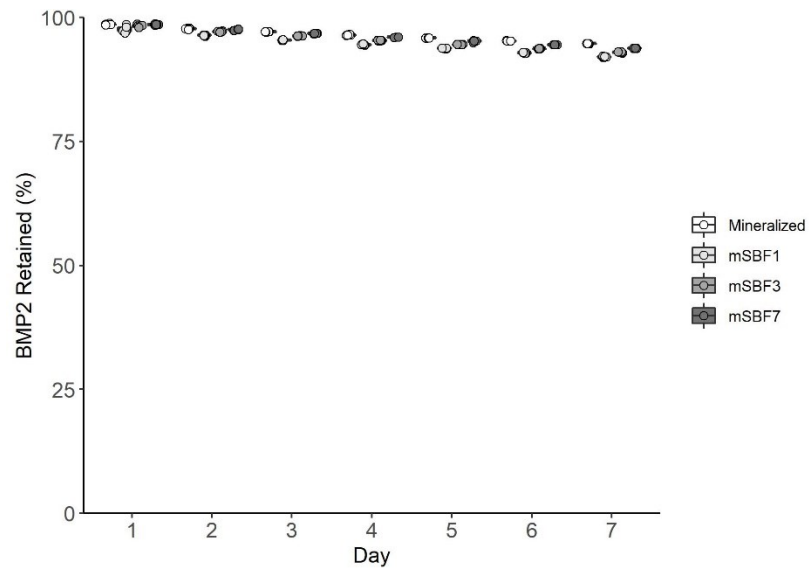


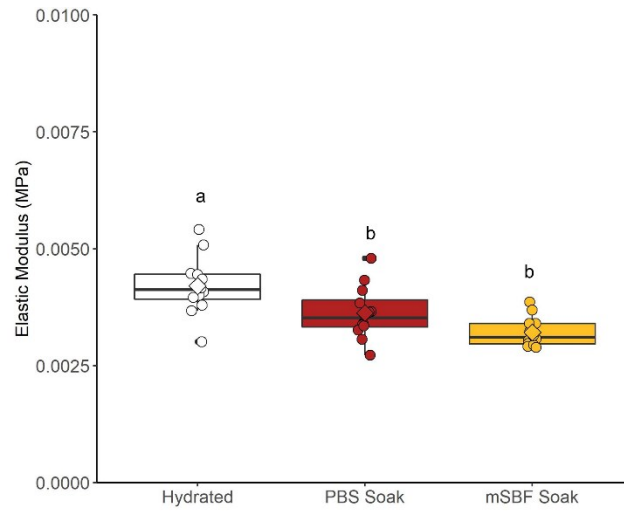
## Supporting Information

### **Sequential sequestrations increase the incorporation and retention of multiple growth factors in mineralized collagen scaffolds**

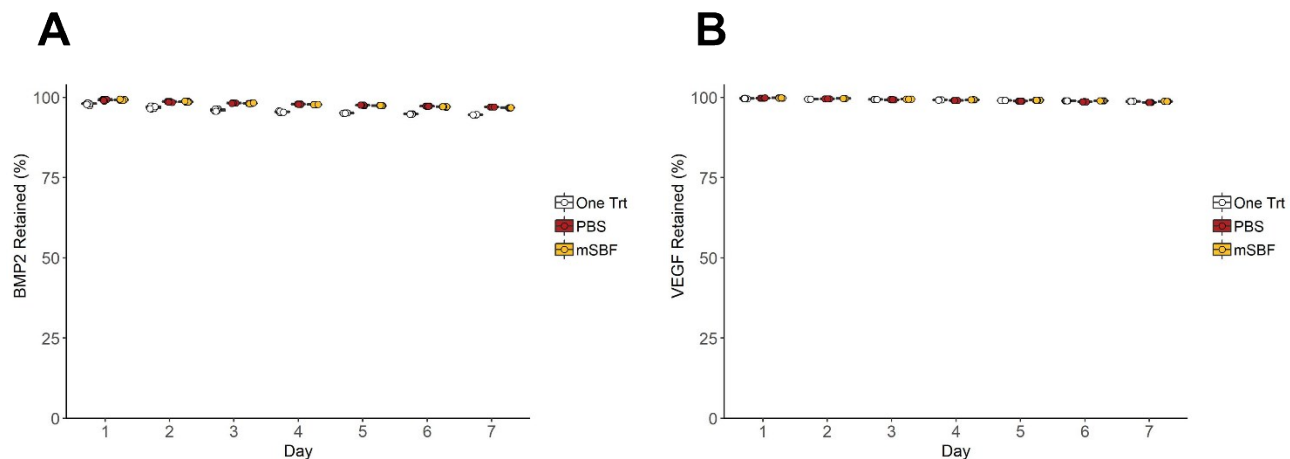
*Aleczandria S. Tiffany, Marley J. Dewey, Brendan A.C. Harley*



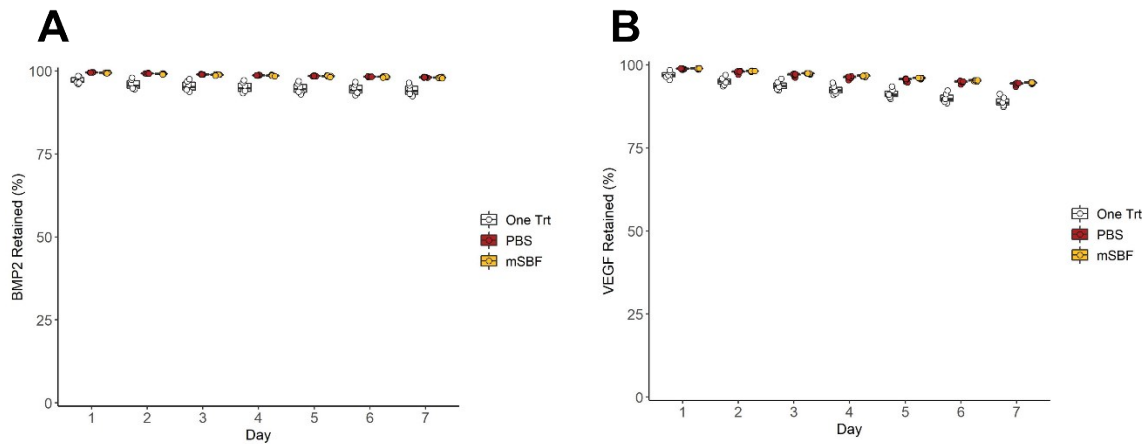
**Supplemental Figure 1: Full BMP2 retention plot for data presented in Figure 1C.** Boxplots overlaid with individual data points are used to represent data. Retention of BMP2 in scaffolds with the y-axis starting at 0%.



**Supplemental Figure 2: Compression testing of hydrated, mSBF soaked, and PBS soaked scaffolds.** Boxplots overlaid with individual data points are used to represent data. Elastic modulus of treated scaffolds. All scaffolds groups are softer than 5 kPa. Groups that share a letter are not significantly different ( $p < 0.05$ ).



**Supplemental Figure 3: Full retention plots for data presented in Figure 2.** Boxplots overlaid with individual data points are used to represent data. (A) Retention of BMP2 in scaffolds with the y-axis starting at 0%; corresponding to data presented in Figure 2C. (B) Retention of VEGF in scaffolds with the y-axis starting at 0%; corresponding to data presented in Figure 2E.



**Supplemental Figure 4: Full retention plots for data presented in Figure 3.** Boxplots overlaid with individual data points are used to represent data. (A) Retention of BMP2 in scaffolds with the y-axis starting at 0%; corresponding to data presented in Figure 3C. (B) Retention of VEGF in scaffolds with the y-axis starting at 0%; corresponding to data presented in Figure 3E.

**Supplemental Table 1.** PCR primers and assay IDs.

<b>Transcript</b>	<b>Supplier</b>	<b>Assay ID</b>
<i>18S</i>	ThermoFisher (Taqman)	Hs99999901_s1
<i>PPIA</i>	ThermoFisher (Taqman)	Hs04194521_s1
<i>PDGF</i>	ThermoFisher (Taqman)	Hs00966522_m1
<i>HIF1a</i>	ThermoFisher (Taqman)	Hs00153153_m1
<i>ANG1</i>	ThermoFisher (Taqman)	Hs00919202_m1
<i>ANG2</i>	ThermoFisher (Taqman)	Hs00169867_m1

**Supplemental Table 2.** Summary statistics for PDGF and HIF1a PCR data

Day	Group	Platelet Derived Growth Factor		Hypoxia Inducible Factor 1 $\alpha$	
		Fold Change*	Sample Size	Fold Change*	Sample Size
1	Blank	1.310 $\pm$ 2.086	7	3.565 $\pm$ 4.796	7
1	mSBF	1.246 $\pm$ 0.902	8	1.902 $\pm$ 0.696	8
1	One Trt	2.191 $\pm$ 0.921	7	2.208 $\pm$ 0.845	8
1	Soluble	0.440 $\pm$ 0.199	8	1.363 $\pm$ 0.417	8
4	Blank	2.461 $\pm$ 1.381	7	3.645 $\pm$ 3.468	8
4	mSBF	3.592 $\pm$ 2.005	7	1.248 $\pm$ 0.379	8
4	One Trt	18.95 $\pm$ 28.48	4	10.76 $\pm$ 9.932	8
4	Soluble	4.430 $\pm$ 3.353	8	2.226 $\pm$ 1.506	8
7	Blank	2.123 $\pm$ 2.003	8	3.014 $\pm$ 5.937	8
7	mSBF	1.624 $\pm$ 0.654	6	0.647 $\pm$ 0.126	8
7	One Trt	3.619 $\pm$ 1.184	5	1.916 $\pm$ 1.561	8
7	Soluble	2.514 $\pm$ 1.543	6	1.249 $\pm$ 1.669	8

\*mean  $\pm$  standard deviation.

**Supplemental Table 3.** Summary statistics for ANG1 and ANG2 PCR data

Day	Group	Angiopoietin 1		Angiopoietin 2	
		Fold Change*	Sample Size	Fold Change*	Sample Size
1	Blank	5.360 ± 5.702	7	1.104 ± 1.462	7
1	mSBF	4.176 ± 1.991	8	0.737 ± 0.290	8
1	One Trt	2.182 ± 1.427	4	0.455 ± 0.186	8
1	Soluble	5.309 ± 4.315	8	0.302 ± 0.086	8
4	Blank	2.508 ± 2.410	7	2.742 ± 2.994	8
4	mSBF	2.030 ± 0.550	7	3.865 ± 7.248	8
4	One Trt	4.324 ± 3.557	3	15.12 ± 12.86	8
4	Soluble	2.126 ± 1.365	7	1.251 ± 0.656	8
7	Blank	1.035 ± 0.335	6	6.024 ± 12.18	8
7	mSBF	1.495 ± 0.447	8	0.936 ± 0.286	8
7	One Trt	2.335 ± 1.438	4	4.058 ± 6.275	8
7	Soluble	1.718 ± 1.195	6	1.666 ± 1.115	7

\*mean ± standard deviation.