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.

| Transcript | Supplier          | Assay ID          |
|------------|-------------------|-------------------|
| 18S        | ThermoFisher (Taqman) | Hs99999901_s1   |
| PPIA       | ThermoFisher (Taqman) | Hs04194521_s1   |
| PDGF       | ThermoFisher (Taqman) | Hs00966522_m1   |
| HIF1a      | ThermoFisher (Taqman) | Hs00153153_m1   |
| ANG1       | ThermoFisher (Taqman) | Hs00919202_m1   |
| ANG2       | ThermoFisher (Taqman) | Hs00169867_m1   |
Supplemental Table 2. Summary statistics for PDGF and HIF1α PCR data

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

*mean ± 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.