Pectin self-assembly and its disruption by water: Insights into plant cell wall mechanics

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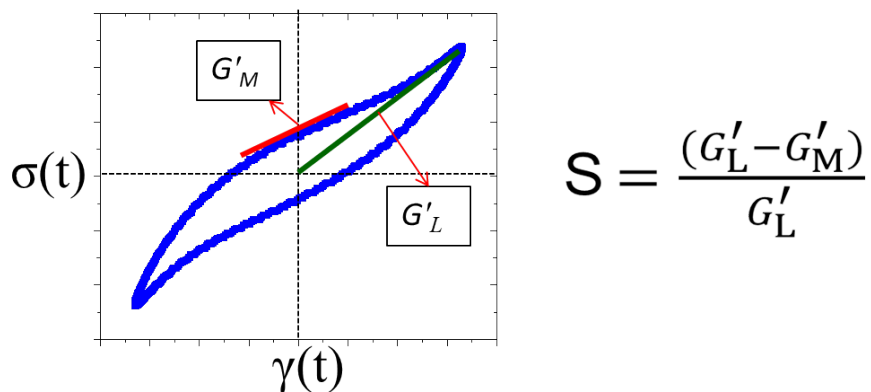
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Figure S1: Storage modulus at large strain (G'_L) and storage modulus at minimum strain (G'_M), within a cycle of oscillation used for calculating the strain-stiffening index, 'S'.

In Oscillatory shear, the mode of deformation involves the application of a cyclic deformation. Generally, a sinusoidal variation of strain, as shown in equation (1) is imposed on the material.

$$\gamma(t) = \gamma_o \sin(\omega t) \tag{1}$$

In the input strain, γ_0 is the strain amplitude and ω is the frequency of the sinusoidal strain. When the strain amplitude (γ_0) is increased from a small to a large value at a fixed ω and progressive transition from linear to non-linear response occurs, the test is called large amplitude oscillatory shear (LAOS). At small strains the output stress is also sinusoidal and has the same frequency as the input strain. For an elastic solid, since the stress is linearly related to strain, the output stress will be in-phase with the strain. Hence the phase lag (δ) is zero. In the case of a Newtonian fluid, stress is proportional to the shear rate and hence the lag is $\pi/2$. For a viscoelastic material, the phase lag is $0<\delta<\pi/2$. Hence, the resulting stress output for a viscoelastic material at small strains is given by,

$$\sigma(t) = \sigma_o \sin(\omega t + \delta) \tag{2}$$

Expansion of Equation 2 results in:

$$\sigma(t) = (\sigma_o \cos \delta) \sin \omega t + (\sigma_o \sin \delta) \cos \omega t \tag{3}$$

Based on Equation 3, material functions storage modulus (G') and loss modulus (G'') can be defined as follows:

$$G' = (\sigma_o/\gamma_o) \cos \delta \tag{4}$$

$$G'' = (\sigma_o/\gamma_o) \sin \delta \tag{5}$$

The in-phase component of the response is called the storage modulus (G'), which indicates the energy stored in the material. The out-of-phase component called the loss modulus (G''), characterizes the viscous behavior and hence the energy that is dissipated. In the non linear regime, the stress response may have contributions from higher harmonics. The stress output in oscillatory shear response at large amplitudes can be represented as,

$$\sigma(t) = \sum_{n=1}^{N} \sigma_n \sin(n\omega t + \delta_n) \tag{6}$$

where, σ_n and δ_n represents the stress amplitude and phase lag respectively of the nth harmonic. Therefore a series of storage modulus and loss modulus corresponding to each harmonics can be defined. Therefore Equation 6 can also be written as,

$$\sigma(t) = \gamma_0 \sum_{n=1}^{N} G'_n \sin(n\omega t) + G''_n \cos(n\omega t) \tag{7}$$

In the linear regime, only the first harmonic exists, n=1. Although, in the non-linear regime, the higher order coefficients exist, rheometers report G'_1 and G''_1 as the measured G' and G''.

However, the time series data, at an imposed strain amplitude and frequency is not based on a single harmonic, rather contains information of all the harmonics. Therefore, the stress vs strain data within a cycle of deformation at steady state can provide more useful information about the non-linear rheological response of a material. Such plots of $\sigma(t)$ vs $\gamma(t)$ within an oscillation are called Lissajous-Bowditch curves, as shown in Figure S1.

Analysing intra-cycle stress vs strain data can provide modulus based on contributions from all the harmonics. Moduli based on the intra-cycle stress-strain data are defined as,

$$G'_M = \left[\frac{d\sigma}{d\gamma} \right]_{\gamma=0} . \tag{8}$$

and,

$$G'_L = \left[\frac{d\sigma}{d\gamma} \right]_{\gamma=\gamma_0} . \tag{9}$$

where, G'_M is the tangent modulus at zero strain and G'_L is the secant modulus at maximum strain. Definition of G'_M and G'_L are schematically shown in Figure S1. In the linear viscoelastic regime, G'_M and G'_L are the same and they are equal to G'. Using the slopes G'_M and G'_L, these changes in material response can be quantified. The strain-stiffening/softening response of a material can be quantified using 'strain-stiffening index', S, which is defined as,

$$S = \frac{G'_L - G'_M}{G'_L} . \tag{10}$$

S greater than zero indicates strain-stiffening and S lesser than zero indicates strain-softening response of a material.
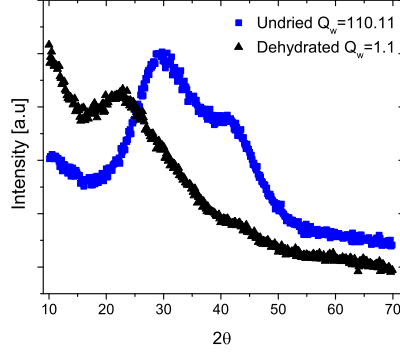

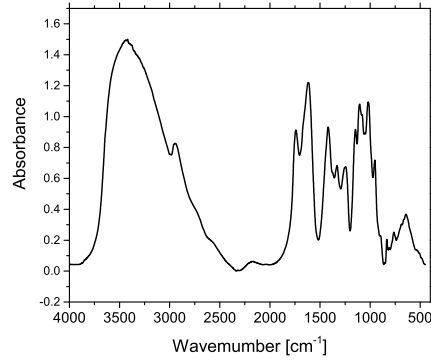Figure S2: X-ray diffraction patterns for undried and dehydrated pectin-Ca gels with R=0.5.

Figure S3: FTIR spectra of the pectin used in this study in the region 3900-500 cm^{-1}

Table 1: β values obtained from fitting the stress-strain data using the Dobrynin model for pectin-Ca gels with R=0.5 at different swelling ratios, Q_w, achieved by dehydration at different dehydration rates.

dehydration rate (k) g/h	Q_w=58.17	Q_w=42.48	Q_w=30.25
0.0033	0.76±0.01	0.74±0.11	0.28±0.05
0.0075	0.69±0.07	0.23±0.08	0.12±0.05
0.0331	0.22±0.08	0.12±0.09	0.13±0.02

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Table 2: β values obtained from fitting the stress-strain data in the first quarter of the Lissajous curves using the Dobrynin model for pectin-Ca gels rehydrated from different extents of dehydration to Q_w=110.11.

dehydration rate (k) g/h	Q_w=58.17	Q_w=42.48	Q_w=30.25
0.0033	0.77±0.03	0.14±0.02	0.08±0.02
0.0075	0.42±0.10	0.25±0.03	0.17±0.01
0.0331	0.17±0.09	0.08±0.05	0.06±0.01

Table 3: Different absorption bands termed as Water matrix Coordinates (WAMACS) and the corresponding bonds that undergo vibrations.

Wavelength (nm)	Bond/species	Description
1343	ν_3 of H$^+$.(H$_2$O)$_3$	asymmetric stretching
1358	H$^+$.(H$_2$O)$_8$	protonated water cluster
1367	ν_1 and/or OH$^-$.(H$_2$O)	symmetric stretching
1371	H$^+$.(H$_2$O)$_5$	protonated water cluster
1382	OH$^-$.(H$_2$O)$_5$ and/or O$_2$$^-$.(H$_2$O)$_5$	water cluster
1395	dehydrating water	-
1408	S$_0$	free water/ ion hydration
1425	polysaccharide hydration	hydration shell
1438	S$_r$ / H$^+$.(H$_2$O)$_2$	protonated water cluster
1447	S$_1$	water molecule with 1 H-bond
1464	S$_2$	water molecule with 2 H-bonds
1475	S$_3$	water molecule with 3 H-bonds
1492	S$_4$	water molecule with 4 H-bonds
1518	OH of H-bonded H$_2$O	symmetric stretching and bending

Table 4: Summary of the analysis of the SANS data for pectin-Ca gels with R=0.5, dehydrated to different extents, at two different dehydration rates (k).

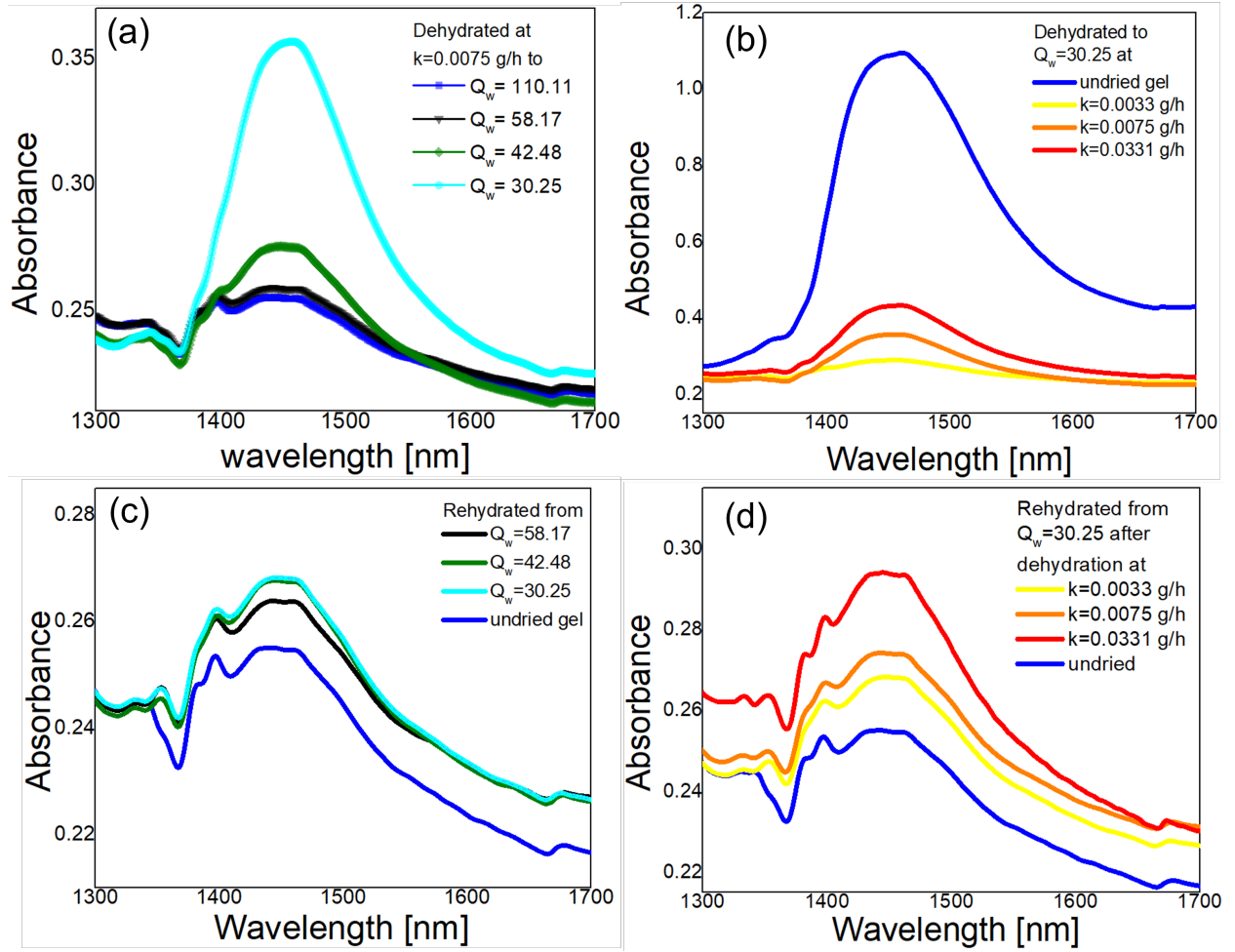| Dehydrated to	k=0.0033 g/h		k=0.0331 g/h	
	d_f	R_g (Å)	d_f	R_g (Å)
Q_w=58.17	1.03	17.7	1.58	35.9
Q_w=42.48	1.12	21.5	1.62	36.1
Q_w=30.25	1.83	31.9	1.78	38.9

Figure S4: NIR spectra of pectin-Ca gels (a) dehydrated at k=0.0075 g/h to different Q_w values. (b) dehydrated to Q_w=30.25 at different dehydration rates (c) rehydrated to Q_w=110.11 after dehydration at k=0.0075 g/h to different Q_w values. (d) rehydrated to Q_w=110.11 after dehydration at different rates to Q_w=30.25.

Table 5: Summary of the analysis of the SANS data for pectin-Ca gels with R=0.5, rehydrated from different extents of dehydration for two different dehydration rates (k).

Rehydrated from	k=0.0033 g/h		k=0.0331 g/h	
	d_{f}	R_g (Å)	d_{f}	R_g (Å)
Q_w=58.17	1.13	20.3	2.03	15.6
Q_w=42.48	1.17	25.1	2.05	15.8
Q_w=30.25	1.21	25.9	2.20	16.1