

Supporting informations

A natural multifunction and multiscale hierarchical matrix as a drug-eluting scaffold for biomedical applications

Gabriela Graziani,^{1,1,§} Carla Triunfo,^{1,2} Giulia Magnabosco,^{1,2,&} Simona Fermani,^{2,3} Devis Montroni,² Daniele Ghezzi,⁴ Martina Cappelletti,⁴ Nicola Baldini,^{*,1,5} Giuseppe Falini^{*,2}

¹Biomedical Science, Technologies, and Nanobiotechnology Lab, IRCCS Istituto Ortopedico Rizzoli, Bologna, Italy.

²Department of Chemistry “Giacomo Ciamician”, University of Bologna, Bologna, Italy. ³Interdepartmental Centre for Industrial Research Health Sciences & Technologies, University of Bologna, 40064 Bologna, Italy. ⁴Department of Pharmacy and Biotechnology, University of Bologna, Bologna, Italy. ⁵University of Bologna, Department of Biomedical and Neuromotor Sciences, Bologna, Italy.

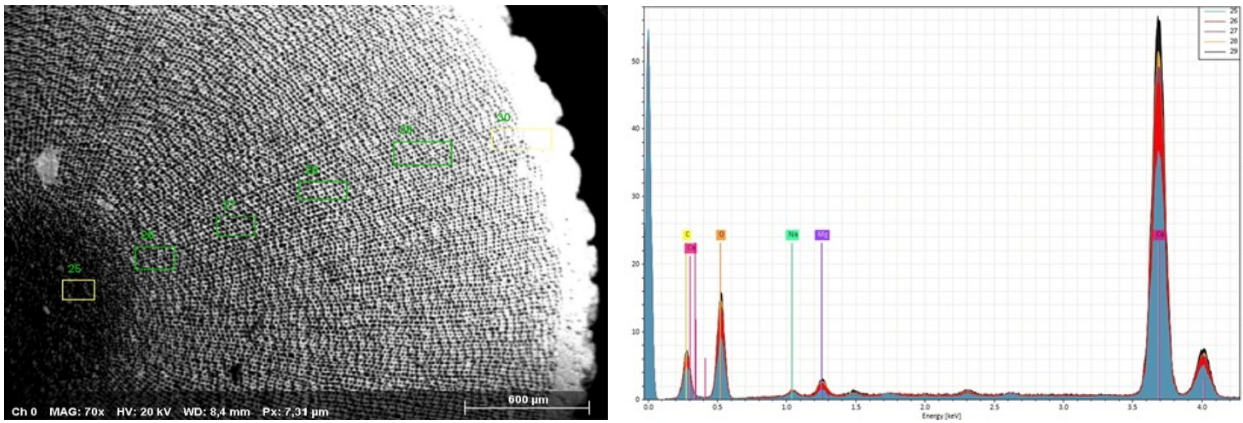


Figure S11. (left) Scanning electron microscopy image using back scattered signal. The squares indicate the regions where EDX spectra were collected. (right) EDX spectra from the squares indicated in the left image.

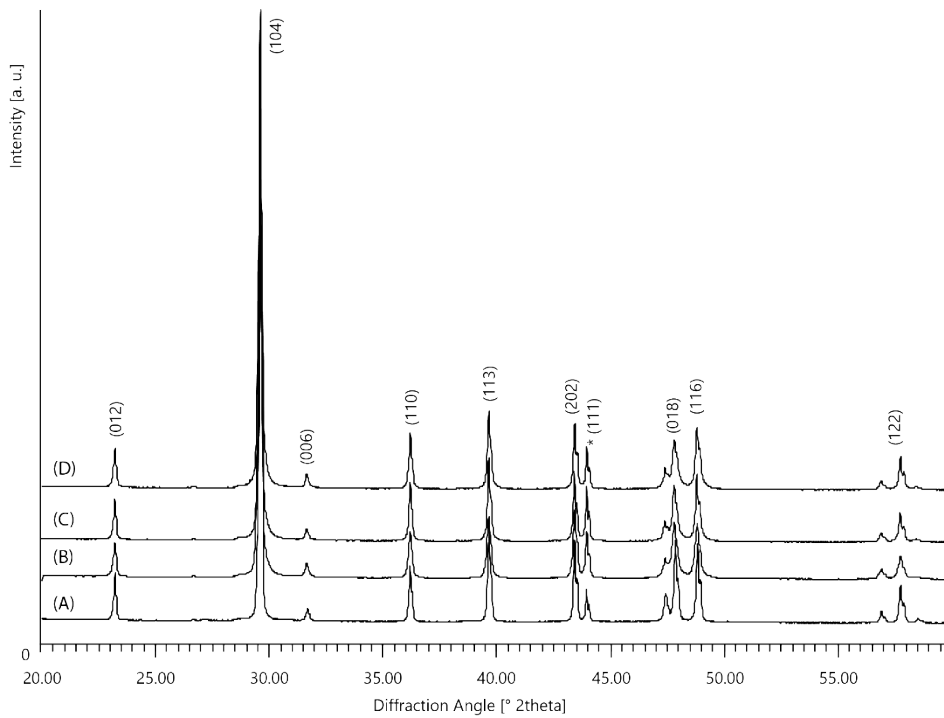


Figure S12. X-ray diffraction patterns of powders from *P. imperialis* spines before (A) and after thermal treatment at 250 $^{\circ}$ C for 8 hours (B), 24 hours (C) and 48 hours (D). Only Mag-calcite was detected. The * symbol indicates the diffraction peak of diamond, which was added as an internal standard. The Miller indices of calcite are reported.

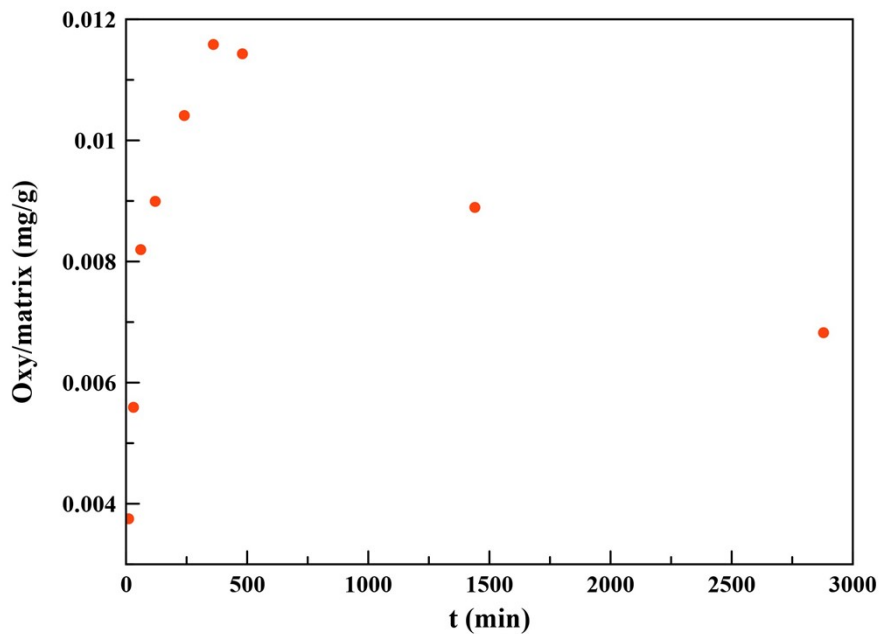


Figure SI3. Isotherm of the desorption kinetics of oxytetracycline from thermally treated sea urchin spines in a PBS solution. The samples were kept in dark conditions to minimize the potential for photodegradation.

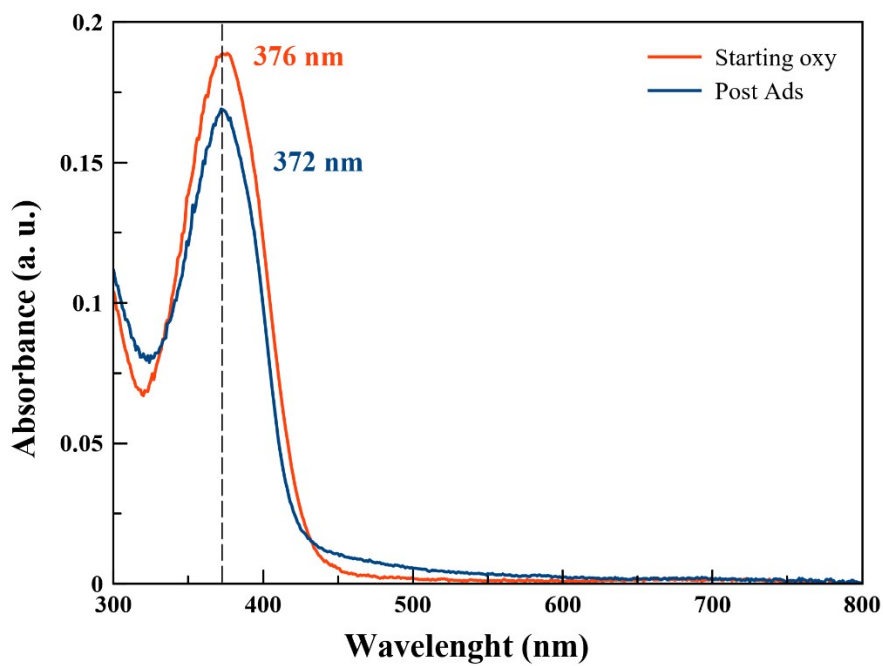


Figure SI4. Visible absorption spectrum of oxytetracycline in PBS before (red line) and after (blue line) adsorption on the sea urchin spine matrix. The profile of the adsorption band remains unchanged.

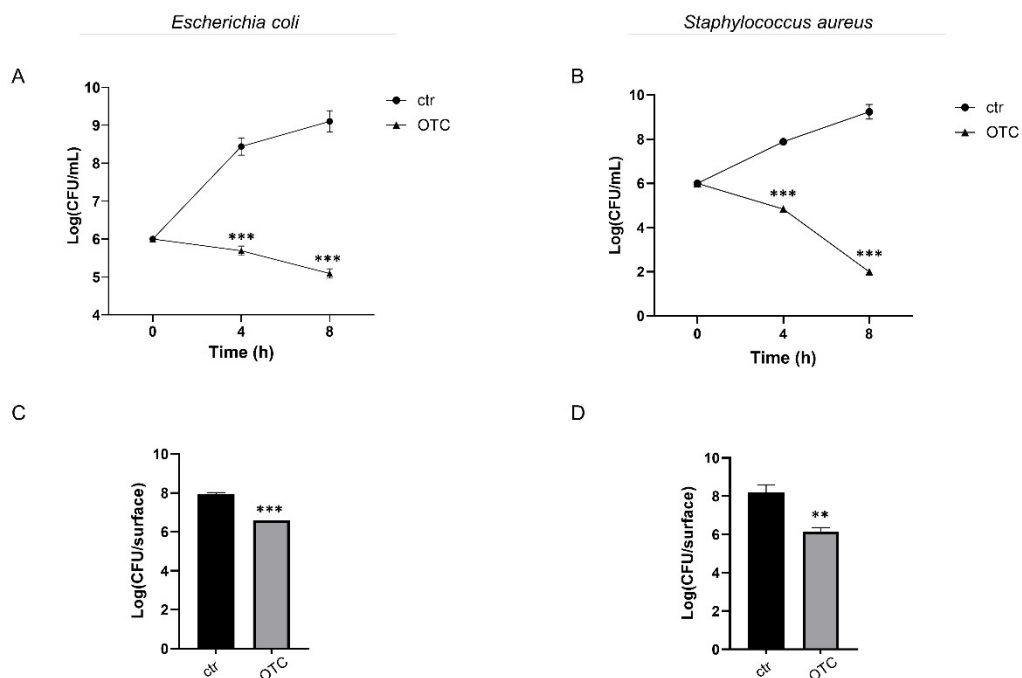


Figure SI5. Planktonic growth and adhesion capacity of *Escherichia coli* ATCC 8739 (panels A and C) and *Staphylococcus aureus* ATCC 6538P (panels B and D) inoculated in the culture medium added with oxytetracycline at the same concentration used to load the sea urchin spines. Significance is indicated as follows: ** $p < 0.01$ and *** $p < 0.001$.

Table SI1. The crystallographic lattice parameters of Mg-calcite of *P. imperialis* samples measured after being thermally treated at 250 °C for different lengths of time. The quantity of amorphous calcium carbonate (ACC) present was also reported.

Sample	a-axis (nm)	c-axis (nm)	ACC (wt %)
0 h	0.49629 ± 0.00002	1.6948 ± 0.0001	8.4 ± 0.6
8 h	0.49667 ± 0.00002	1.6977 ± 0.0001	5.1 ± 0.7
24 h	0.49662 ± 0.00003	1.6977 ± 0.0001	4.8 ± 0.8
48 h	0.496670 ± 0.00003	1.6980 ± 0.0002	4.9 ± 0.6

Table SI2. Maximum compressive strength (σ_c) and Young's modulus (E) from the compression tests of spines from *P. imperialis* before (A) and after the thermal treatment at 250 °C for 8 hours (B), 24 hours (C) and 48 hours (D). At least 10 specimens were used for each sample set.

Sample	Time	σ_c	E
	(hours)	(MPa)*	(GPa)*
A	0	100 ± 20	1.9 ± 0.7
B	8	120 ± 10	2.1 ± 0.7
C	24	120 ± 20	1.9 ± 0.4
D	48	90 ± 20	1.8 ± 0.5

*These differences are not statistically significant.