Electronic Supplementary Material (ESI) for Biomaterials Science.

Gene activated scaffolds incorporating star-shaped polypeptide-pDNA nanomedicines rapidly accelerate bone tissue regeneration in *vivo*

David P. Walsh[1-4], Rosanne M. Raftery[2-4], Robert Murphy[6], Gang Chen[7], Andreas Heise[4-6], Fergal J. O'Brien[1-5], Sally-Ann Cryan [1-3,5]*

[1]Drug Delivery & Advanced Materials Team, School *of Pharmacy & Biomolecular Sciences, RCSI, Dublin, Ireland*

[2]Tissue Engineering Research Group, Department of Anatomy & Regenerative Medicine, RCSI, Dublin, Ireland

[3]Trinity Centre for Biomedical Engineering, Trinity College Dublin (TCD), Dublin, Ireland

[4] SFI Advanced Materials and Bioengineering Research (AMBER) Centre, RCSI & TCD

[5]SFI Centre for Research in Medical Devices (CURAM), RCSI, Dublin and National University of Ireland, Galway, Ireland

[6]Department of Chemistry, RCSI, Dublin, Ireland

[7]Centre for the Study of Neurological Disorders, Microsurgical Research and Training Facility (MRTF), RCSI, Dublin, Ireland

**Corresponding Author:*

Professor Sally-Ann Cryan

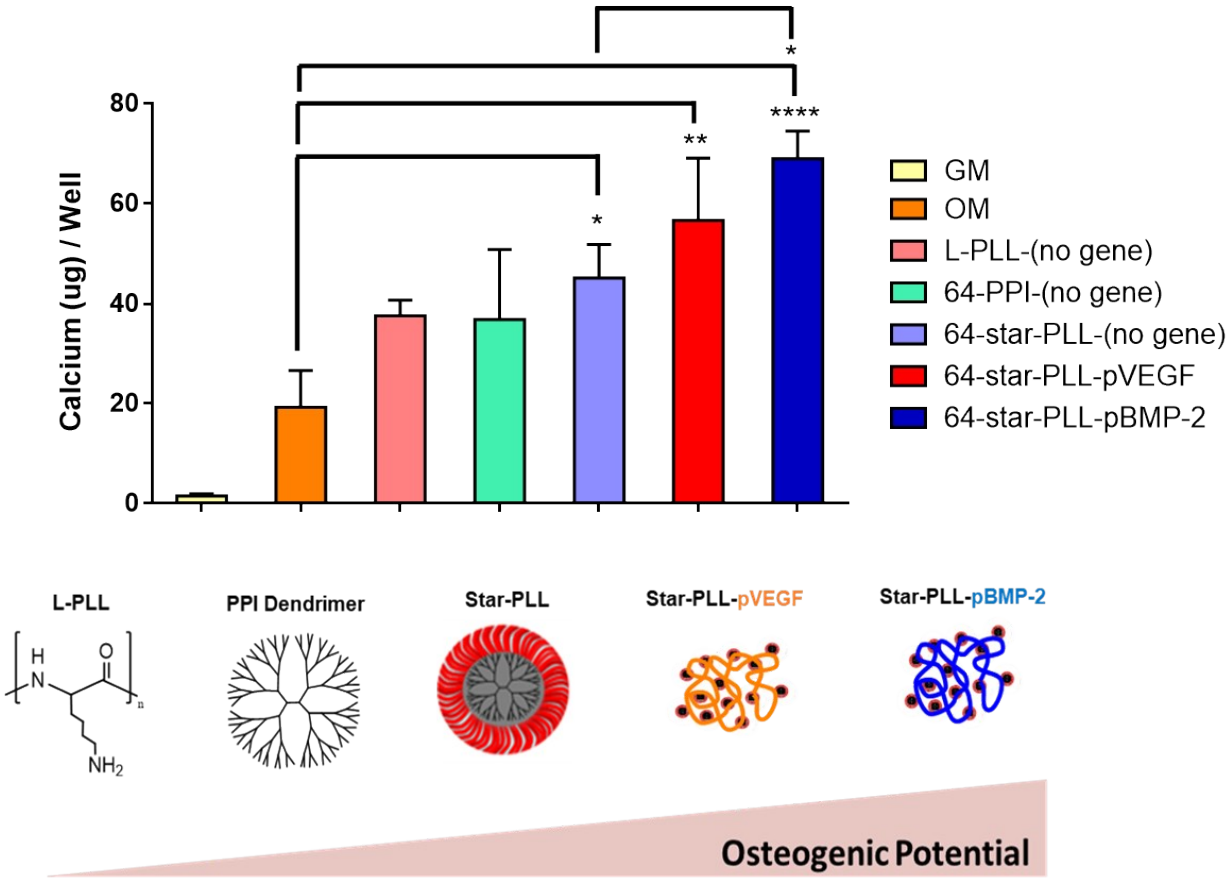
School of Pharmacy,

Royal College of Surgeons in Ireland,

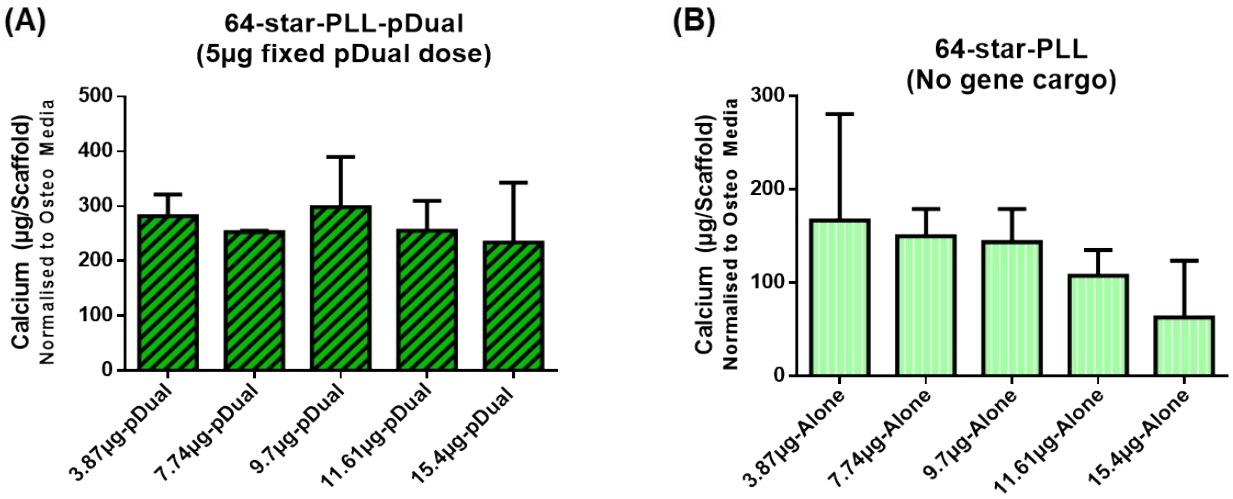
1st Floor, Ardilaun House (Block B), Dublin 2, Ireland

Tel: 00353-(0)1-4022741

Email: scryan@rcsi.ie



Supplementary Information Figure 1: ***Assessment of the osteogenic potential of the star-PLL vector.*** Illustrated above is a systematic study assessing the bioactive potential of the star-PLL architecture or its various components in monolayer MSC culture. The 64-star-PLL vector alone, when unbound to any pDNA cargo can enhance calcium deposition compared to an OM control suggesting a bioactive nature to the star-PLL vector. This osteogenic potential was further enhanced when the vector was complexed with a gene i.e. pVEGF or pBMP-2. L-PLL = Linear poly(L-lysine), 64-PPI = poly(propyleneimine) dendrimer with 64 terminal lysine units. Results are expressed as the mean ± SD (n=3) where *p<0.05, **p<0.01 & ***p<0.001.



Supplementary Information Figure 2: The effect of star-PLL delivered dose on MSC mediated osteogenesis in 3D scaffolds. Illustrated above is calcium deposition by MSCs 28 days post seeding on a series of (A) 64-star-PLL-pDual collagen-HA gene activated scaffolds with an increasing amount of 64-star-PLL vector but constant (5µg) pDual dose or (B) a series of 64-star-PLL-(no gene) collagen-HA activated scaffolds with an increasing 64-star-PLL dose. This data demonstrates that the therapeutic cargo, rather than the star-PLL is the main factor driving MSC osteogenesis. Calcium levels have been normalised to the effects of a gene-free collagen-HA scaffold maintained in osteogenic media for 28 days. Data is expressed as the mean ± SD (n=3).