Xyloglucan remodelling enzymes and the mechanics of plant seed and fruit biology

Tina Steinbrecher and Gerhard Leubner-Metzger

Department of Biological Sciences, Royal Holloway University of London, Egham, Surrey TW20 0EX, UK

* Correspondence: Gerhard.leubner@rhul.ac.uk

This article comments on:


The developmental transition from flowers to the mature diaspores (seeds or fruits) depends on cell growth and differentiation (Finch-Savage et al., 2006; Balanza et al., 2016). The plant cell wall is a dynamic nanoscale network for which the classical model and role of xyloglucan–cellulose tethers in wall structure and cell growth was challenged by recent results from genetics, biomechanics, and advanced imaging (Moula, 2013; Cosgrove, 2018; B. Zhang et al., 2021). Xyloglucan (XyG), the predominant hemicellulose, is composed of a β-1,4-glucan backbone that is consecutively substituted with α-1,6-linked xylose residues (Frankova et al., 2013; Pauly et al., 2016). Di Marzo et al. (2022) demonstrated that the MADS-box transcription factor SEEDSTICK (STK) specifically controls seed and fruit biology by α-xylosidase (XYL) mediated XyG remodelling.

Specific cell wall remodelling is decisive for generating the diversity in morphological, biomechanical, and physiological traits of dispersed diaspores during seed and fruit development (Steinbrecher et al., 2017; Landrein et al., 2019; Seale et al., 2020; Arshad et al., 2021; Huss et al., 2021). It is of similar importance in the control of germination timing via dormancy, seed responses to abiotic stresses including heat (thermoinduction), and seedling growth required for plant establishment and survival in a particular environment (Finch-Savage et al., 2006; Shigeyama et al., 2016; Finch-Savage et al., 2017). A representative structural unit of XyG is composed of four β-1,4-linked glucose molecules (backbone) of which three have α-1,6-linked xylose side chains in Arabidopsis thaliana (XXXG; see Box 1 for nomenclature). The xylosyl residues are often modified with β-1,2-linked galactosyl residues which may be additionally α-1,2-linked with fucosyl residues (Box 1). A machinery of specific glycosyl transferases, transglycosidases, and hydroxylases generates the diversity in XyG structures, with XyG α-1,6-xylosyltransferases (XXTs) adding α-Xyl residues, and α-xylosidases (αXYLs) cleaving xylosyl residues from the non-reducing end of XyG cell wall components and XyG oligosaccharides (Frankova et al., 2013; Pauly et al., 2016; B. Zhang et al., 2021). Interestingly, while XyG-deficient A. thaliana xxt mutants exhibit only minor morphological phenotype changes, xyl1 mutants lacking α-xylosidase enzyme activity exhibit altered XyG side chains, free XyG oligosaccharide accumulation, and specific phenotypic defects during reproduction, seed dispersal, germination, and seedling growth. Di Marzo et al. (2022) demonstrate that the expression of the XYL1 gene is directly regulated in developing seeds and fruits by the STK transcription factor.

Box 1 summarizes seed- and fruit-associated morphological, biochemical, biomechanical, and physiological changes of xyl1 and stk mutants, including reduced siliqua elongation growth and increased cell wall stiffness in both, as well as altered XyG side chains, accumulation of free XXXG oligosaccharides, lack of seed dormancy, and increased seed thermostolerance of the xyl1 mutant (Sampedro et al., 2010; Günl et al., 2011; Sechet et al., 2016; Shigeyama et al., 2016; Di Marzo et al., 2022). Likewise, results from bgd10, bgd6 (mum2), axy8, and bgcl1 mutants are presented which have reduced β-galactosidase, α-fucosidase, and β-glucosidase enzyme activities, respectively. They all have cell wall XyG with altered side chains and free XyG oligosaccharide accumulation (Iglesias et al., 2006; Dean et al., 2021).
Box 1. Xyloglucan remodelling and cell wall biomechanics during Arabidopsis thaliana seed and fruit biology

Specific XyG remodelling by a battery of enzymes (A) has profound roles during reproduction, seed dispersal, and germination (B–E). The control of reproduction by the MADS-box transcription factor STK is achieved in part by αXYL-mediated cell wall remodelling (B) combined with other pathways which may differ between seed and fruit development (see cited references and figure 7 in Di Marzo et al., 2022). The control of silique growth (C) by STK, for example, requires XYL1 with a reduced silique size and increased valve cell wall stiffness in both the stk and the xyl1 mutant. There were no obvious morphological phenotype changes observed in axy8 and bglc1 mutants. In contrast to this, bgal and xyl1 mutants exhibited specific seed- and fruit-associated phenotype changes. As for the xyl1 mutant, reduced silique elongation growth was also observed in the bgal10 mutant (Sampedro et al., 2012); however, in contrast to the non-dormant xyl1 mutant seeds, the seeds of bgal10 mutants are dormant. The seeds of bgal6 (mum2) (Dean et al., 2007), stk (Ezquer et al., 2016), and stk/xyl1 mutants are impaired in mucilage production (B), whereas xyl1 mutant seeds have wild-type (WT) phenotype and produce mucilage (Di Marzo et al., 2022). As in the xyl1 mutant, increased cell wall stiffness (C) was also observed in developing seeds of the stk mutant (Ezquer et al., 2016) and may lead to its smaller seed size as well as the defects in seed coat development in that stk, but not xyl1, mutant seeds are impaired in mucilage production (B) and impaired seed abscission [D; from Balanza et al. (2016) with permission (https://doi.org/10.1242/dev.135202)] required for seed dispersal (Balanza et al., 2016). STK seems to achieve this via the MUM2 gene encoding a βGAL6 involved in pectin and possibly also XyG remodelling (Dean et al., 2007; Ezquer et al., 2016). The bgal10 mutant is also reduced in silique growth (C), impaired in seed mucilage production, and XyG remodelling (Sampedro et al., 2012). The production of dormant seeds (E) is not affected in the bgal10 and axy8 (the AXY8 gene encodes an αFUC) mutants, but xyl1 mutant seeds are non-dormant (Sechet et al., 2016). Interestingly, the non-dormant xyl1 mutant seeds are thermoinhibition resistant (E) and have increased hypocotyl cell wall stiffness in creep-extension analysis (Shigeyama et al., 2016). Altered XyG in cell walls and the accumulation of free XyG oligosaccharides (C, E) were associated with the altered fruit and seed phenotypes of the xyl1 (Iglesias et al., 2006; Sampedro et al., 2010; Günl and Pauly, 2011; Sechet et al., 2016; Shigeyama et al., 2016; Di Marzo et al., 2022), bgal10 (Sampedro et al., 2012), axy8 (Günl et al., 2011), and bglc1 (Sampedro et al., 2017) mutants. DAP, days after pollination.
XYL1 and the transcriptional regulation of its expression by STK plays a major role in the control of seed and fruit mechanical properties by XyG remodelling (Box 1); however, depending on the specific process or tissue, other interacting pathways may dominate.

An integrated approach combining genetics with biomechanical and image analysis appears to be important for advancing...
our understanding of XyG remodelling and cell wall mechanics in seed and fruit biology (Sechet et al., 2016; Shigeyama et al., 2016; Di Marzo et al., 2022). Using atomic force microscopy (AFM) to analyse silique valve cell wall stiffness, Di Marzo et al. (2022) demonstrate that developmentally regulated XYL1 gene expression is required for maintaining wall integrity during silique growth. Using creep-extension analysis with elongating stem segments, Shigeyama et al. (2016) reported that xyl1 mutant cell wall stiffness was higher than in wild-type plants. This work also demonstrated that epidermal cells of xyl1 mutant siliques are longitudinally shorter and horizontally enlarged, a finding which fits with the increased cell wall stiffness in xyl1 mutant siliques reported by Di Marzo et al. (2022). Although different biomechanical methods were used, in both cases the same conclusion about the role of αXYL in controlling cell wall mechanical properties (stiffness) was obtained. Interestingly, the silique elongation growth is reduced in XyG-deficient xxt1/xxt2 mutants (Sechet et al., 2016), and the cell wall stiffness tested by microtensile assays of hypocotyls was also decreased compared with the wild type (Cavalier et al., 2008). The importance of the right balance in XyG remodelling enzymes (Box 1) seems crucial, and both XXT-mediated incorporation and αXYL-mediated removal of xylosyl residues can lead to the same biomechanical changes.

The αXYL-catalysed cleavage of xylosyl residues from the non-reducing ends of cell wall XyG chains and XyG oligosaccharides has been shown to be the limiting step in XyG oligosaccharide degradation (Iglesias et al., 2006; Shigeyama et al., 2016; Sampedro et al., 2017). Released XyG oligosaccharides can also alter cell wall properties by incorporation catalysed by XyG endotransglycosylase (XET) enzyme activity (Box 1). In grass caryopses, this may lead to coleorhiza-enforced dormancy due to tissue stiffening (Holloway et al., 2021) and in tomato and other endospermic seeds tissue to weakening of the micropylar endosperm (Finch-Savage et al., 2006; Steinbrecher et al., 2017). XyG oligosaccharides were also proposed to directly or indirectly mediate cell wall signalling which can result in altered hormonal biosynthesis or signalling (Frankova et al., 2013; Pauly et al., 2016; Sechet et al., 2016; Shigeyama et al., 2016; B. Zhang et al., 2021). The structure of XyG differs between plant species especially in diversity of the side chains; however, despite this, conservation in XyG remodelling mechanisms and enzymes was also established (Pauly et al., 2016; Rubianes et al., 2019; Holloway et al., 2021). Mutants in XyG remodelling enzymes, such as in STK and XYL1 in the work of Di Marzo et al. (2022), are indeed highly suited to advance our understanding of the mechanisms of cell wall biochemistry and biomechanics (Box 1).

Within the Brassicaceae, the dimorphic diaspores of Aethionema arabicum offer another interesting approach into cell wall biology during reproduction (Box 2). In Ae. arabicum, the developmental control and plasticity of fruit and seed morphs is associated with morphological, biomechanical, gene expression, and physiological differences between the morphs (Lenser et al., 2016; Wilhelmsson et al., 2019; Arshad et al., 2029, 2021). Comparing the distinct seed and fruit morphs of heteromorphic species therefore provides very interesting systems for future research into cell wall biochemistry and biomechanics including for XyG remodelling enzymes (Box 2).

Environmental conditions play a key role in seed and fruit biology (Finch-Savage et al., 2017; Fernandez-Pascual et al., 2019). Temperature during reproduction can shift the ratios and numbers of the Ae. arabicum fruit and seed morphs (Lenser et al., 2016). Temperature and photoperiod contribute to population fitness by affecting seed coat cell wall properties (thickness, proanthocyanidin content) and thereby dormancy in other species (Finch-Savage et al., 2006; Mizzotti et al., 2014; MacGregor et al., 2015; Fernández Farnocchia et al., 2021).

The cell wall is a highly dynamic and adjustable structure, and its biomechanical properties are determined by specific cell wall compositions for which new modelling approaches are being pursued (B. Zhang et al., 2021; Y. Zhang et al., 2021). Integrating molecular work with morphological and biomechanical analysis, as exemplified by Di Marzo et al. (2022), and further with such novel modelling approaches are promising prospects for future research into this fascinating topic.

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References


Moula B. 2013. Plant biomechanics and mechanobiology are convergent paths to flourishing interdisciplinary research. Journal of Experimental Botany 64, 4617–33.


