Supporting Information

Injectable MnSr-doped brushite bone cements with improved biological performance

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Table S1. Evaluation of the homogeneity of the CPC pastes [initial liquid to powder ratio (LPR) = 0.28 mL·g^{-1}] upon extrusion. To evaluate filter pressing effects, paste samples were collected every 30 ± 2 s counted from the beginning of the extrusion process. Any remaining paste in the syringe (when complete extrusion did not occur) was also collected. These samples were weighed before and after complete drying at 120ºC for 24 h, to determine their LPR values. The filter pressing effect was evaluated by the significance of the variations in LPR (three independent experiments) undergone upon the extrusion of each paste and quantified from repeated measures. ANOVA with Greenhouse Geisser correction was used to determine statistical significance ($p < 0.05$).

Cement Pastes	LPR of A1 [a] (± 0.01) (mL g^{-1})	LPR of A2 [a] (± 0.01) (mL g^{-1})	LPR of A3 [a] (± 0.01) (mL g^{-1})	LPR of remaining paste not extruded (± 0.01) (mL g^{-1})	p-value
Sr5CPC–A	0.3	-	-	0.26	0.029[b]
Sr5CPC–AS	0.27	0.28	0.29	-	0.156
Sr5CPC–AF	0.27	0.29	0.28	-	0.103
Mn0.5Sr5CPC–A	0.28	0.3	-	0.25	0.011[b]
Mn0.5Sr5CPC–AS	0.27	0.28	0.28	-	0.055
Mn0.5Sr5CPC–AF	0.28	0.29	0.29	-	0.100
Mn1Sr5CPC–A	0.28	0.3	-	0.26	0.029[b]
Mn1Sr5CPC–AS	0.28	0.28	0.29	-	0.065
Mn1Sr5CPC–AF	0.27	0.29	0.29	-	0.057

[a] A1, A2 and A3 – samples of paste injected during the extrusion.

[b] Presence of filter pressing effect ($p < 0.05$).

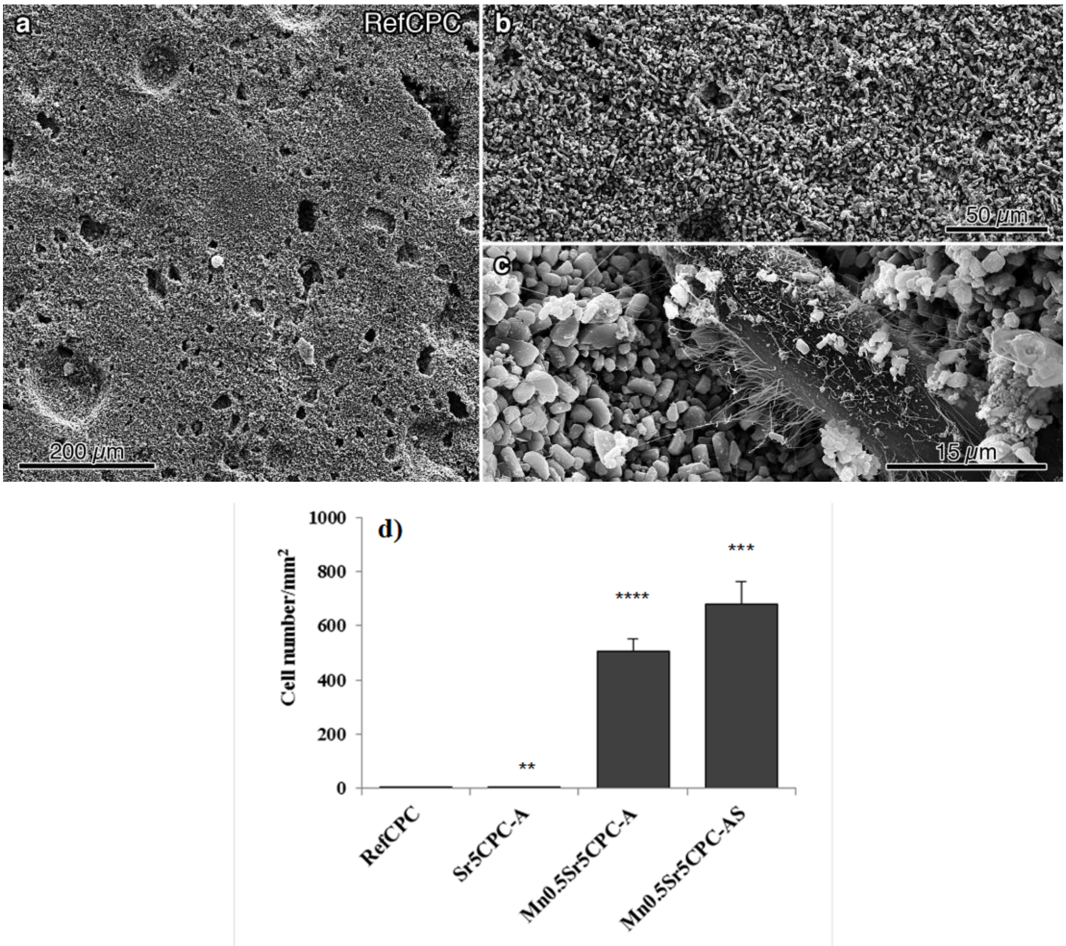

Figure S1. SEM images of MG63 human osteoblasts on disc surface of RefCPC after 7 days of culturing (**a**-**c**). The area shown in **a**) is 0.5 mm^2. **a**)-**b**) Surface texture at two magnifications. No cells are visible in the area displayed. **c**) Detail of one of the rare cells found on the disk surface, showing numerous thin pseudopodia extending toward surface particles from a relatively thick, elongated cell body (compare with Figure 7). **d)** Plot of cell densities obtained under each of the four experimental conditions, estimated from cell counts over at least 3 mm^2 of disk surface visualized as in **a)**. Symbol (*) denotes significantly different values for co-doped CPCs-A and –AS with respect to RefCPC-A; **, $p<0.01$; *****, $p<0.001$; ****, $p<0.0001$). Statistic significances in-between doped CPCs: Sr5CPC-A vs Mn0.5Sr5CPC-A $p < 0.0001$; Sr5CPC-A vs Mn0.5Sr5CPC-AS $p < 0.0002$; Mn0.5Sr5CPC-A vs Mn0.5Sr5CPC-AS $p = 0.0518$.

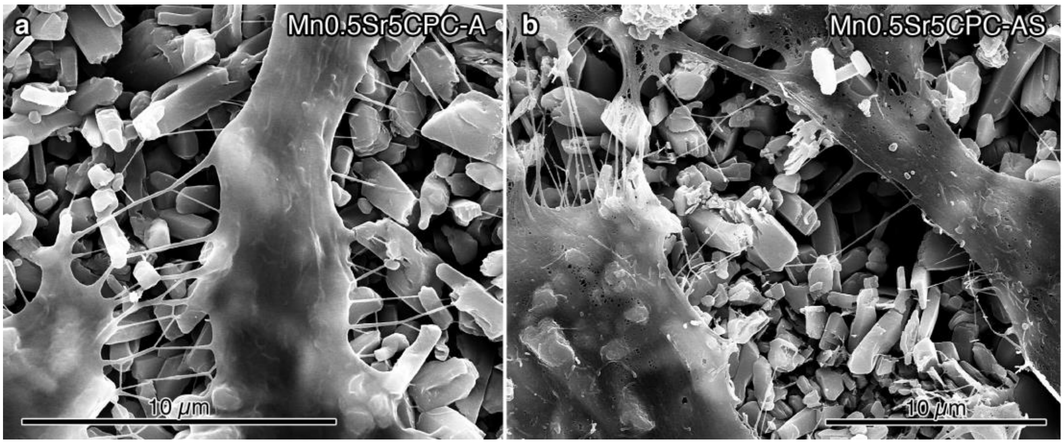

Figure S2. Details of MG63 cells colonizing the disc surface of Mn0.5Sr5CPC-A **(a)** and Mn0.5Sr5CPC-AS **(b)** using SEM. Both cell-to-cell and cell-to-substrate connections by thin pseudopodia are abundant. The cells are rather flat and somewhat electron-translucent, allowing the distinction of underlying substrate particles.

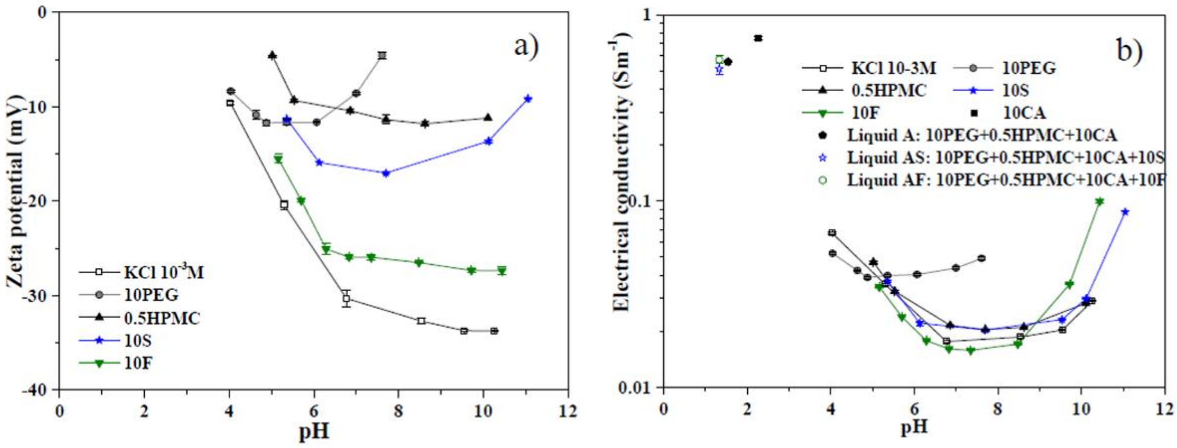

Figure S3. Since Sr5CPC cements presented the most significant differences of injectability percentage when sugars were added to the setting liquids, the Sr5-TCP powder was selected to investigate the effects of liquids' composition on their dispersing ability. The influences of different aqueous electrolyte solutions and pH on **a)** Zeta Potential (ZP) and **b)** Electrical conductivity (EC) were therefore surveyed. Except for molar concentration in KCl (M), the other numerical coefficients in the sample codes stand for weight percent concentration (wt.%) in the electrolyte in solution. The error bars of standard deviations are almost imperceptible in some cases, meaning excellent reproducibility of data. The measurements were carried out at several pH values using a Zeta Sizer (Nano ZS, Malvern, Worcestershire, UK). For this, 13 mg of powder was ultrasonically dispersed for 15 min into 50 mL of the background electrolyte solution (1 mM KCl) and adding the other additives. Each dilute suspension was divided into two equal parts, one for increasing and the other for decreasing pH runs, and the pH values were adjusted using 0.1 M NaOH or 0.1 M HCl solutions, respectively. Results revealed that ZP is significantly affected by pH and the type of additive, while EC is mostly dependent on pH (except in the presence of PEG). The results for 15 wt.% CA in **a)** are not shown because fast dissolution of particles hindered any reliable measurement, in agreement with observations reported elsewhere [36]. The ZP curves for the Sr5-TCP powders suspended in 0.5 wt.% HPMC, 10 wt.% of fructose and KCl 10^{-3} M as electrolytes, showed a decreasing trend as pH increases, more pronounced for KCl 10^{-3} M. ZP curves in the presence of 10 wt.% PEG or 10 wt.% sucrose present minima of ~12 mV at pH range 5−6 and of ~17 mV at pH≈8, respectively. In the presence of fructose, higher absolute ZP values were registered in comparison to sucrose. High EC values **b)** were measured when particles are dispersed in the complete setting liquids, irrespective of their additive contents. When the same additives were dissolved in the KCl 10^{-3} M used for electrophoretic measurements, EC decreased for about 1.5 order of magnitude.