1. Title of Case Study: The Molecular Architecture of Plant Cell Walls

2. Grant Reference Number: Contract: PR140003

3. One sentence summary: The arrangement and molecular interactions of polymers within plant cell walls as evidenced by high-field solid-state NMR.

4. One paragraph summary:

Dupree and co-workers have made use of the improved resolution obtained at high magnetic field to investigate cell wall architecture in the model plant *Arabidopsis* and in spruce wood grown in air containing ^{13}C-enriched CO_2. The aim of this study was to understand the molecular basis of plant cell wall properties, such as strength and recalcitrance, which influence the use of plant biomass for timber, paper and pulp, renewable materials and fuel. Wild-type *Arabidopsis,* cell wall-defective mutants and spruce were investigated using multi-dimensional ^{13}C MAS NMR. Specifically, we showed that the structure of xylan is important for assembly of the cell wall. We substantially revised the understanding of softwood molecular architecture, proposing an arrangement of xylan-coated cellulose embedded with mannan and lignin. The work is important for improving the processing and application of wood-derived materials.

5. Key outputs in bullet points:

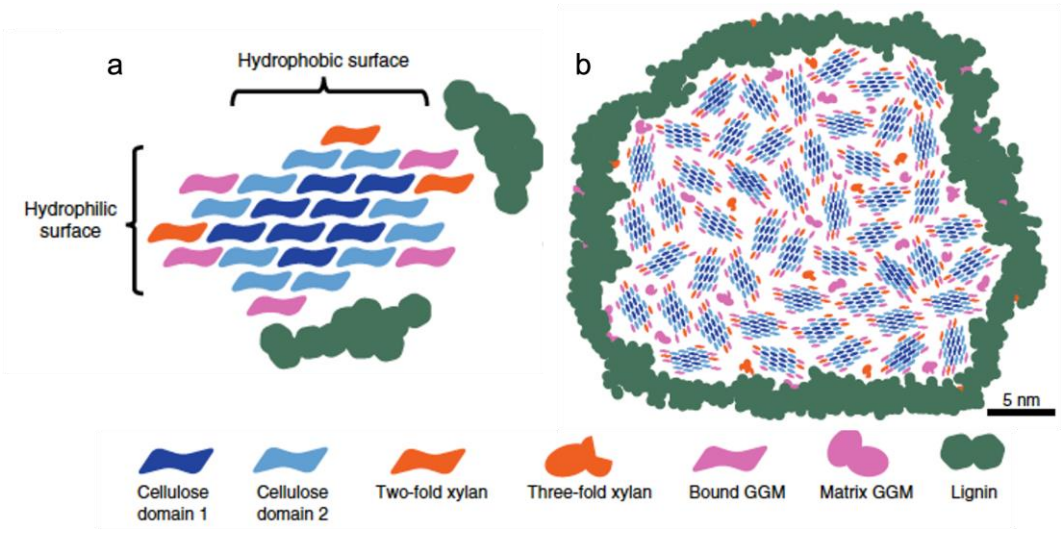
- *A model was derived for the molecular architecture of softwood*
- *Deeper insight into the structure of cellulose fibrils was obtained.*
- *Discoveries will allow increased use and improved applications of woody materials for building construction, energy, materials and food*
- *Training, notably in solid-state NMR of plant cell walls, of a postdoctoral worker Dr Rosalie Thompson and Ph.D. students Oliver Terrett and Jan Lyczakowski*
- *Instigated a new collaboration with Dr Mathias Sorieul, Scion, New Zealand, on NMR studies of wood*
- *Further funding to develop smart sustainable plastics from plants (UKRI: NE/V010565/1)* with Prof J Elliott, Dr J Cullen.

6. Main body text

Materials from plants have been used by humans over millennia for their food, to feed animals, for clothing, and for building construction as timber. As the largest available resource of renewable carbon on the planet, in the future plants are likely to provide sustainable ways to avoid fossil fuel use. However, increased exploitation of this plant cell wall biomass is hampered by our ignorance of the molecular basis for its properties such as strength and digestibility. Cellulose is the main component of the plant cell wall material, and it is present as long, strong fibrils, set like steel reinforcement rods within a mixture of other components. These other components include long chains of other sugars, such as xylan and galactoglucomannan (GGM). Xylan is the principal hemicellulose in many plant secondary cell walls where it binds tightly to cellulose microfibrils. The precise atomic-scale nature of this interaction remains unclear, despite the likely importance in providing strength to timber and in preventing digestion of woody plants.

In this project our aim was to study plant cell walls, to determine their molecular architecture, and to obtain information about how cellulose binds to xylan. We developed techniques to study intact

plant cell walls with solid-state NMR spectroscopy so that information on the shape of the components and the distance between them could be obtained. The very high resolution obtained at the 850 MHz NMR Facility enabled us to distinguish many of the different components in the cell wall. We showed (*Nature Plants* **3** 859–865(2017)) that an even pattern of xylan substitution is critical for its interaction with cellulose in plant cell walls, and were able to present a model (see the figure) for the cell wall architecture of softwood (N*at. Comm*. **10**, 13902 (2019). (This paper is already highly cited being in the top 1% in plant sciences according to the Web of Science)



A possible model of the molecular architecture of softwood. a) A microfibril showing the two cellulose domains with 2-fold xylan and GGM bonded to the surfaces. b) A model of the macrofibril containing groups of cellulose microfibrils with bound GGM and xylan. Lignin is localised mainly to the surface of the macrofibril and interacts predominantly with GGM, xylan and cellulose domain 2

The discovery of how polysaccharides influence plant cell wall assembly provides new principles to understand woody cell wall properties. This method and discovery will now lead to development of better processes for paper and packaging production, and also in improvements to digestion of plant materials for animal feed and bioenergy. Since timber is used widely for building construction, we believe this model will also allow development of better methods for wood modification and preservation.

7. Names of key academics and any collaborators:

Professor Paul Dupree (University of Cambridge)
Professor Ray Dupree (University of Warwick)
Professor Steven P. Brown (University of Warwick)

8. Sources of significant sponsorship (if applicable):

Contract for the High Field Solid State Nuclear Magnetic Resonance Facility (EPSRC)
BBSRC (The BBSRC Sustainable Bioenergy Cell Wall Sugars Programme, and BB/R015783/1)

9. Who should we contact for more information?

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