

Supplementary Information

For

Biomimetic cues from poly(lactic-co-glycolic acid)/hydroxyapatite nano-fibrous scaffolds drive osteogenic commitment in human mesenchymal stem cells in the absence of osteogenic factor supplements

Mohammad Soheilmoghaddam^a, Harish Padmanabhan^a and Justin J. Cooper-White^{*,a,b,c}

^{a.} *Tissue Engineering and Microfluidics Laboratory (TE&M), Australian Institute for Bioengineering and Nanotechnology (AIBN), University Of Queensland, St Lucia, QLD, Australia*

^{b.} *The UQ Centre in Stem Cell Ageing and Regenerative Engineering (StemCARE), Australian Institute for Bioengineering and Nanotechnology, University Of Queensland, St Lucia, QLD, Australia*

^{c.} *School of Chemical Engineering, University Of Queensland, St Lucia, QLD, Australia*

* To whom correspondence should be addressed email: j.cooperwhite@uq.edu.au

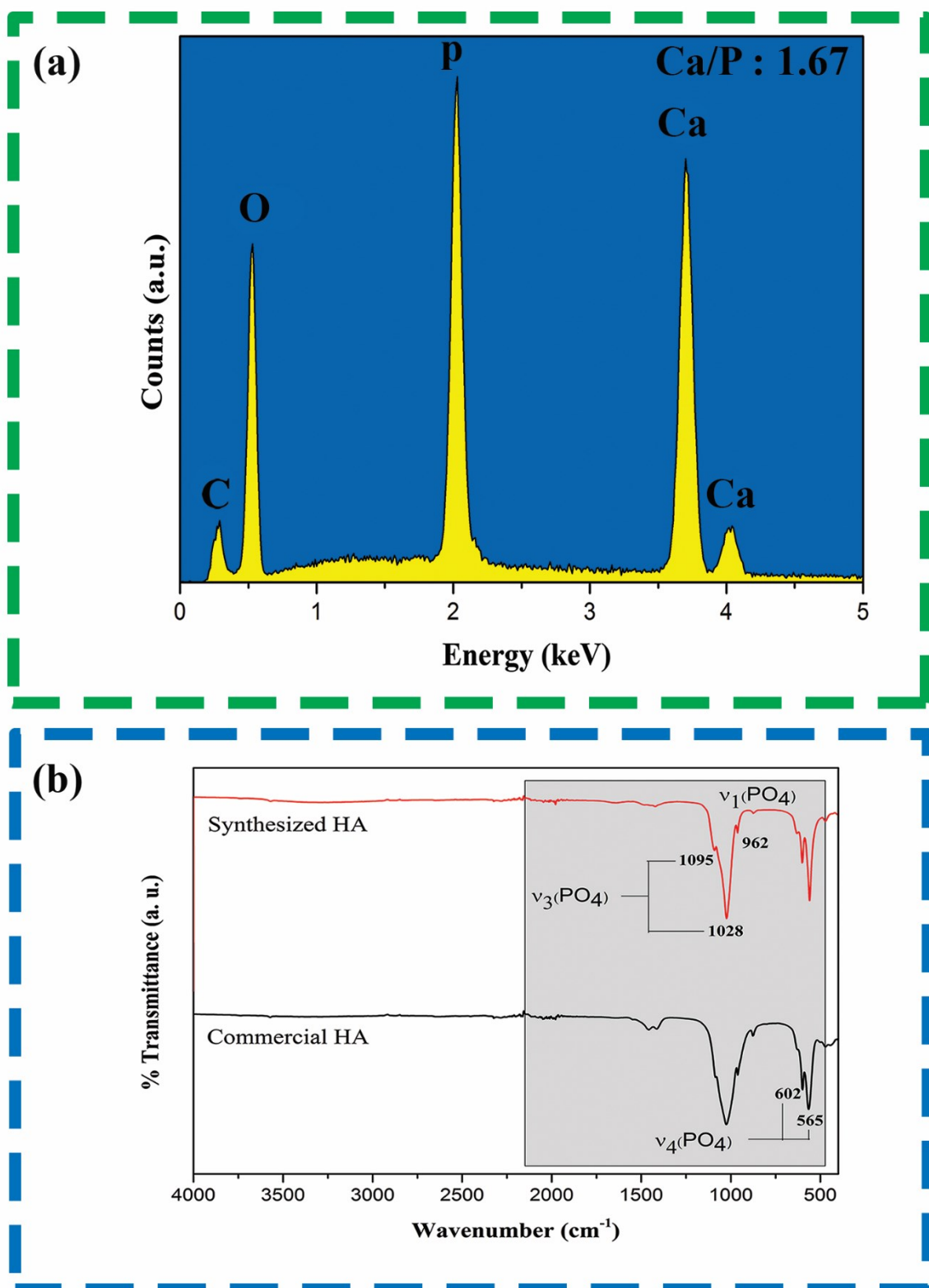


Fig. S1 (a) EDX spectra of synthesised nHA with the Ca/P ratio of 1.67 and (b) FTIR spectra obtained for synthesised nHA, compared with commercial nHA.

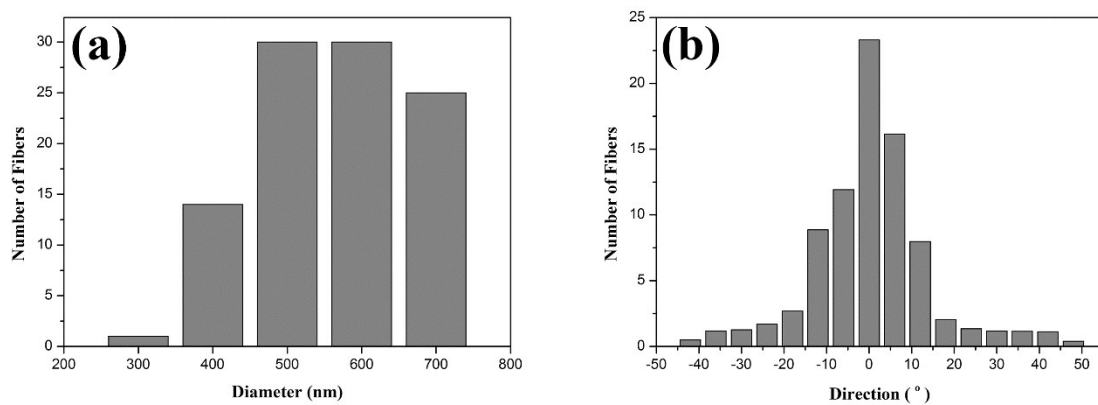


Fig. S2 (a) Size distribution of the fabricated electrospun PLGA mat and (b) the fiber alignment to the reference direction.

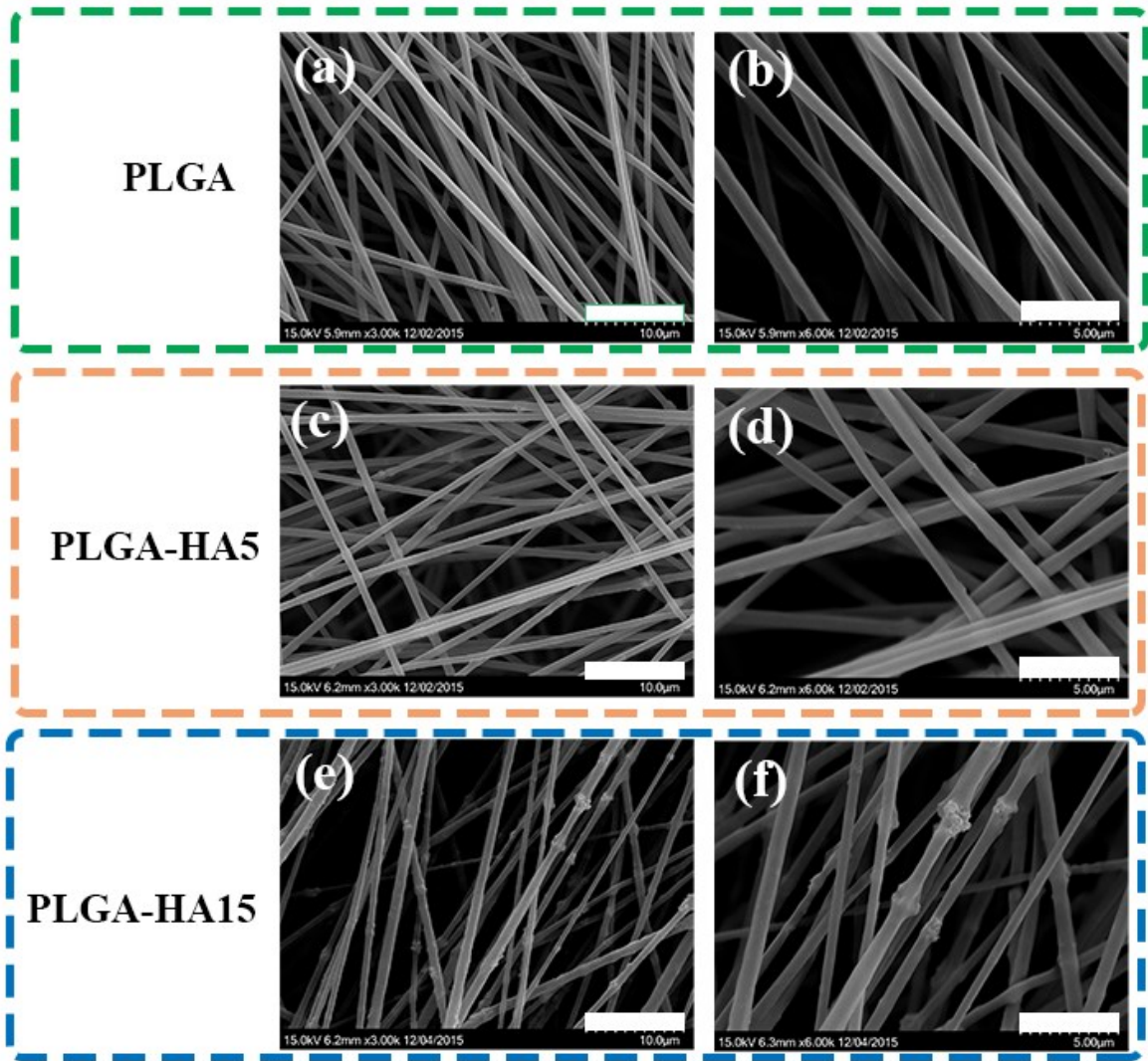


Fig. S3 SEM pictures of (a and b) PLGA and (c and d) PLGA-HA5 nanofibrous scaffold. PLGA showed continuous and smooth fibers while (e and f) HA agglomeration occurred along the PLGA- HA15 fibers.

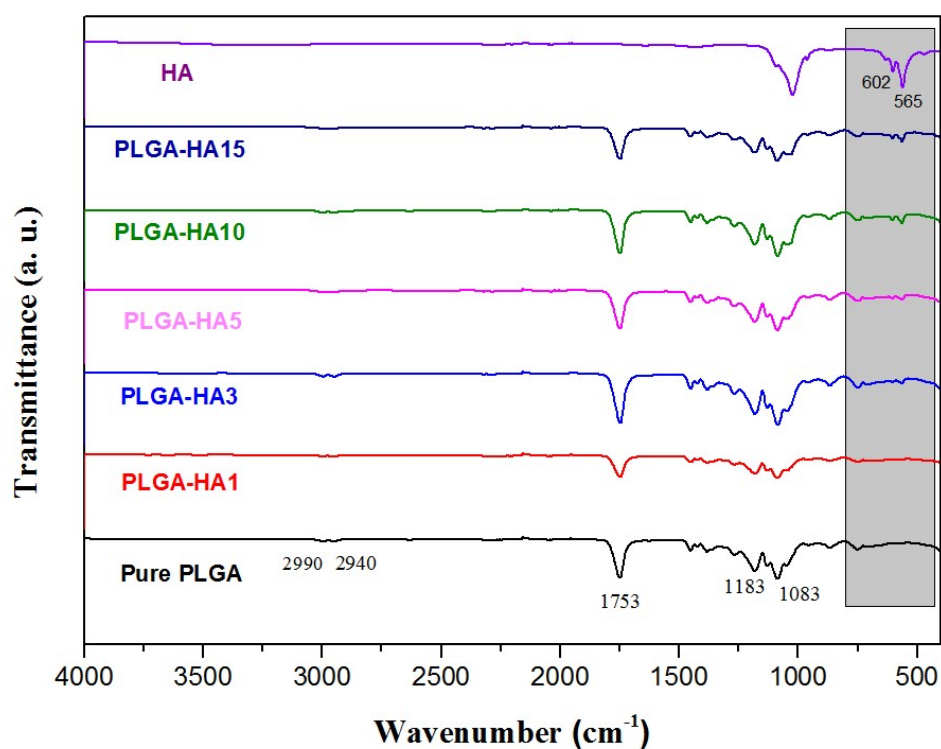


Fig. S4 FTIR spectra of nHA, PLGA and PLGA nanocomposites modified with different amounts of nHA. The PLGA-HA electrospun fibers showed new bands that attributed to the nHA.

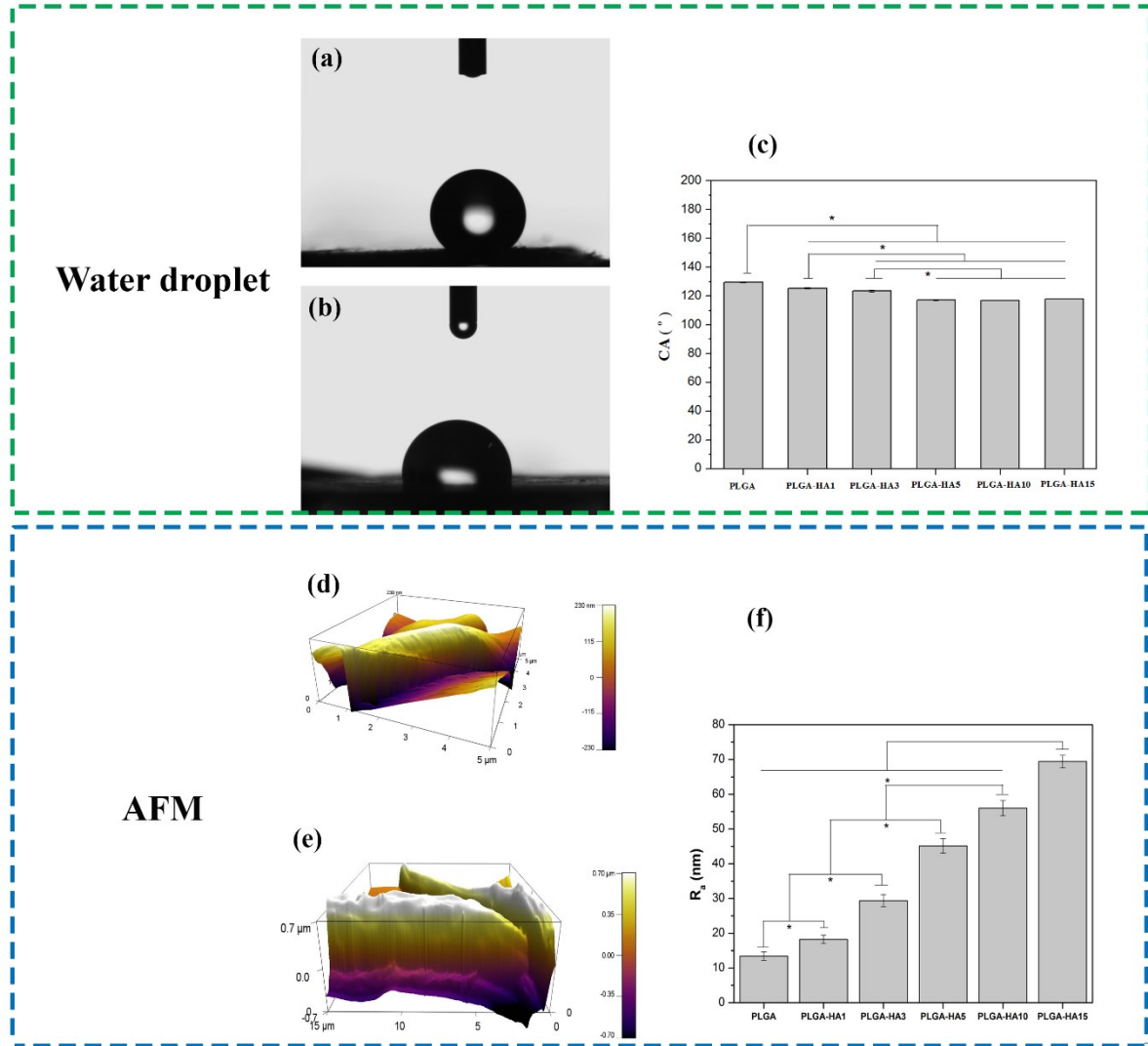


Fig. S5 Water droplet images of (a) PLGA and (b) PLGA-HA10. (c) Water contact angle results showed the higher hydrophilicity of the PLGA-HA fibers compared to PLGA. AFM topography images of (d) PLGA nanofibers, (e) PLGA-HA10 nanocomposites and (f) the surface roughness (R_a) of the nanocomposite scaffolds. * indicates a significant difference of $p \leq 0.05$.

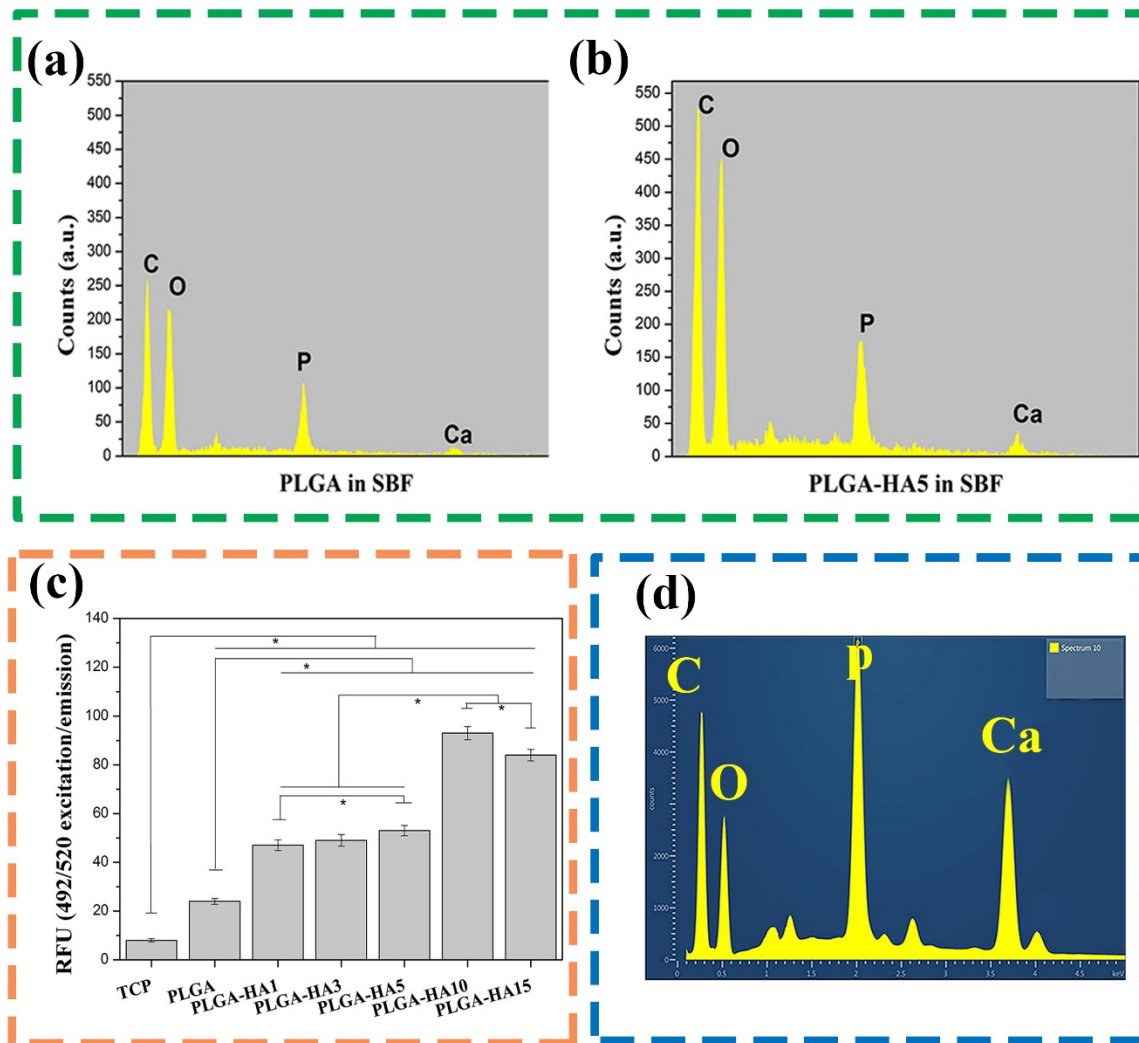


Fig. S6 EDX analysis of the formed bioapatite on the surface of (a) PLGA and (b) PLGA-5HA scaffolds after submerging for 1 day in SBF, indicating a Ca/P ratio of 1.67. (c) Quantitative measurement of HA content using the Osteoimage assay (Lonza, Walkersville, MD). The fluorescent staining reagent (green) binds to the HA portion of the mineralised matrix and the fluorescence is measured at 495/519 nm (Ex/Em); * $P < 0.05$. (d) EDX analysis of the deposited minerals on PLGA -HA5 after 21 days.

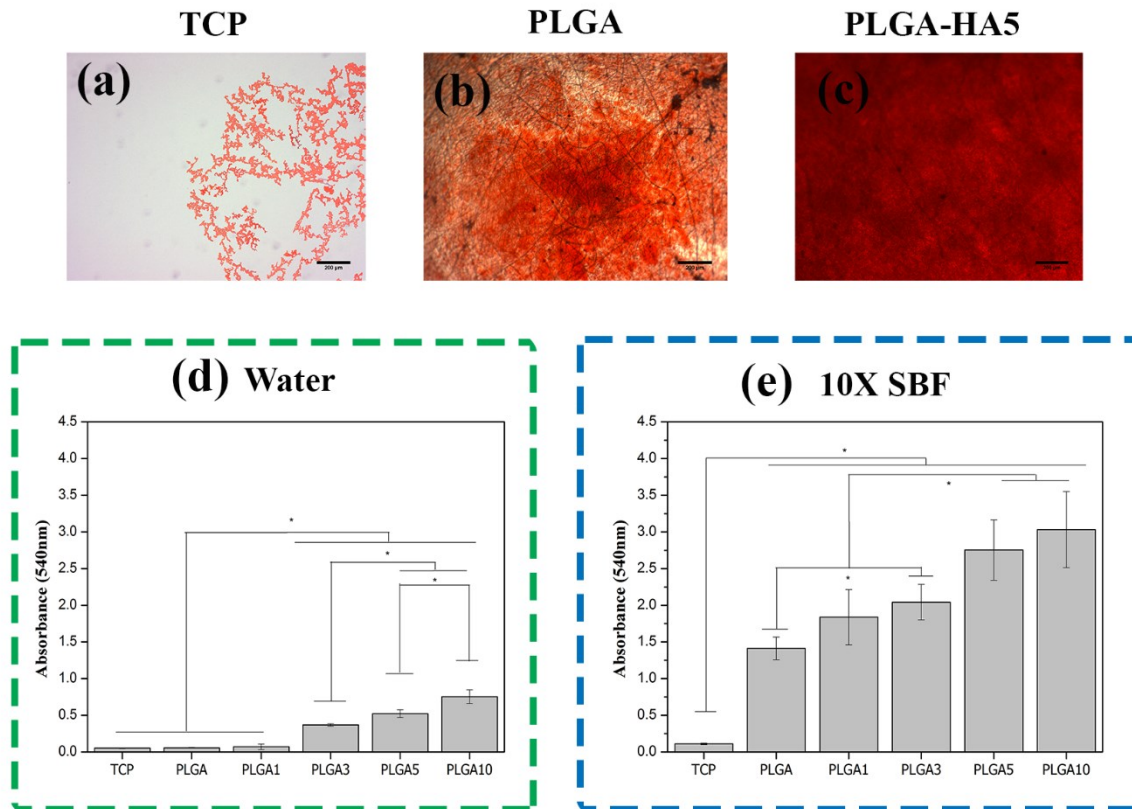


Fig. S7 Alizarin Red staining of calcium deposited on (a) TCP, (b) PLGA and (c) PLGA- HA5 scaffolds. Quantitative analysis of the calcium content that formed on scaffolds in (d) water and (e) 10X SBF. Data represents the mean 6 standard deviations, n = 6. Scale bars, 200 μ m; *P<0.05.

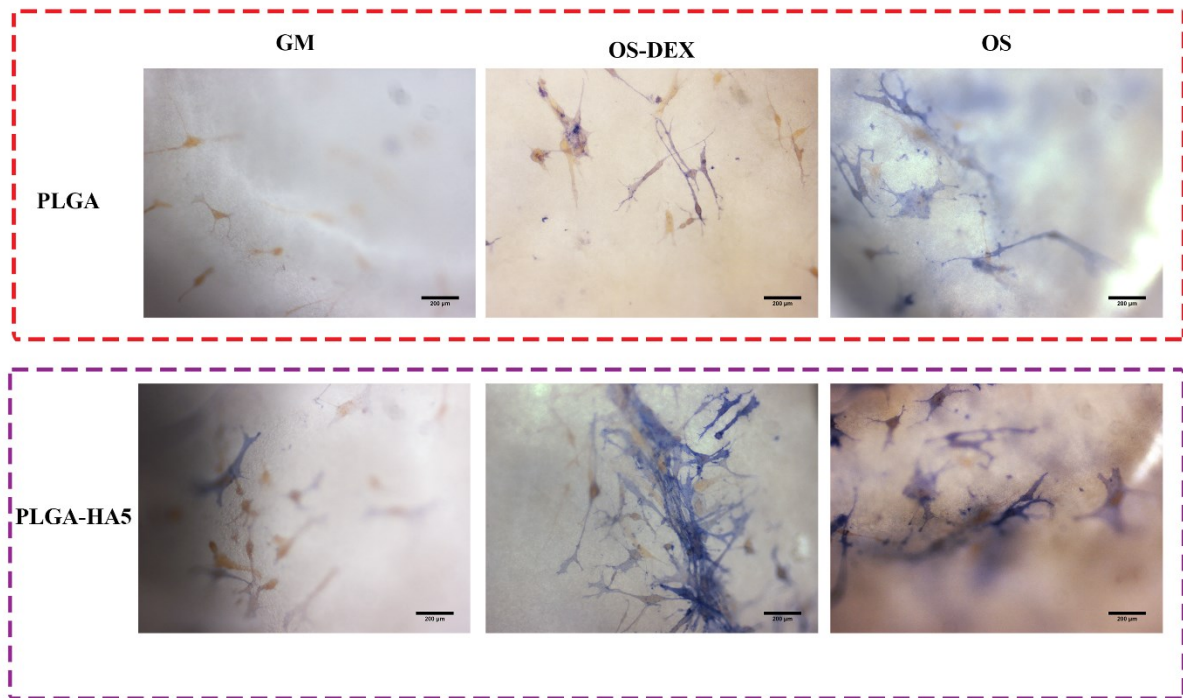


Fig. S8 ALP staining of leftover hBMSCs on PLGA and PLGA-HA5 scaffolds after harvesting. The scaffolds were seeded in different media conditions including GM, OS-DEX and OS. Scale bars, 200 μ m.

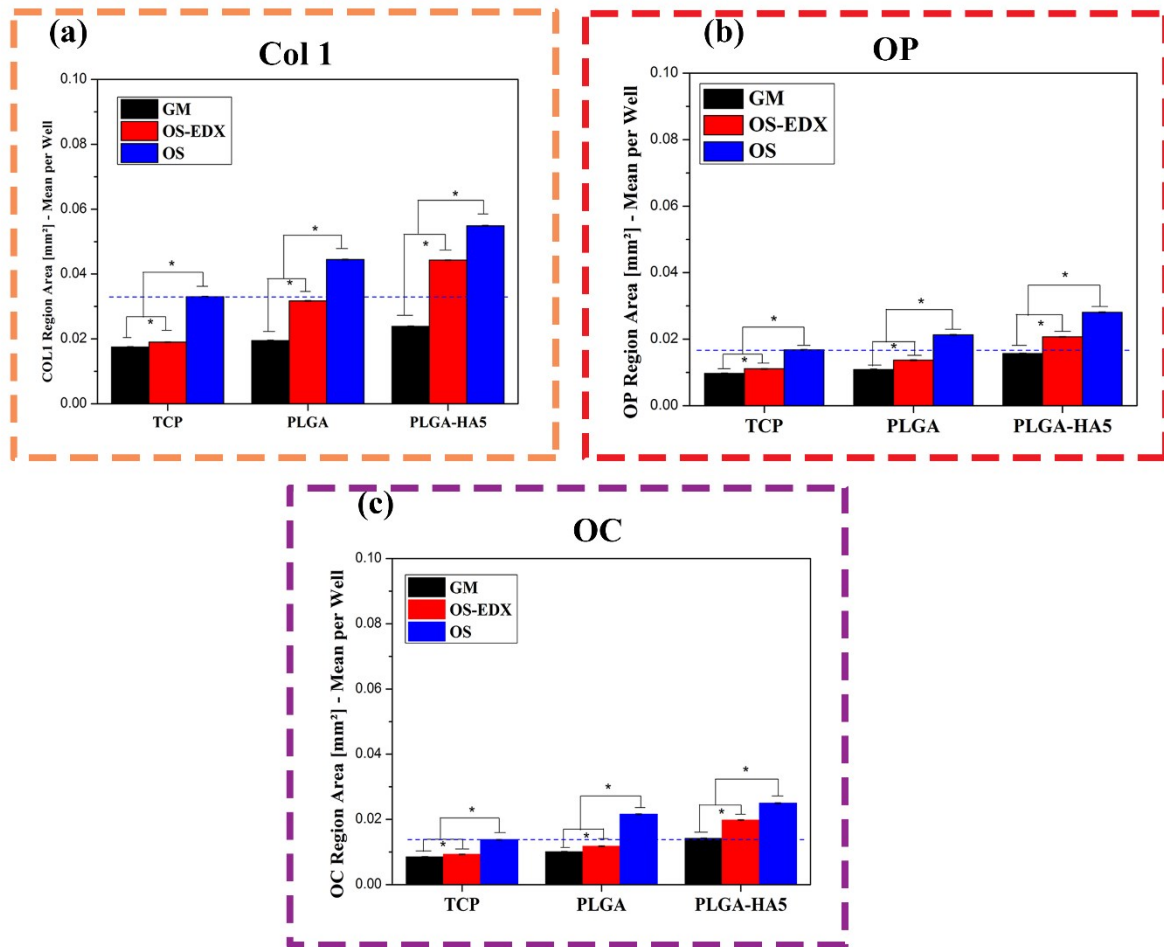


Fig. S9 (a) Col1 was quantified based on the area of positive staining on TCP, PLGA and PLGA-HA5 after 7 days. (b) Quantification of OP protein expressed after 21 days on TCP, PLGA and PLGA-HA scaffolds. (c) Quantification of OC protein expressed after 21 days on TCP, PLGA and PLGA-HA scaffolds. *P<0.05.

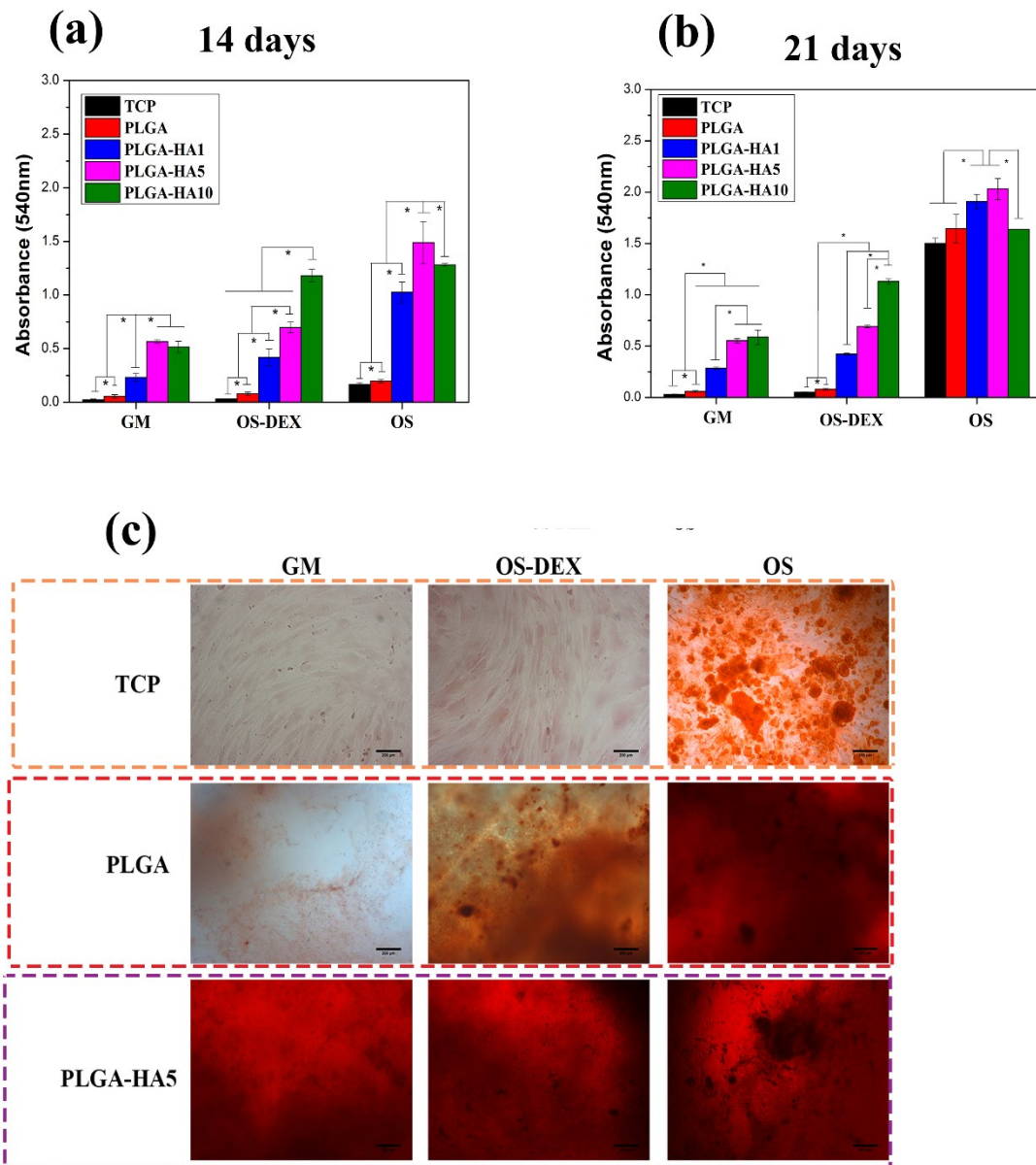


Fig. S10 Quantitative analysis of the mineralisation of hBMSCs on TCP, PLGA and PLGA-HA scaffolds cultured in GM, OS-DEX and OS for (a) 14 days and (b) 21days. (c) Alizarin Red staining of hBMSCs cultured on TCP, PLGA and PLGA-HA5 in different medium conditions after 21 days. Scale bars, 200 μ m; *P<0.05.

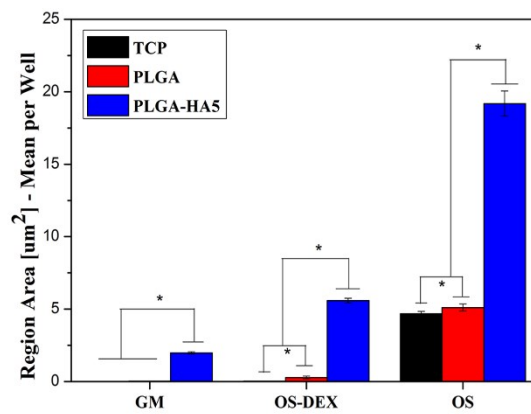
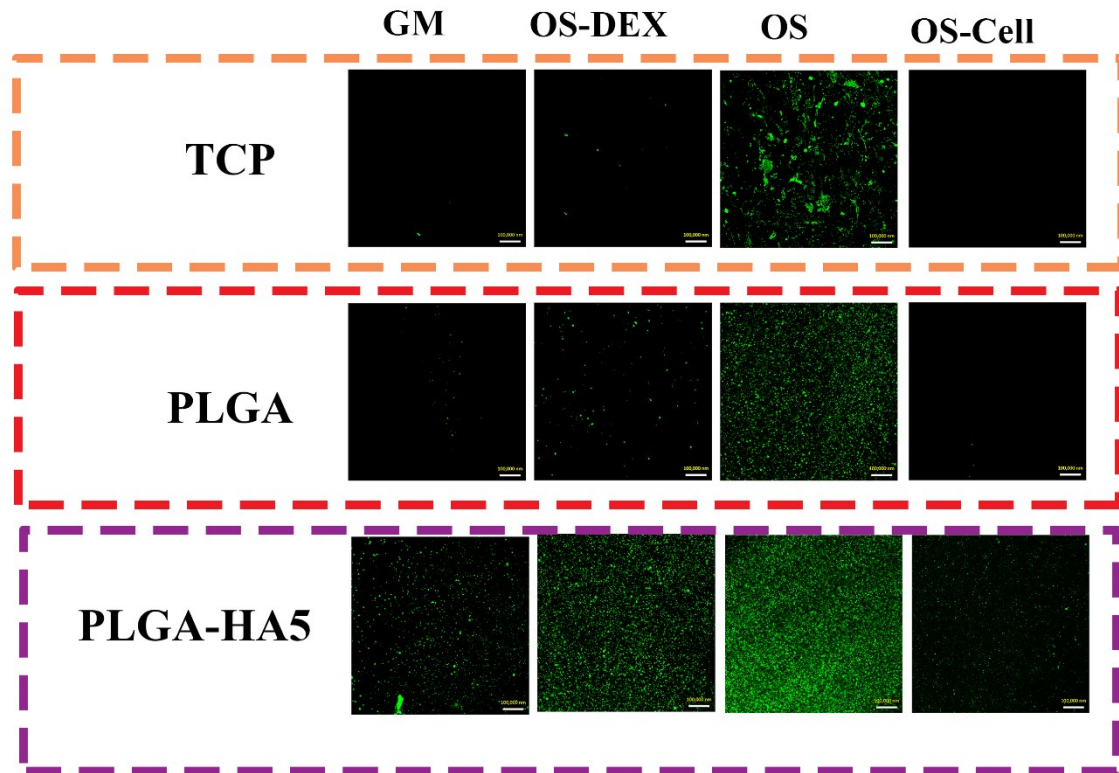


Fig. S11 HA formation on TCP, PLGA and PLGA-HA5 scaffolds cultured in GM, OS- DEX and OS for 21 days. Apatite formation was also quantified. The result has been normalised based on the same samples before cell culture in similar medium conditions and time points. Scale bars, 100 μm .