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SUPPLEMENTARY INFORMATION



Supplementary Figure 1. (A) UV spectra of the different 1 wt% BLKpec solutions mixed with known RGD quantities and the 1 wt% RGDpec solution and (B) the calibration curve obtained using the endpoint readings at 230 nm.



Supplementary Figure 2. Gelation kinetics of PURpec solutions using different ratios between the calcium ions and D-glucono- δ -lactone (GDL) content (R_{Ca-GDL} values of 0.5, 0.25 and 0.125, Equation 2), keeping the R-value constant (R = 1, Equation 1). The bar graph summarizes the cross-over times (δ = 45°, G' < G'').



Supplementary Figure 3. Frequency and amplitude strain sweeps of the BLKpec and RGDpec hydrogels (at 1.5 wt% and 2.5 wt%) after stabilization in cell culture medium for 1h. The frequency sweeps were performed at a constant strain of 1% and the amplitude sweeps were performed at a constant frequency of 0.1 Hz.



οδ

Δ δ (with cells)

• G'

▲ G' (with cells)

С

• G''

▲ G" (with cells)

	1.5 wt%			2.5 wt%		
	cross-over	G' (kPa)	G" (kPa)	cross-over	G' (kPa)	G" (kPa)
	time (min)	after	1h	time (min)	afte	er 1h
RAWpec	~ 20	0.047	0.003	< 2	0.513	0.017
PURpec	~ 2	0.102	0.009	< 1	0.972	0.031
BLKpec	< 4	0.589	0.016	< 2	1.390	0.041
RGDpec	< 4	0.349	0.010	< 2	1.698	0.053
BLKpec (cells) < 5	0.197	0.009	< 3	0.933	0.033
RGDpec (cells	5) < 5	0.188	0.008	< 3	0.792	0.028

Shear Moduli, G' (elastic component) and G" (viscous component) (kPa)

10

0.01

0.001

90

Phase

s Angle, δ (°)

G" (BLKpec)

• G" (RGDpec)

120

2.5 wt%

60

Time (min)

30

o δ (BLKpec)

δ (RGDpec)

90

- 90

Phase

Angle, δ (°)

10

120

Supplementary Figure 4. Gelation kinetics of the different pectin solutions, at two different final concentrations (1.5 wt% and 2.5 wt%). (A) RAWpec vs. PURpec, (B) BLKpec vs. RGDpec, and (C) BLKpec vs. RGDpec, with and without embedded human mesenchymal stem cells (hMSCs) (at 8 x 10⁶ cells/mL). Table (D) summarizes the main data from the gelation kinetics studies.

MESH SIZE OF PECTIN HYDROGELS

The mesh size (ξ) of pectin hydrogels (after being swollen in cell culture medium at 37°C), was calculated using rheometry data to obtain the molecular weight between crosslinks (M_c) [S1]. The following figure illustrates these two parameters:



ADAPTED FROM: Zaidel et al. Biocatalysis and Agricultural Biotechnology, 2012, 1, 207-219 and Pourjavadi et al. Starch, 2009, 61, 173-187

Immediately after the ionic crosslinking, the pectin matrices were (1) incubated in cell culture medium for 24h at 37°C to reach the swelling equilibrium [S2], (2) weighed, (3) freeze-dried, and (4) weighed again. The swelling ratio (q_F) and polymer volume fraction (v_2) were calculated using the following equations [S3]:

 q_F = (mass of the hydrogel after swelling) / (mass of the hydrogel after freeze-drying)

where ρ_P is the density of pectin (approximately 1.515 g/cm³ [S4]) and ρ_S the density of the cell culture medium at 37°C (0.99 g/ cm³). The molecular weight between crosslinks (M_c) was calculated from the value of the shear modulus elastic component (G', Pa) of the swollen hydrogels using the following equation [S1]:

$$M_c = c_p RT / G'$$

where c_p is the concentration of pectin in solution (1.5wt% = 15000 g/m³ or 2.5wt% = 25000 g/m³), R is the gas constant (8.314 m³.Pa.mol⁻¹.K⁻¹), and T is the temperature at which the measurement was performed (310.2 K). The mesh size (ξ) was then calculated as [S1]:

$$\xi = v_2^{-1/3} / (2 \text{ M}_c/\text{M}_r)^{1/2} \text{ C}_n^{1/2}$$

where M_r is the molecular weight of the monomeric galacturonic acid unit (194 g/mol [S5]), *I* is the length of the repeating unit (4.35Å [S5]), and C_n is the characteristic ratio ($C_n = 0.021M_n + 17.95$ calculated as for alginate [S6], considering the well-known analogies between the two polyuronides). For the studied hydrogels, the mesh size is $\xi_{1.5wt\%} \approx 707$ nm (±12.4) and $\xi_{2.5wt\%} \approx 380$ nm (±0.6).

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