

1 Article

# 2 Synthesis and application of scaffolds of 3 Chitosan-Graphene oxide by the Freeze-drying 4 method for tissue regeneration

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## 17 1. Experimental

### 18 1.1. Deacetylation degree and characterization of CS by potentiometric method

19 For the DD calculation, a potentiometric titration of CS with a WTW pHmeter pH 3310 was  
20 performed, a NaOH solution of 0.101 M previously standardized with C<sub>8</sub>H<sub>5</sub>KO<sub>4</sub> as titrating agent  
21 was used and a solution of 0.253 g of CS in 20 ml HCl 0.3M.

### 22 1.2. Capillary viscosimetry for the calculation of the M<sub>v</sub>.

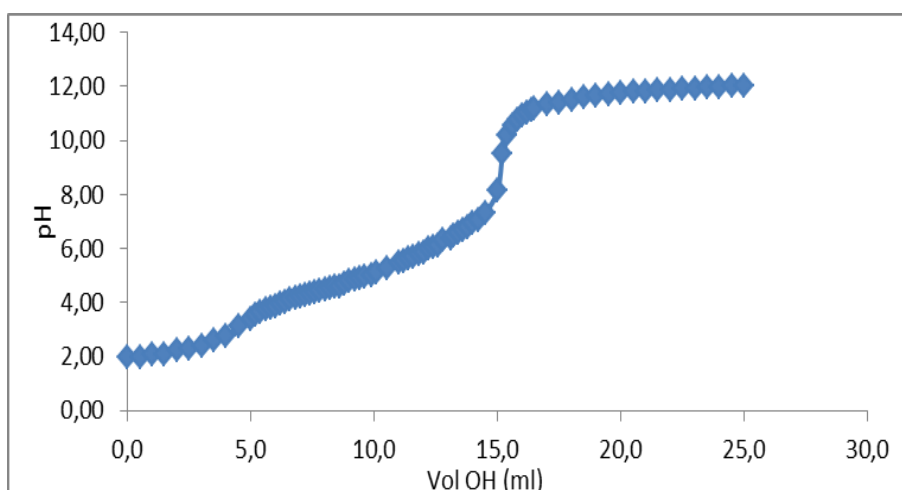
23 A standard solution of 100 mL of CH<sub>3</sub>COOH 0.1 M, NaCl 0.2 M and 0.02 g/mL of CS was prepared  
24 for the calculation of the M<sub>v</sub> of CS. From this 5 solutions of 25 mL were prepared with the  
25 concentrations shown in figure 3 and the density was measured with a pycnometer of 1 mL and the  
26 fall time with a viscometer of ubbelohde viscometer with constant β of 3.31x10<sup>-5</sup> that is calculated  
27 with the time of fall of the water.

## 28 1. Supporting tables

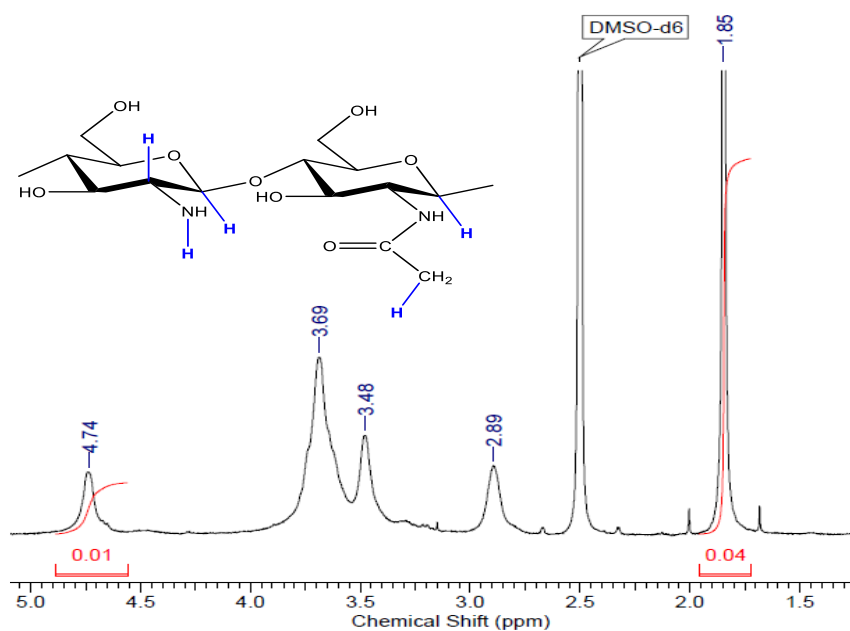
29 **Table S1.** Concentrations and drop times of solutions taken in an ubbelohde viscometer.

Concentration (mL/g)	Time (s)	ρ (g/mL)
Solvent	277	1.0320
0.0008	279	1.0324
0.0010	282	1.0327
0.0013	283	1.0332
0.0015	284	1.0348
0,0018	285	1.0360
0.0020	286	1.0368

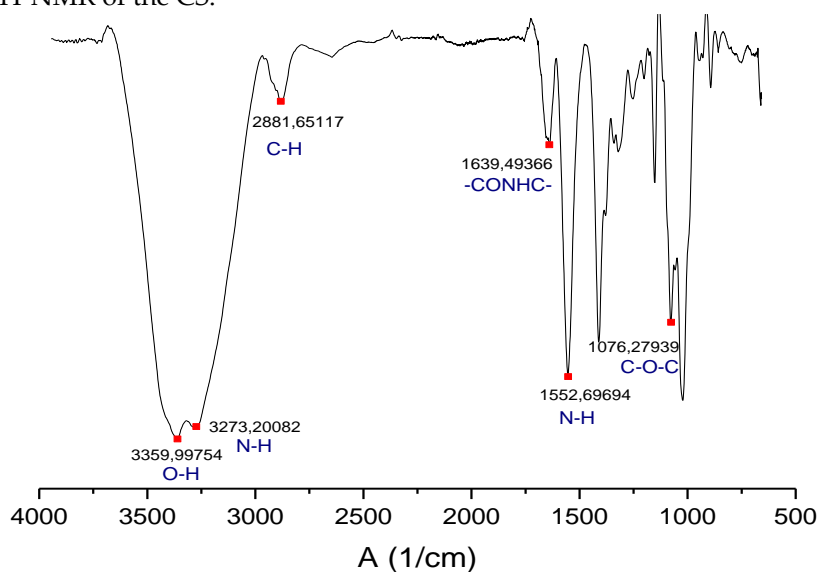
## 30 2. Supporting images



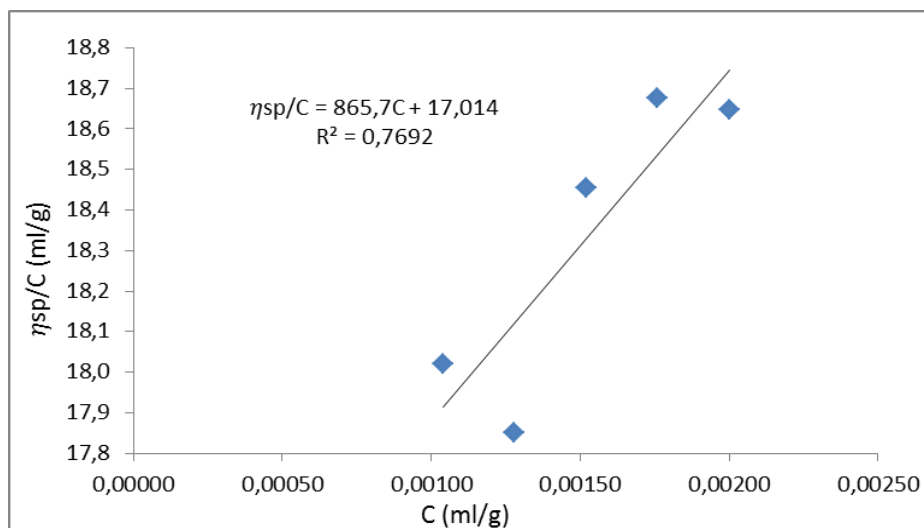
31 **Figure S1.** Potentiometric titration of the CS.



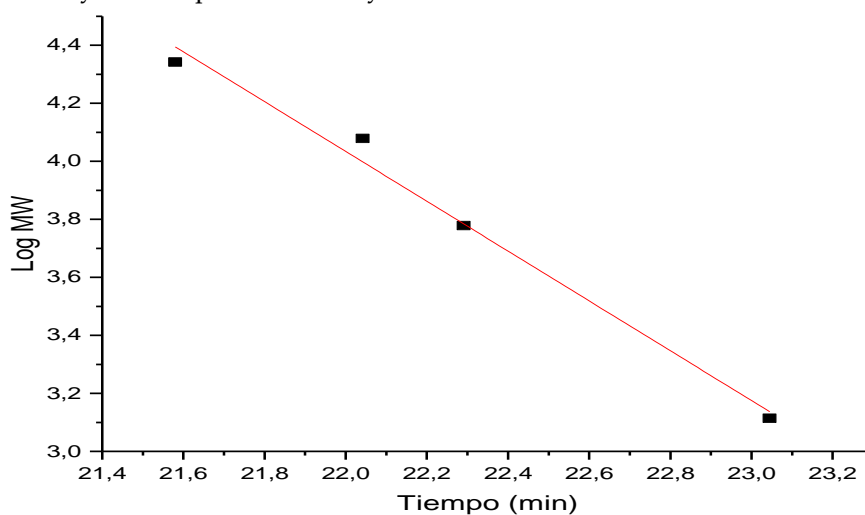
32 **Figure S2.**  $^1\text{H-NMR}$  of the CS.



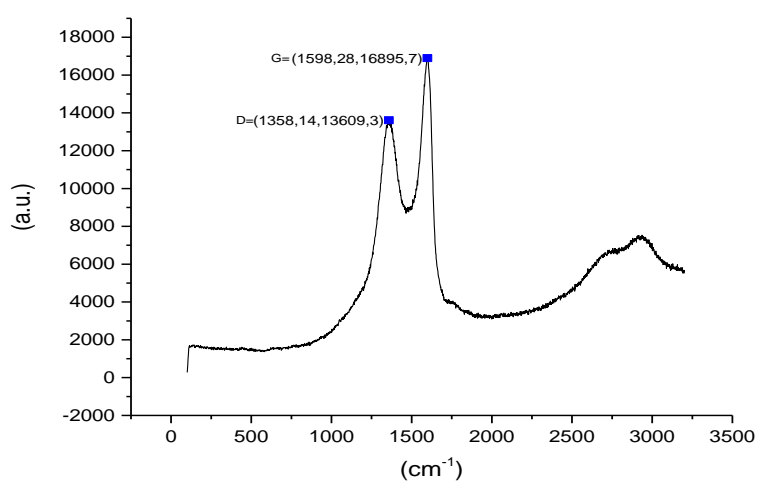
33 **Figure S3.** FTIR of the CS.



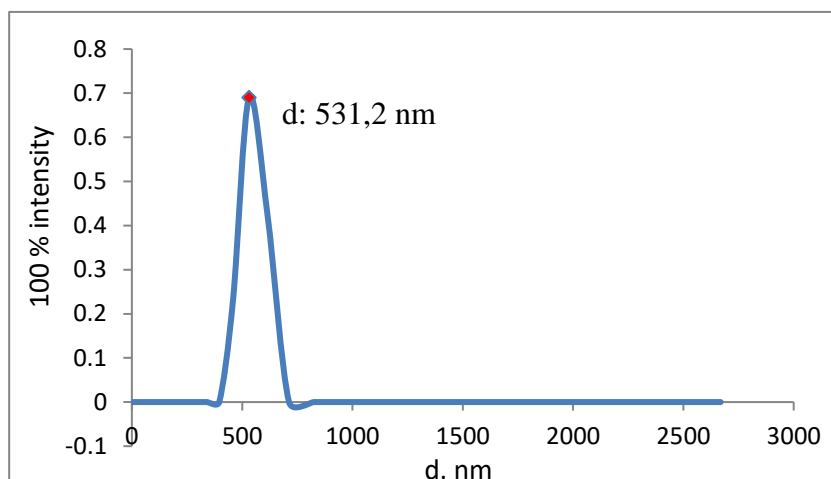
34 **Figure S4.** Viscosity curve. Specific viscosity vs concentration.



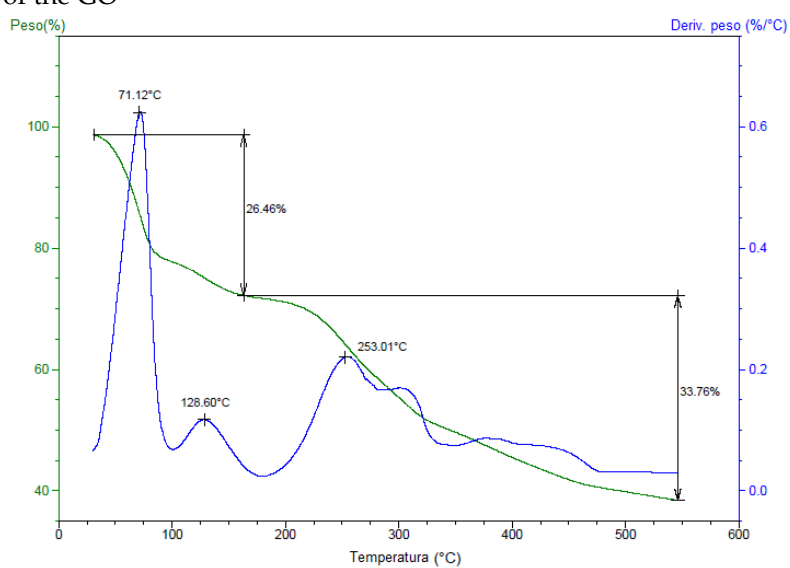
35 **Figure S5.** Calibration curve of GPC with pululan standards.



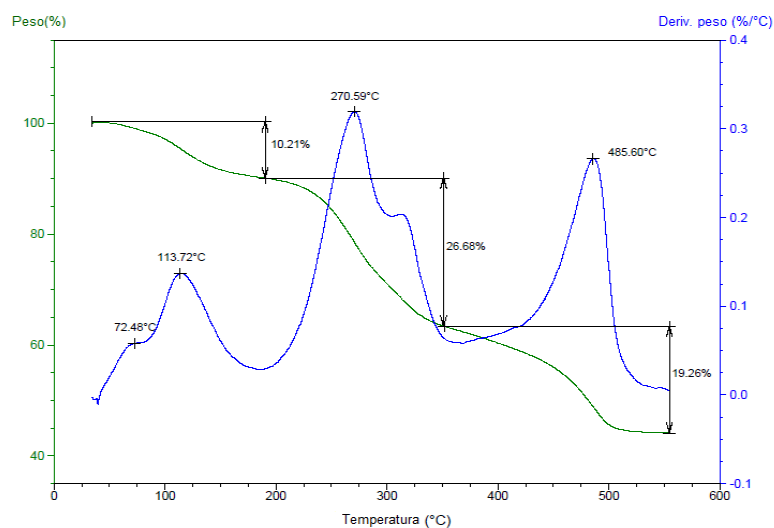
36 **Figure S6.** Raman spectrum of the GO



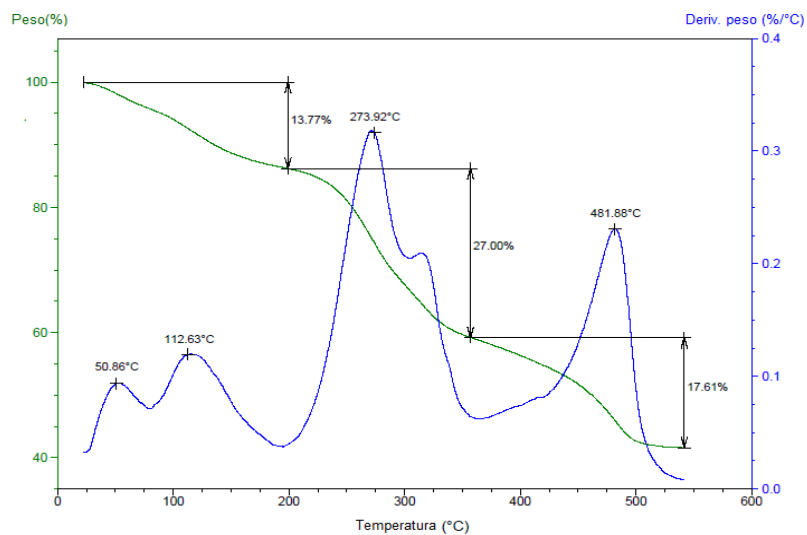
37 Figure S7. DLS of the GO



(a)

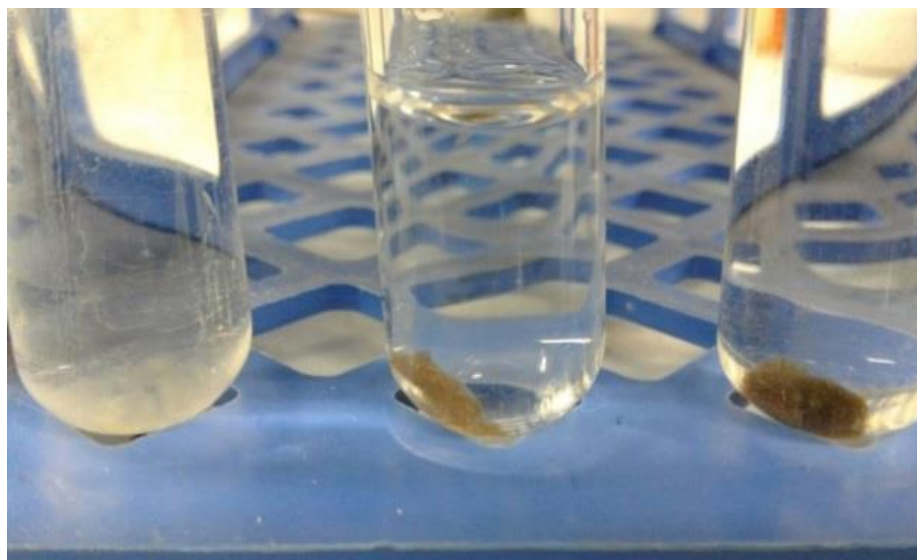


(b)



(c)

38 **Figure S8.** Thermogravimetric curve of CS scaffolds (a) without GO, (b) with 0.5% GO and (c) with  
39 1.0% GO.



(a)

(b)

(c)

40 **Figure S9.** Degradability test of CS scaffolds in physiological serum (a) without GO, (b) with 0.5%  
41 GO and (c) with 1.0% GO added.