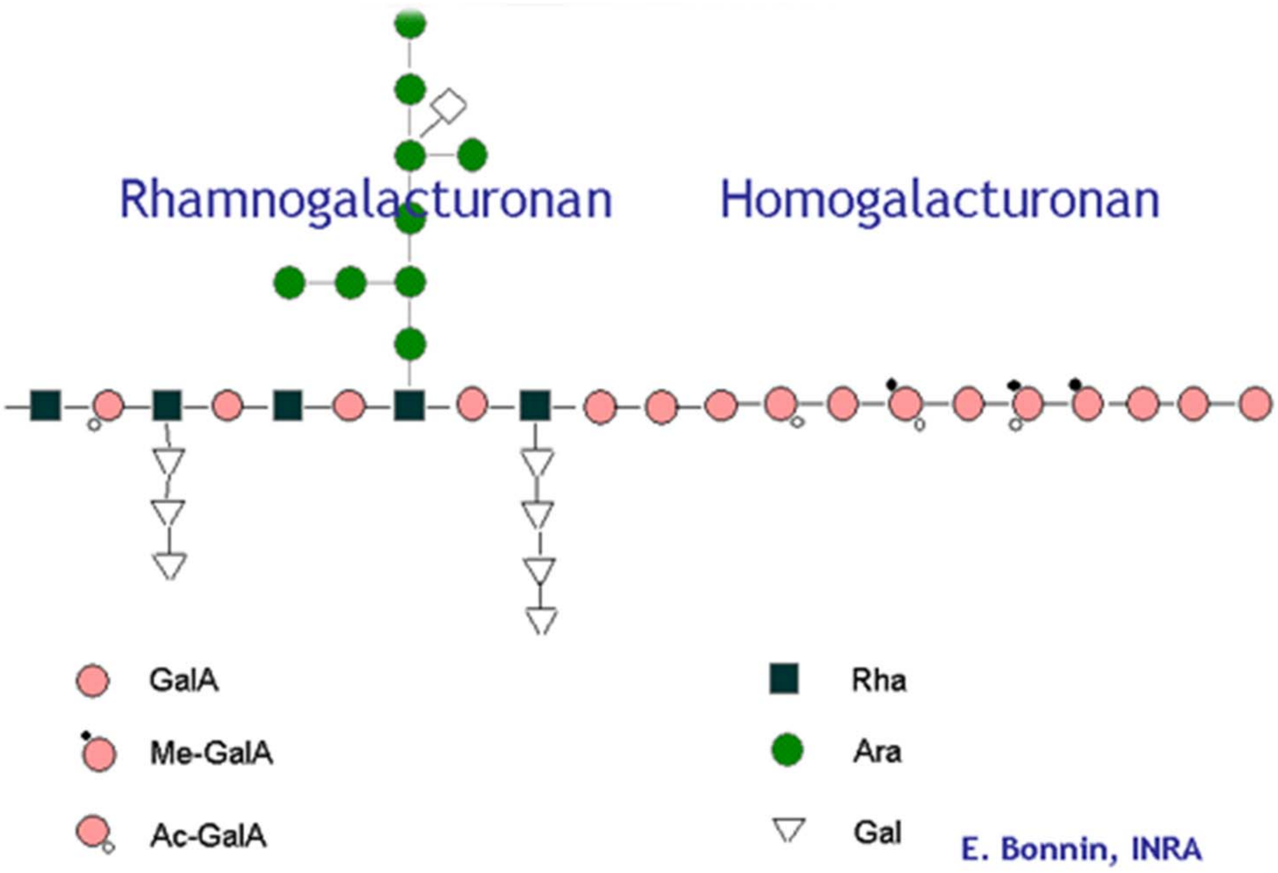
mannans



Plant cell wall is composed mainly by polysaccharides

homogalacturonans
rhamnogalacturonans

Pectins



Take home messages

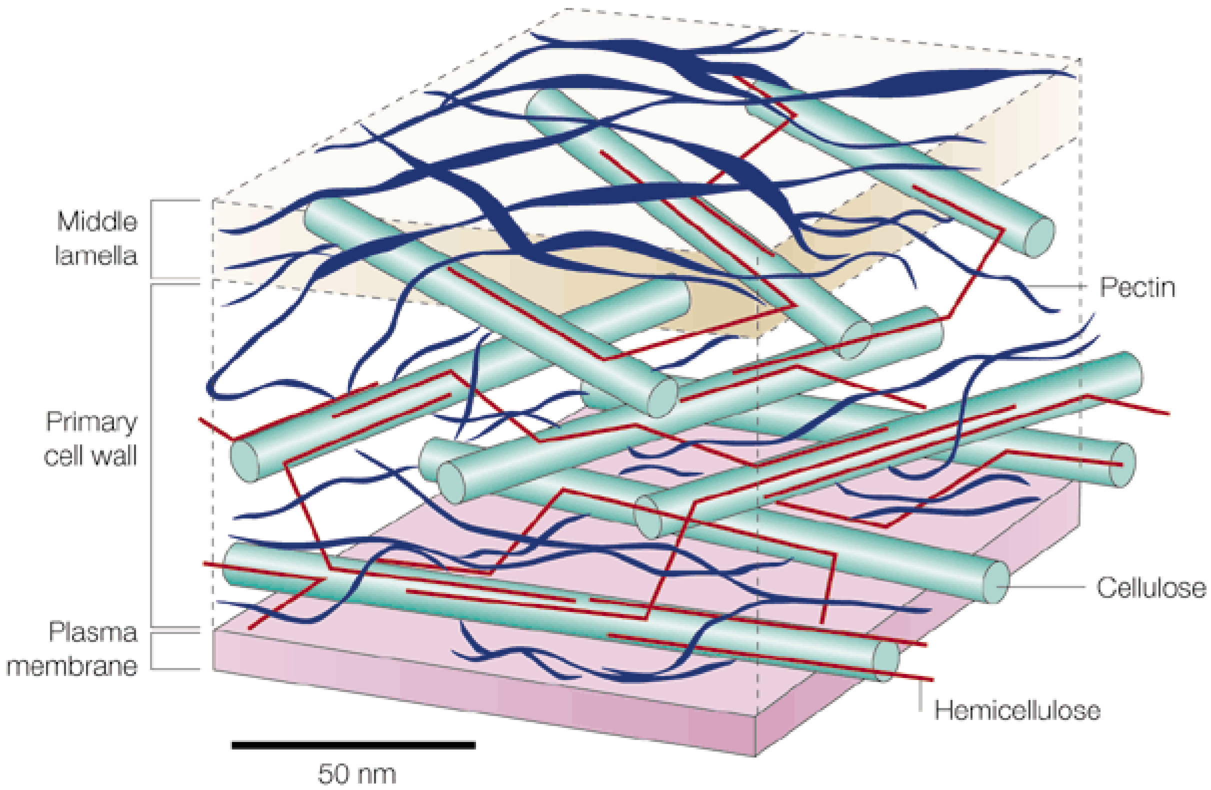
- Irreversible cell expansion is an essential aspect of plant growth and morphogenesis.
- Growth may be highly localised, as in tip-growing cells, or more diffuse growth
- Expansive growth of plant cells requires simultaneous uptake of water into the cell and irreversible expansion of the cell wall.
- Cell growth begins with cell wall loosening which leads to ‘stress relaxation’ of the cell wall which in turn creates the water potential difference needed for water uptake by the cell, resulting in the physical enlargement of the cell.
- Deposition of new polymers to the wall is usually coordinated with surface expansion, but these are separable processes.
- The growing cell wall is composed of a network of cellulose microfibrils embedded in pectins and hemicelluloses that make up the wall matrix;
- these materials combine to form a load-bearing structure that controls cell mechanics and physically limits cell growth.

Is it enough?

Plant cell wall

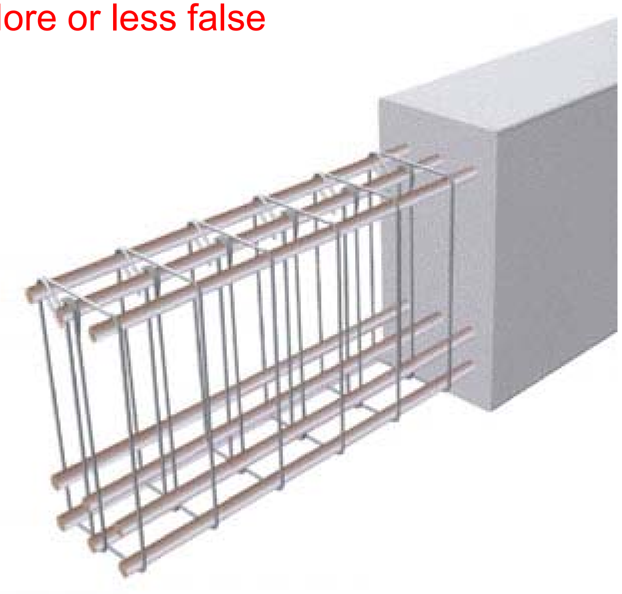
Creation of mechanical properties

Creation of mechanical properties



More or less true

More or less false



The tethered network

captif

Plant cell division: building walls in the right places Laurie G. Smith Nature Reviews Molecular Cell Biology 2, 33-39 (2001) doi:10.1038/35048050

Toward new models

386 Biophysical Journal Volume 103 August 2012 386–394

Atomic Force Microscopy Stiffness Tomography on Living *Arabidopsis thaliana* Cells Reveals the Mechanical Properties of Surface and Deep Cell-Wall Layers during Growth

Ksenija Radotić,[†*] Charles Roduit,[§] Jasna Simonović,[†] Patricia Hornitschek,[¶] Christian Fankhauser,[¶] Dragosav Mutavdžić,[†] Gabor Steinbach,[||] Giovanni Dietler,[‡] and Sandor Kasas[‡§]
[†]Institute for Multidisciplinary Research, University of Belgrade, Belgrade, Serbia; [‡]Laboratoire de Physique de la Matière Vivante, École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland; [§]Département de Biologie Cellulaire et de Morphologie and [¶]Center for Integrative Genomics, Université de Lausanne, Lausanne, Switzerland; and [||]Hungarian Academy of Sciences, Biological Research Centre, Szeged, Hungary

Order of magnitude of E:

0.5 MPa during growth

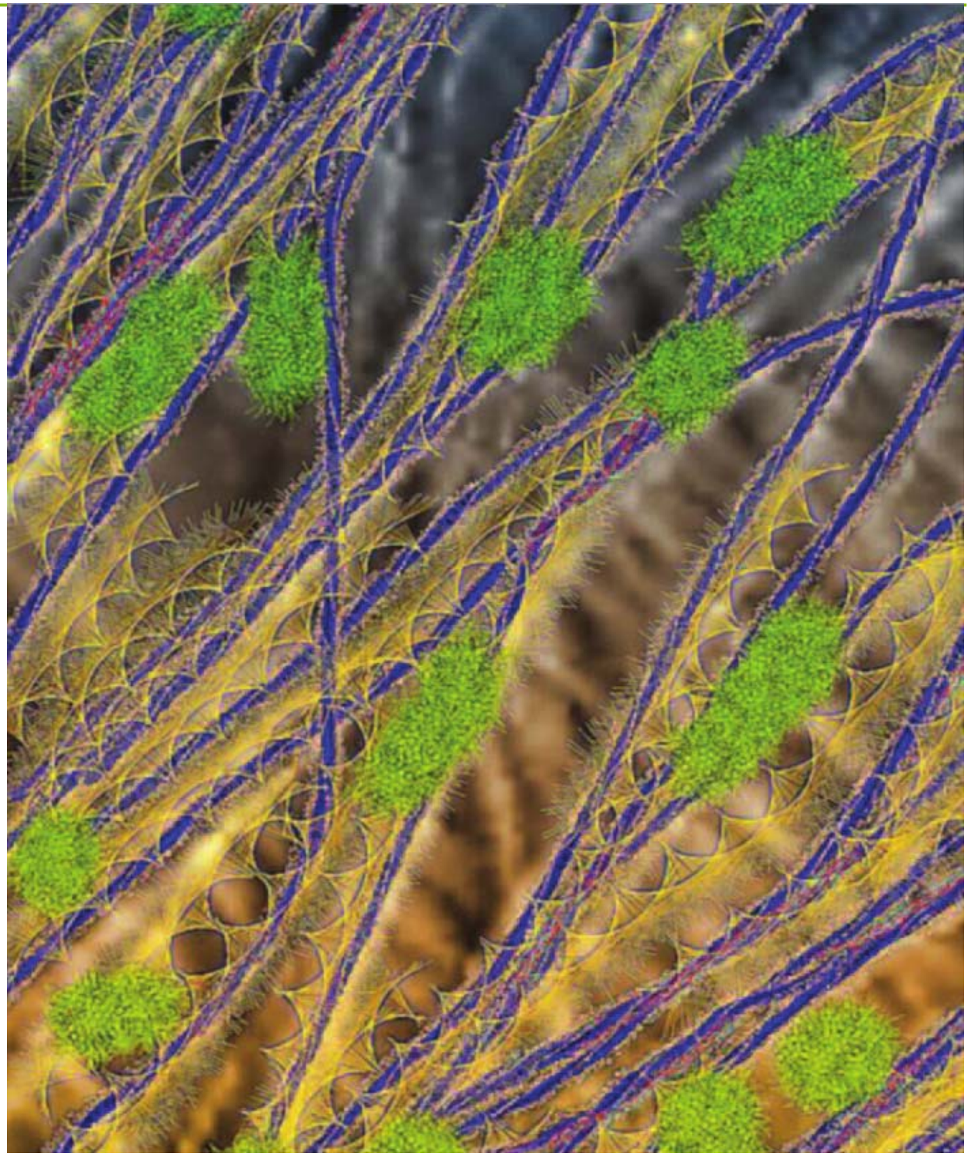
21GPa for steel

Potential Cellulose_ cellulose junctions

xyloglucan

pectins

microfibrills



Cosgrove, D.J. (2016) Plant cell wall extensibility: Connecting plant cell growth with cell wall structure, mechanics, and the action of wall-modifying enzymes. J. Exp. Bot., 67, 463–476.

But it is not so simple

Copyedited by: OUP

Journal of Experimental Botany
doi:10.1093/jxb/erx329
This paper is available online free of all access charges (see http://jxb.oxfordjournals.org/open_access.html for further details)

RESEARCH PAPER

Examining the contribution of cell wall polysaccharides to the mechanical properties of apple parenchyma tissue using exogenous enzymes

Pauline Videcoq[1,†], Adelin Barbacci[1,*,†], Carole Assor[1,3], Vincent Magnenet[2], Olivier Arnould[3], Sophie Le Gall[1] and Marc Lahaye[1,*]

[1] INRA, UR1268 Biopolymères Interactions et Assemblages, F-44316 Nantes, France
[2] Université de Strasbourg, UMR 7357 Laboratoire des Sciences de l'Ingénieur, de l'Informatique et de l'Imagerie (ICube), CNRS, Illkirch, France
[3] Université de Montpellier, LMGC, CNRS, Montpellier, France

* Correspondence: marc.lahaye@inra.fr or adelin.barbacci@toulouse.inra.fr

† These authors contributed equally to this work.

Received 24 March 2017; Editorial decision 24 August 2017; Accepted 24 August 2017

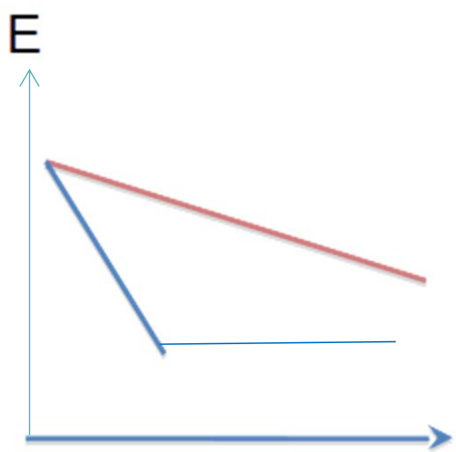
Editor: Bruno Moulia, INRA-University Blaise Pascal

Abstract

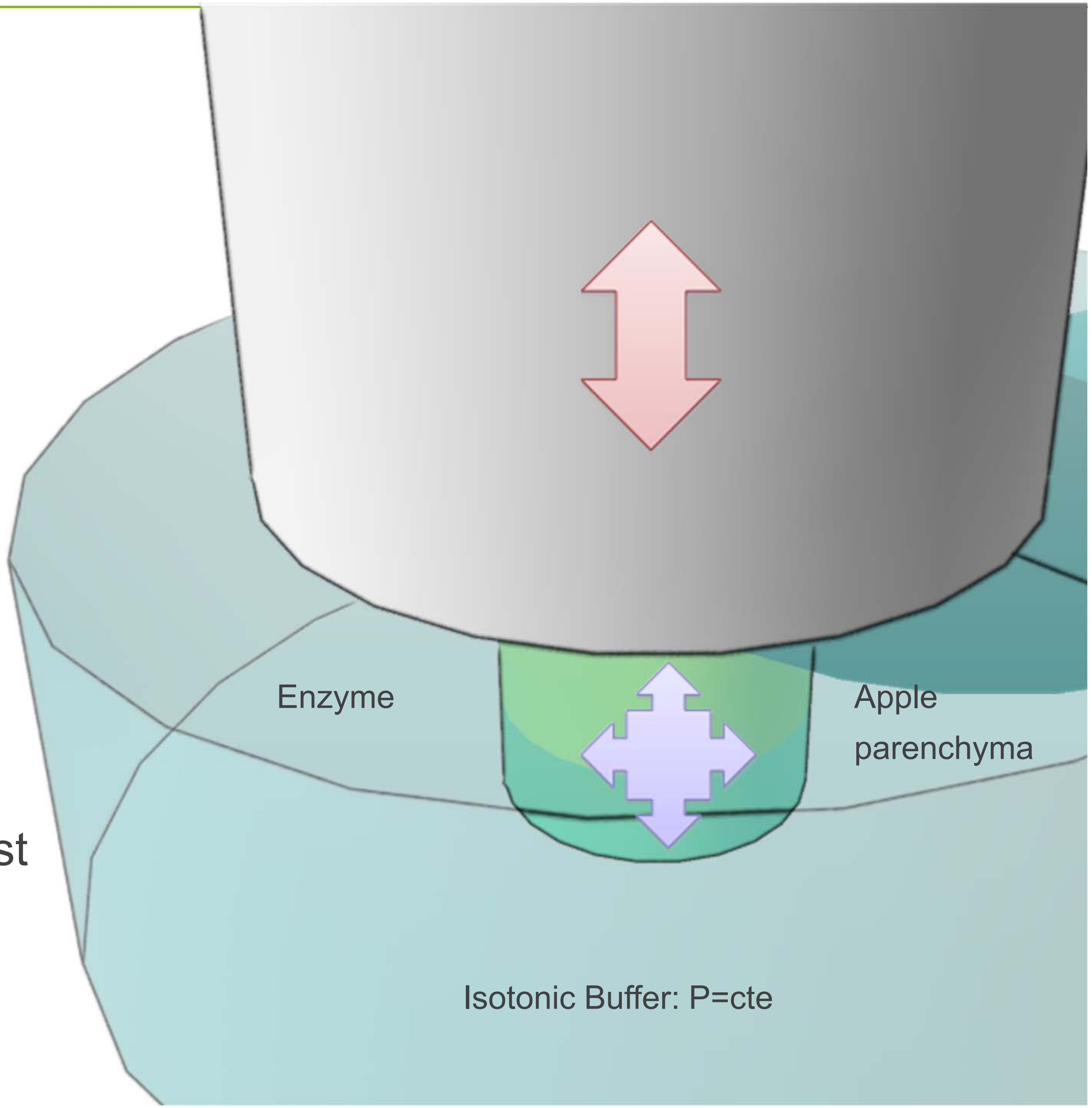
The viscoelastic mechanical properties of water-rich plant tissues are fundamental for many aspects of organ physiology and plant functioning. These properties are determined partly by the water in cellular vacuole and partly by the mechanical properties of the cell wall, the latter varying according to the composition and organization of its polysaccharides. In this study, relationships between the viscoelastic properties of apple cortex parenchyma tissue and cell wall pectin, hemicelluloses, and cellulose structures were studied by infusing the tissue with selected sets of purified enzymes in a controlled osmoticum. The results showed that tissue elasticity and viscosity were related, and controlled to variable extents by all the targeted polysaccharides. Among them, pectic homogalacturonan domains, crystalline cellulose, and fucosylated xyloglucan were revealed as being of prime importance in determining the viscoelastic mechanical properties of apple cortex tissue.

Key words: Apple, biomechanics, cell wall, enzyme, mechanical properties, model, parenchyma.

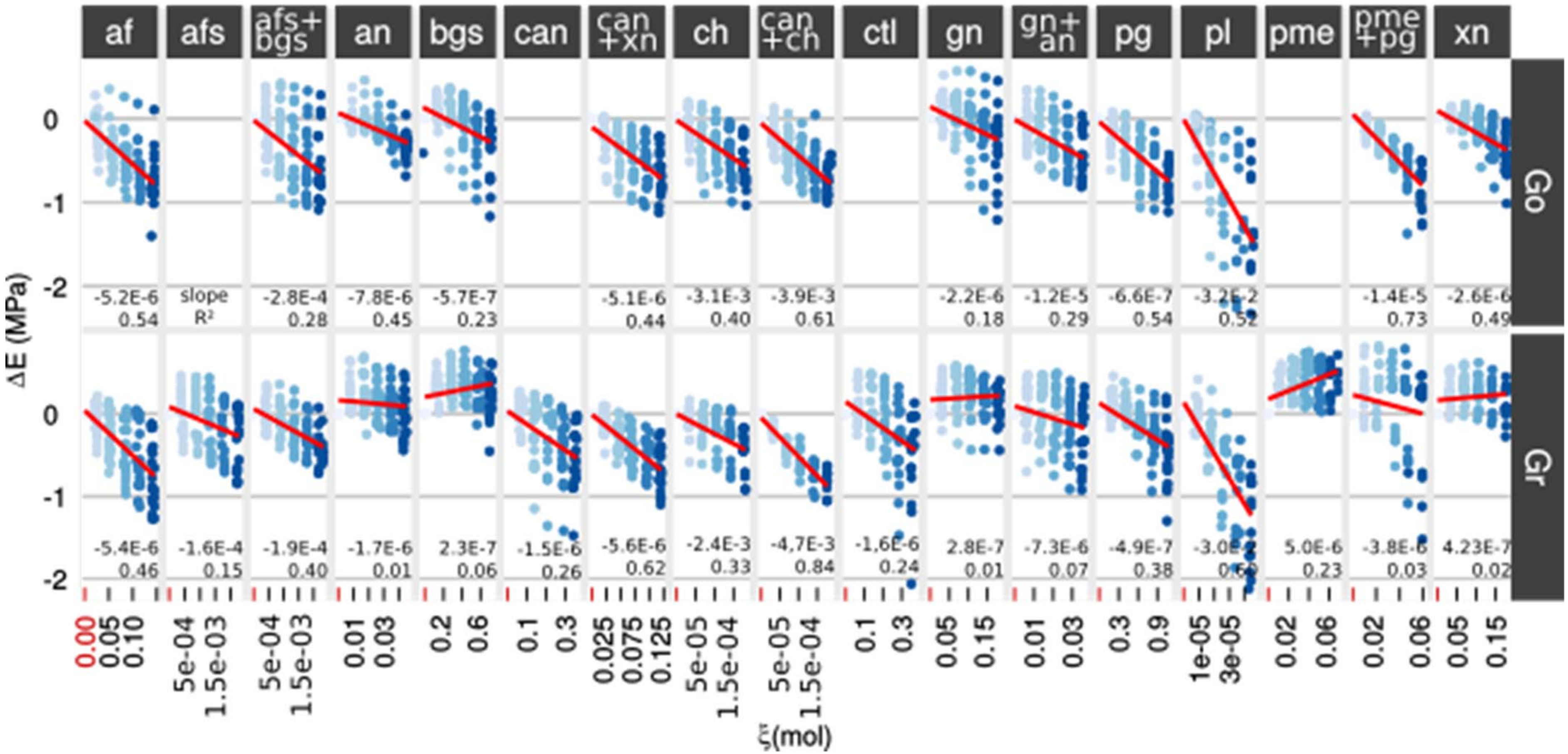
Methods



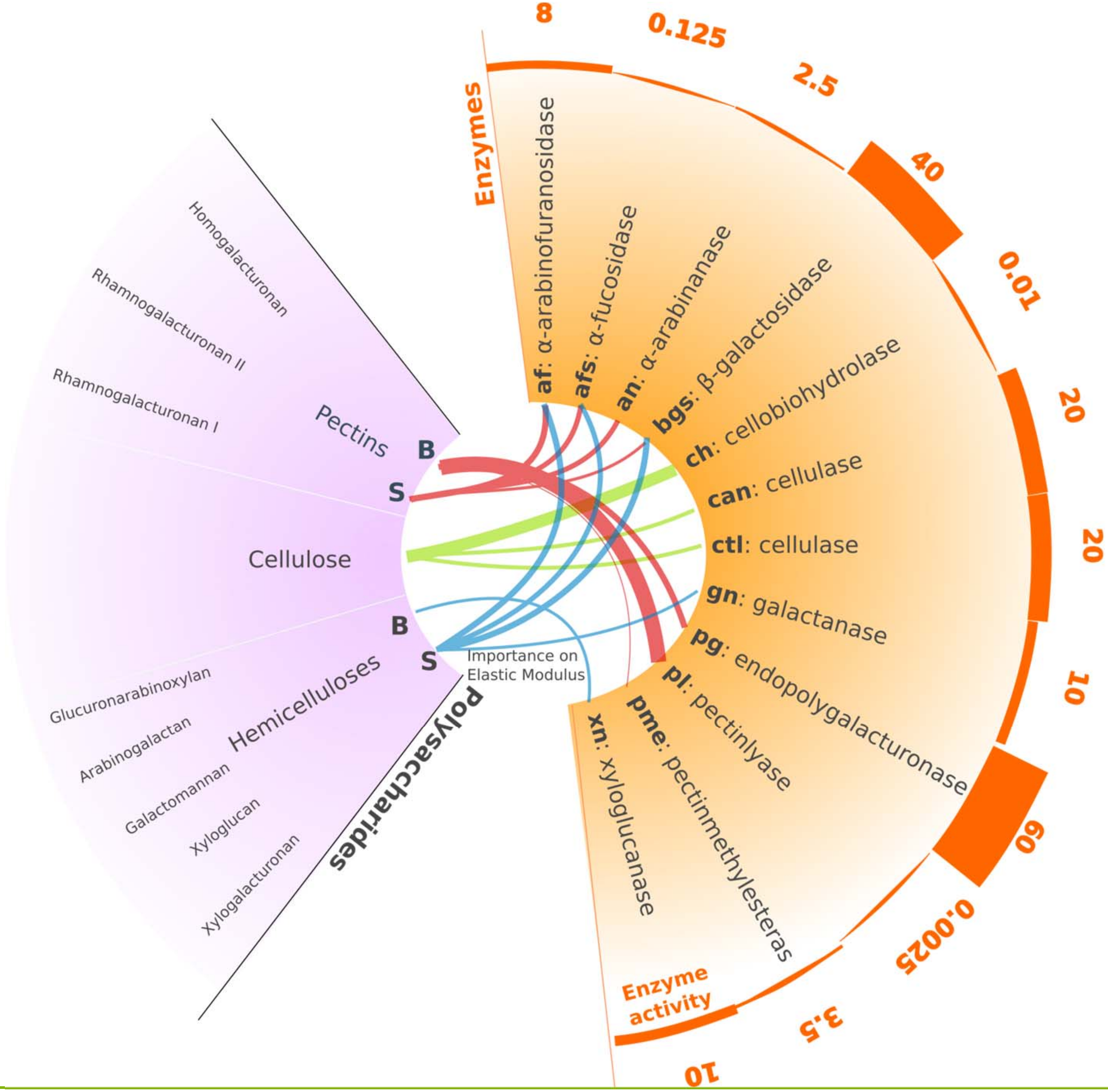
Which polysaccharide family contributes the most to the mechanical properties?



HG, cryst cell, fuc xyg are important for mechanical properties



HG, cryst cell, fuc xyg are important for mechanical properties



af: α-arabinofuranosidase
afs: α-fucosidase
an: α-arabinanase
bgs: β-galactosidase
ch: cellobiohydrolase
can: cellulase
ctl: cellulase
gn: galactanase
pg: endopolygalacturonase
pl: pectinlyase
pme: pectinmethylesterase
xn: xyloglucanase

Role of pectin revisited

Re-constructing our models of cellulose and primary cell wall assembly

Daniel J Cosgrove

Cell wall mechanics and growth control in plants: the role of pectins revisited

*Alexis Peaucelle[1,2], Siobhan Braybrook[3] and Herman Höfte[1]**

[1] Institut Jean-Pierre Bourgin, UMR1318 INRA/AgroParisTech, Saclay Plant Sciences, INRA Centre de Versailles, Versailles, France
[2] Laboratoire MSC, UFR de Physique de Paris 7, Université Paris Diderot, Paris, France
[3] Institute of Plant Sciences, University of Bern, Bern, Switzerland

Pectin Methylesterase, a Regulator of Pollen Tube Growth[1[w]]

Maurice Bosch*, Alice Y. Cheung, and Peter K. Hepler

Biology Department (M.B., P.K.H.), Department of Biochemistry and Molecular Biology (A.Y.C.), and Plant Biology Graduate Program (A.Y.C., P.K.H.), University of Massachusetts, Amherst, Massachusetts 01003

Growth control by cell wall pectins

Sebastian Wolf · Steffen Greiner

Mechano-Chemical Aspects of Organ Formation in *Arabidopsis thaliana*: The Relationship between Auxin and Pectin

Siobhan A. Braybrook[1¤], Alexis Peaucelle[2,3]*

Plant cell wall

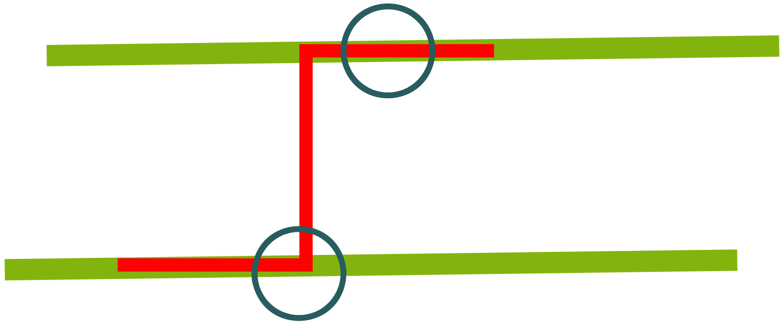
Assembly of new bricks into CW

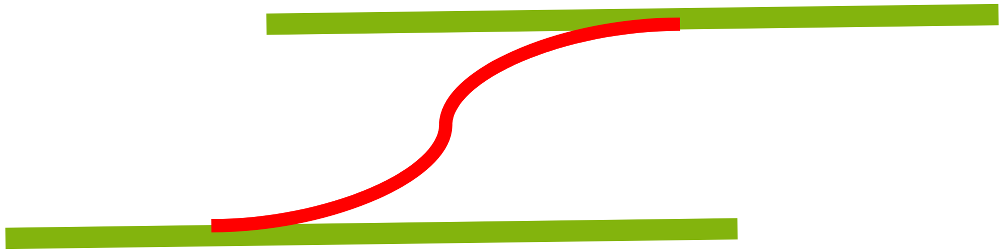
Cell wall lossening mechanism

2 possible mechanisms

slippage

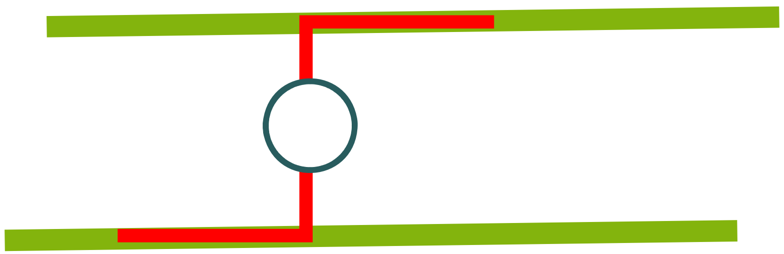
No modification of the stifness





cut & bind

modification of the stifness



Cell wall loosening mechanism

Wall loosening = creep

Expansins

slippage

- α (work @ low pH <-> acid growth auxin induced). Induced creep without reducing wall stiffness
- β. Solubilize matrix polysaccharides (xylan & HG) -> lytic action. Real activity is still fuzzy. ?
- Bacterial. ~ α-expansin

Endoglucanases & endotranglucosidase (Xyg-Cell)

- Cel12A (endoglucanase) ~ similar to α-expansin but slower
- XTH GH9-GH16

cut & bind

Pectin methylesterase and other pectin-modifying enzymes (HG)

- demethylesterase causes wall stiffening

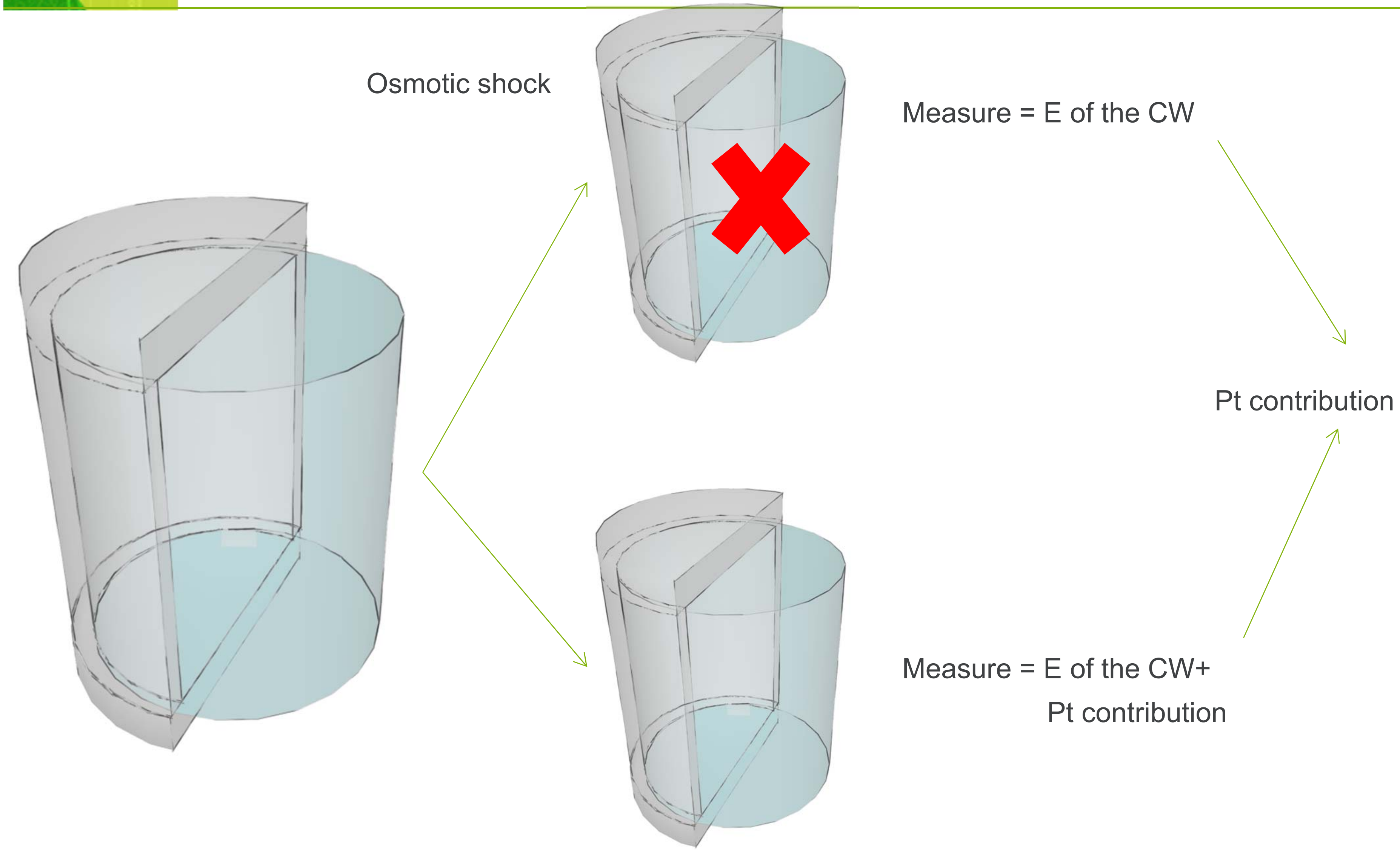
Take home messages

- Irreversible cell expansion is an essential aspect of plant growth and morphogenesis.
- Growth may be highly localised, as in tip-growing cells, or more diffuse growth
- Expansive growth of plant cells requires simultaneous uptake of water into the cell and irreversible expansion of the cell wall.
- Cell growth begins with cell wall loosening which leads to ‘stress relaxation’ of the cell wall which in turn creates the water potential difference needed for water uptake by the cell, resulting in the physical enlargement of the cell.
- Deposition of new polymers to the wall is usually coordinated with surface expansion, but these are separable processes.
- The growing cell wall is composed of a network of cellulose microfibrils embedded in pectins and hemicelluloses that make up the wall matrix;
- these materials combine to form a load-bearing structure that controls cell mechanics and physically controls cell growth.
- Cells growth involves the reshuffling of the cell wall (via proteins)

Plant cell wall

Measuring plant cell wall mechanical properties

Measuring CW properties



Shrinking the hammer: micromechanical approaches to morphogenesis

Pascale Milani[1,2], Siobhan A. Braybrook[3] and Arezki Boudaoud[1,2,*]

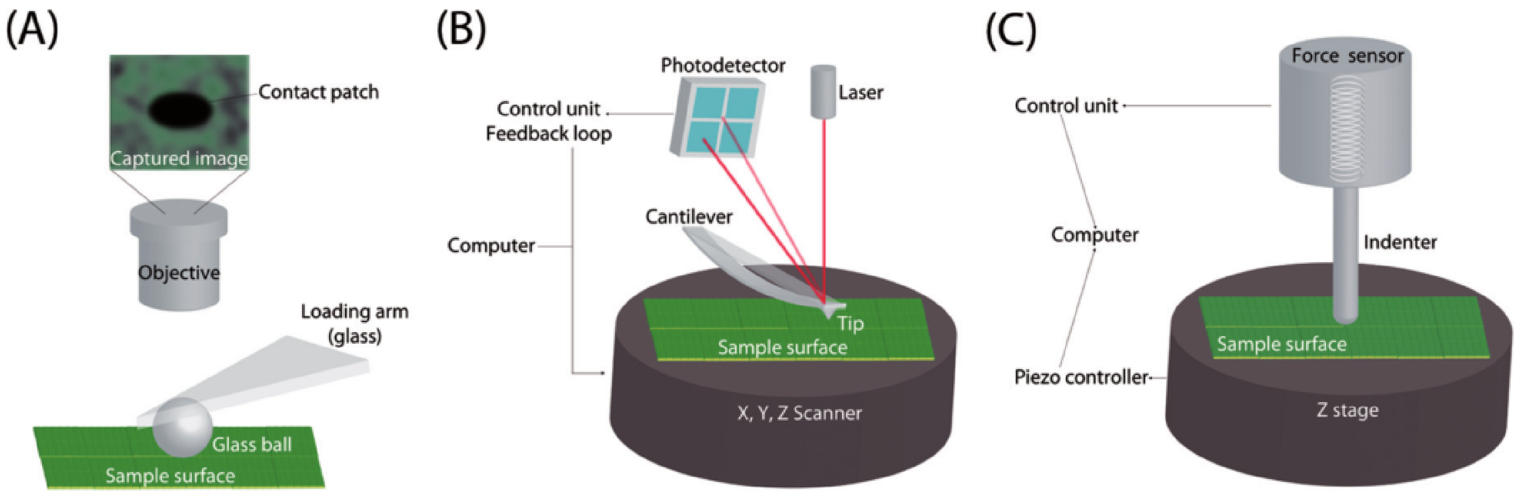


Fig. 1. Schematics of the main indentation set-ups. (A) Ball tonometry. The sample is indented by a glass ball. A microscope objective allows visualization of the contact patch. The load-dependent area of this contact patch is used to determine turgor pressure. (B) Atomic force microscopy. A sharp tip is attached on a flexible cantilever and is used to indent the sample surface. The deformation of the cantilever when a known force is applied on the sample is monitored via a laser beam reflecting from the top surface of the cantilever into a photodetector. This provides force–displacement curves from which are extracted the mechanical properties of the sample. (C) Micro-indentation. A hard micrometric indenter (equipped with a force sensor) penetrates the sample. During indentation, the applied force and depth of penetration are measured. The corresponding force–displacement curves can be used to extract the mechanical properties of the sample. Note that AFM and micro-indenters come in different configurations but with the same working principle.

Shrinking the hammer: micromechanical approaches to morphogenesis

Pascale Milani[1,2], Siobhan A. Braybrook[3] and Arezki Boudaoud[1,2,*]

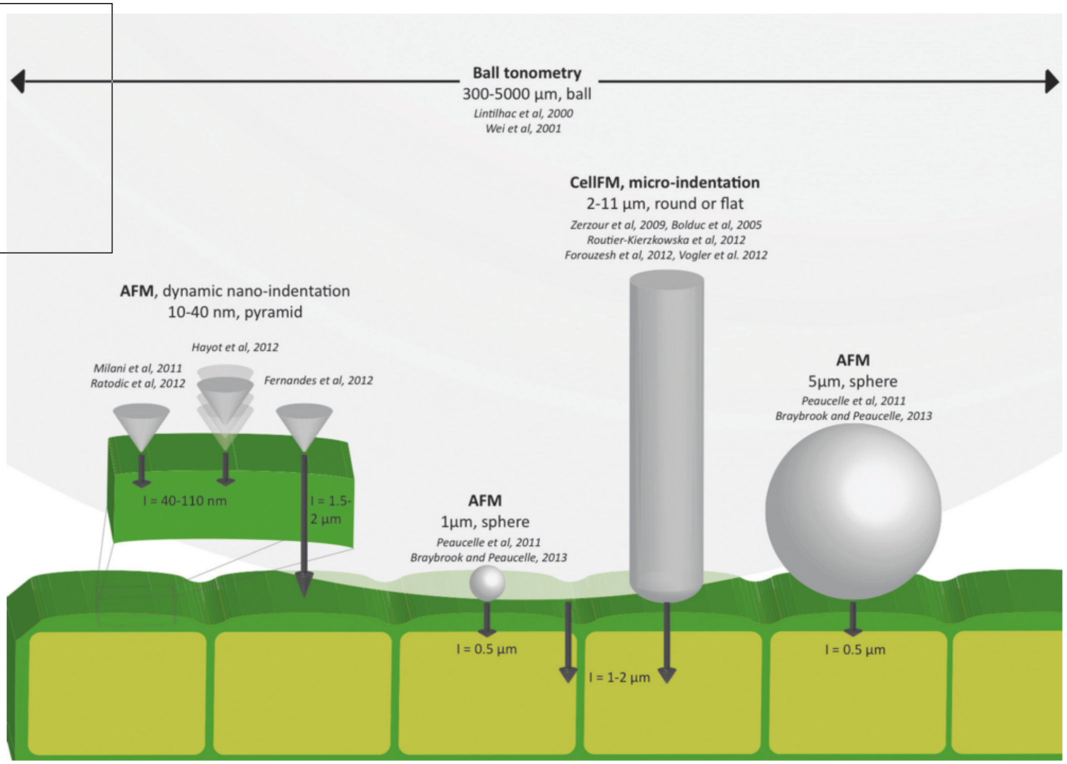


Fig. 2. The panorama of indentation methods applied to plant cells. Schematics of the probes are shown, together with arrows representing typical indentation depths and the reference in which they were used. The cones are proxy for pyramids and the superimposition of cones stands for the dynamic mechanical analysis (DMA) in which the probe is oscillated sinusoidally on top of the vertical translation to the maximal depth. The probe used in ball tonometry is shown for comparison, although it was only used on much bigger cells.

Measuring CW properties

Lab on a Chip

RSCPublishing

PAPER

View Article Online
View Journal | View Issue

Quantification of the Young's modulus of the primary plant cell wall using Bending-Lab-On-Chip (BLOC)

Cite this: *Lab Chip*, 2013, **13**, 2599

Amir Sanati Nezhad,[a] Mahsa Naghavi,[b] Muthukumaran Packirisamy,*[a] Rama Bhat[a] and Anja Geitmann*[b]

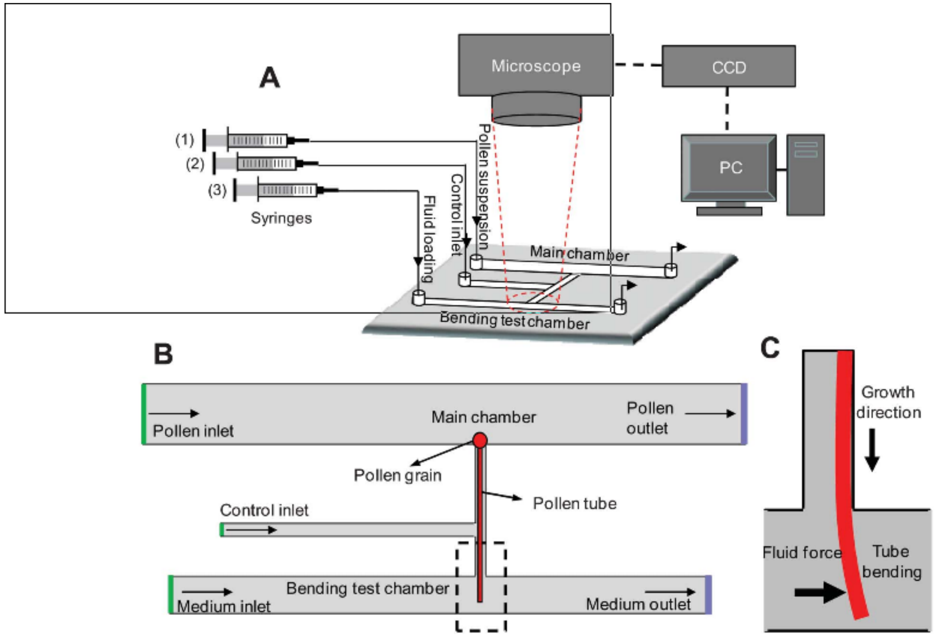


Fig. 2 BLOC design, A) Schematic design of BLOC set-up with three inlets and two outlets for bending test on the pollen tube. B) Details of the BLOC microfluidic network. The design consists of the main chamber to introduce the pollen grain suspension, a growth microchannel into which the elongating tube is guided, a control inlet to position the pollen tube against the opposite side wall and the bending test chamber for the application of flow induced bending force. C) Close up view of pollen tube bending under fluid loading within the bending test chamber.

Plant cell wall

Significant work is still needed

REVIEW PAPER

Plant cell wall extensibility: connecting plant cell growth with cell wall structure, mechanics, and the action of wall-modifying enzymes

Daniel J. Cosgrove*

Department of Biology, 208 Mueller Lab, Pennsylvania State University, University Park, PA 16802, USA

* To whom correspondence should be addressed. E-mail: dcosgrove@psu.edu

Received 18 September 2015; Revised 6 November 2015; Accepted 9 November 2015

Editor: Nadav Sorek, University of California Berkeley

Abstract

The advent of user-friendly instruments for measuring force/deflection curves of plant surfaces at high spatial resolution has resulted in a recent outpouring of reports of the 'Young's modulus' of plant cell walls. The stimulus for these mechanical measurements comes from biomechanical models of morphogenesis of meristems and other tissues, as well as single cells, in which cell wall stress feeds back to regulate microtubule organization, auxin transport, cellulose deposition, and future growth directionality. In this article I review the differences between elastic modulus and wall extensibility in the context of cell growth. Some of the inherent complexities, assumptions, and potential pitfalls in the interpretation of indentation force/deflection curves are discussed. Reported values of elastic moduli from surface indentation measurements appear to be 10- to >1000-fold smaller than realistic tensile elastic moduli in the plane of plant cell walls. Potential reasons for this disparity are discussed, but further work is needed to make sense of the huge range in reported values. The significance of wall stress relaxation for growth is reviewed and connected to recent advances and remaining enigmas in our concepts of how cellulose, hemicellulose, and pectins are assembled to make an extensible cell wall. A comparison of the loosening action of α-expansin and Cel12A endoglucanase is used to illustrate two different ways in which cell walls may be made more extensible and the divergent effects on wall mechanics.

Key words: Atomic force microscopy, Cel12A endoglucanase, cell growth, cell wall, expansin, extensibility, indentation modulus, Young's modulus of elasticity.

ELSEVIER

Available online at www.sciencedirect.com

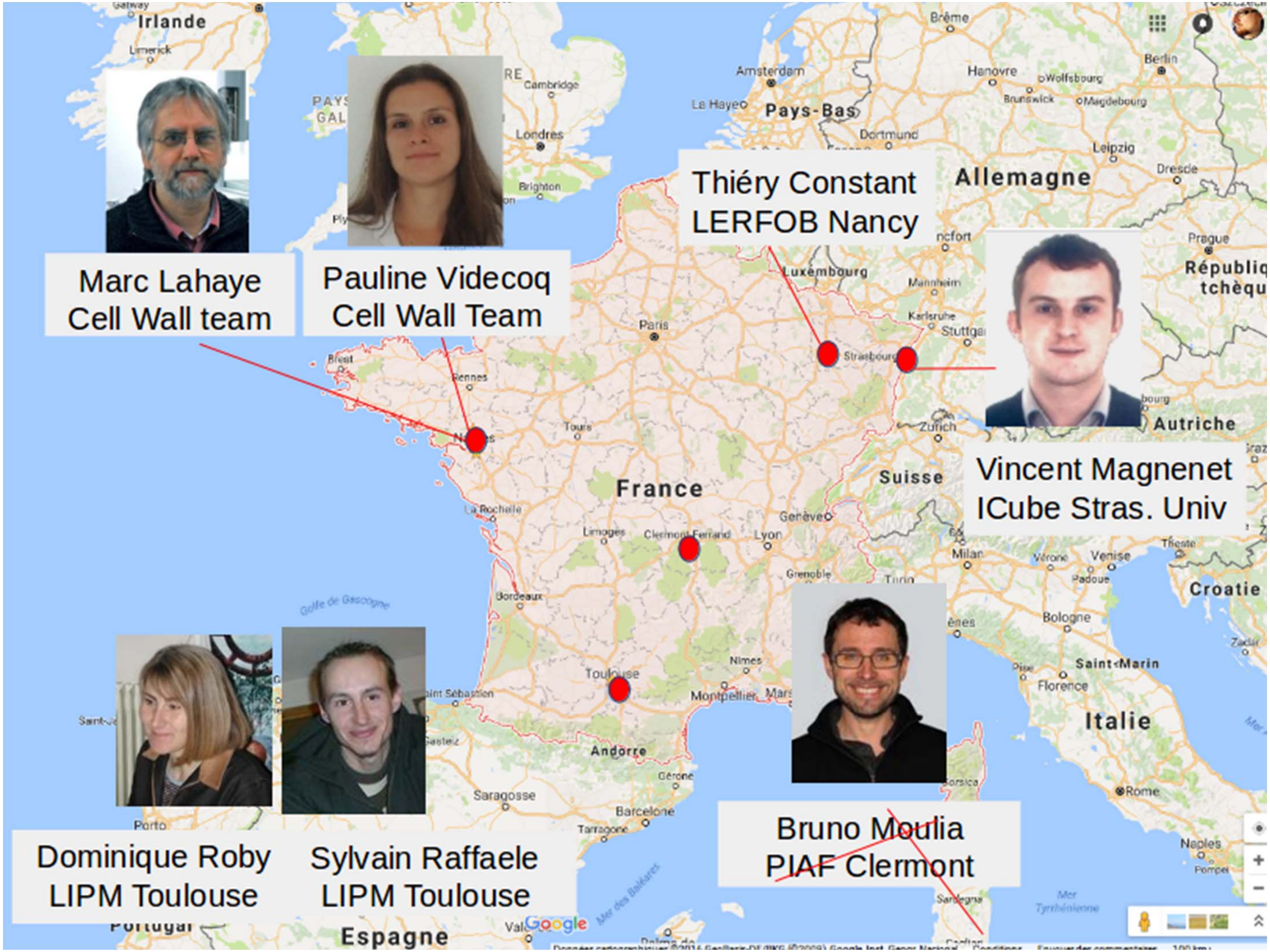
ScienceDirect

Current Opinion in Plant Biology

Understanding plant cell morphogenesis requires real-time monitoring of cell wall polymers

Bara Altartouri and Anja Geitmann

CrossMark



Marc Lahaye
Cell Wall team
Pauline Videcoq
Cell Wall Team
Thiéry Constant
LERFOB Nancy
Vincent Magnenet
ICube Stras. Univ
~~Bruno Moulia~~
~~PIAF Clermont~~
Dominique Roby
LIPM Toulouse
Sylvain Raffaele
LIPM Toulouse
Irlande
Pays-Bas
Allemagne
France
Suisse
Autriche
Croatie
Italie
Andorre
Espagne
Portugal
Paris
Strasbourg
Toulouse
Clermont-Ferrand
Lyon
Bordeaux
Londres

Questions?

Download this slide show @
http://qiplab.weebly.com/