

Supporting Information

On-demand radial electrodeposition of alginate tubular structures

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Materials and Methods

Materials

For use in electrodeposition, alginate was prepared as a solution of 2% (w/v) alginate (Sigma Aldrich, St. Louis, MO) and 0.9%(w/v) NaCl within Dulbecco's phosphate buffered saline (DPBS). Divalent cation particles (CaCO_3 or BaCO_3) were prepared in DPBS, at concentrations corresponding to the desired experiment. To form the electrodeposition solution, alginate solution and divalent cation particle suspensions were mixed in equal parts. To ensure uniform dispersion of the particles (*i.e.*, CaCO_3 or BaCO_3), the mixture was sonicated prior to each deposition experiment. For electrodeposition, a DC power supply (DigiKey), used to control the applied voltage, was wired in series with an Arduino, which served as a 'trigger' and to control deposition duration. Copper wires (diameters 1.00, 0.50, and 0.25 mm) were utilized as electrodes (cathode and anode) during electrolysis (Sigma Aldrich).

Optical Coherence Tomography

Optical coherence tomography (OCT) is a well-established, 3D imaging modality, that enables micrometer resolution of biological structural features [1,2]. Herein, we employed this imaging modality to structurally assess electrodeposited alginate gels. OCT imaging was performed, as previously described [3]. Briefly, a commercial spectral domain OCT system (TEL220C1; Thorlabs Inc.), with a maximum sensitivity of 101 dB and a maximum A-scan line rate of 76 kHz, was used for all imaging. The OCT probe was positioned orthogonally above the sample, and scans were collected along the alginate tube, utilizing a 5.5 kHz line rate (highest sensitivity), with 1024 A-scans per B-scan. Individual B-scans were assessed within the ThorLabs software, using the 'measure' function, to determine the vessel wall thickness and lumen diameter. 3D video renderings of these scans were exported from the ThorLabs software, and are provided for visualization in the Supplemental Videos section.

Logistical curves

OCT-derived measures of electrodeposited gel thickness were plotted vs time, and analyzed using a logistic function-fitting tool in MATLAB (Math-Works Inc., Natick, MA) [4]. This function takes the inputs, time (independent variable) and the thickness (dependent variable), and outputs, $t_{1/2}$ (time to half thickness), Q_{inf} (the gel thickness as t approaches infinity), and alpha (time decay constant), with a 95% confidence interval. These data are summarized in Table 1S.

Statistics

Statistical analyses were performed within MATLAB (Math-Works Inc., Natick, MA). Bar graphs were utilized for exploratory analyses to visualize means, variance, and distributions, and subsequently checked for normality using Anderson-Darling tests. All data sets examined were normally distributed, with groups of comparable sizes, and equal variances, justifying the use of parametric statistical analyses. Significant differences within the data between variables were identified by one-way ANOVA (analysis of variance, using the built-in "anova1" function), with Tukey's HSD post-hoc

analysis (Tukey's Honestly Significant Difference). In all cases, $p < 0.05$ for statistical significance.

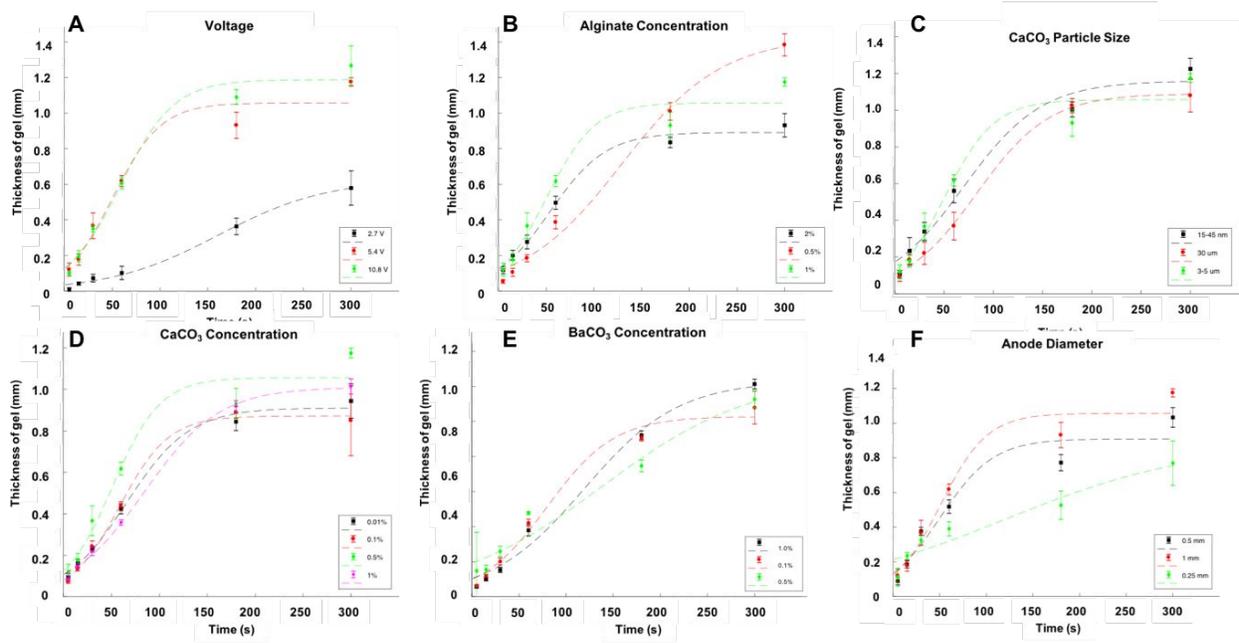


Figure S1. Logistical curves fitted to the deposited gel thickness data for applied voltage (A), alginate concentration (B), CaCO₃ particle size (C), CaCO₃ concentration (D), BaCO₃ concentration (E), and anode diameter (F).

Table S1. Coefficients of the fitted logistical curves from time vs gel thickness data.

$Q(t) = Q_{inf}/(1 + e^{-\alpha(t-T_{half})})$		Logistic Curve Fit Constants with 95% Confidence Intervals		
		T_{half}	Q_{inf}	α
CaCO ₃ Particle Size	15-45 nm	69.4	1.16	0.025
	3-5 μ m	50.9	1.06	0.040
	30 μ m	82.8	1.09	0.027
Anode Diameter	0.25 mm	119.5	0.89	0.010
	0.5 mm	50.4	0.91	0.035
	1.0 mm	50.9	1.06	0.040
Applied Voltage	2.7 V	161.0	0.63	0.018
	5.4 V	50.9	1.06	0.040
	10.8 V	58.9	1.19	0.036
Alginate Concentration	0.5%	129.2	1.43	0.019
	1.0%	50.9	1.06	0.040
	2.0%	54.0	0.89	0.034
CaCO ₃ Weight Percentage	0.01%	66.1	0.91	0.031
	0.1%	58.6	0.87	0.039
	0.5%	50.7	1.06	0.040
	1.0%	84.3	1.01	0.025
BaCO ₃ Weight Percentage	0.10%	119.2	1.23	0.020
	0.50%	80.0	1.03	0.028
	1%	138.9	1.26	0.012

Videos (S1-4). OCT 3D rendering and sectioning of an electrodeposited alginate tube (1), bifurcation (2), T-junction (3), and linear tube with bifurcating ends (4). Uploaded as supporting files.

Supplemental References

- [1] D. Huang, E. Swanson, C. Lin, J. Schuman, W. Stinson, W. Cahng, M. Hee, T. Flotte, K. Gregory, C. Puliafito, Optical Coherence Tomography, *Science*. 254 (1991) 1178–1181.
- [2] J.M. Schmitt, Optical Coherence Tomography (OCT): A Review, *IEEE J. Sel. Top. Quantum Electron.* 5 (1999) 1205–1215.
- [3] D.M. Kingsley, C.L. Roberge, A. Rudkouskaya, D.E. Faulkner, M. Barroso, X. Intes, D.T. Corr, *Acta Biomaterialia* Laser-based 3D bioprinting for spatial and size control of tumor spheroids and embryoid bodies, *Acta Biomater.* (2019). doi:10.1016/j.actbio.2019.02.014.
- [4] J. Conder, fit_logistic(t,Q), (2016). https://www.mathworks.com/matlabcentral/fileexchange/41781-fit_logistic-t-q.

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