

2021 Biomaterials Science Emerging Investigators Issue: Enhanced efficiency of nonviral direct neuronal reprogramming on topographical patterns

Supplementary Materials

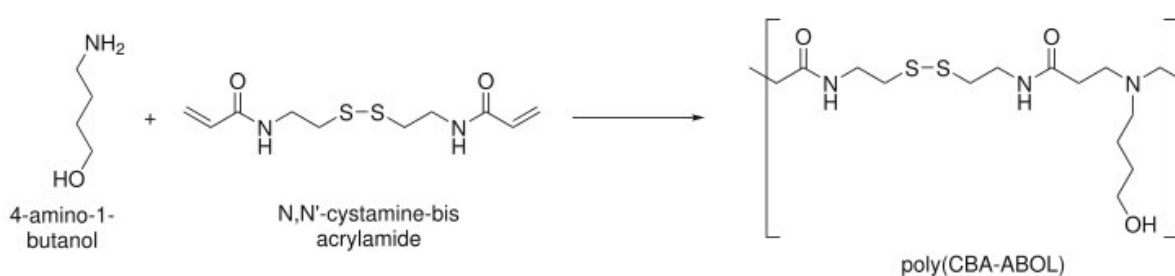


Figure S1 - Michael polyaddition of N,N-cystaminebisacrylamide (CBA) and 4-amino-1-butanol (ABOL) as described by Lin *et al.*¹ The polymeric gene delivery system pABOL is a cationic polymer that can stably envelop plasmids in the extracellular environment and degrades upon entrance to the reductive environment of the cytoplasm. The polyplex formed by pABOL and DNA, is <200 nm in diameter and has a charge of >+20 mV. Readers interested in learning more about pABOL polyplexes and pABOL polyplexes for non-viral neuronal reprogramming are recommended to review the original works of Lin *et al.* and Adler *et al.*^{1, 2}

DAPI TUJ1 MAP2

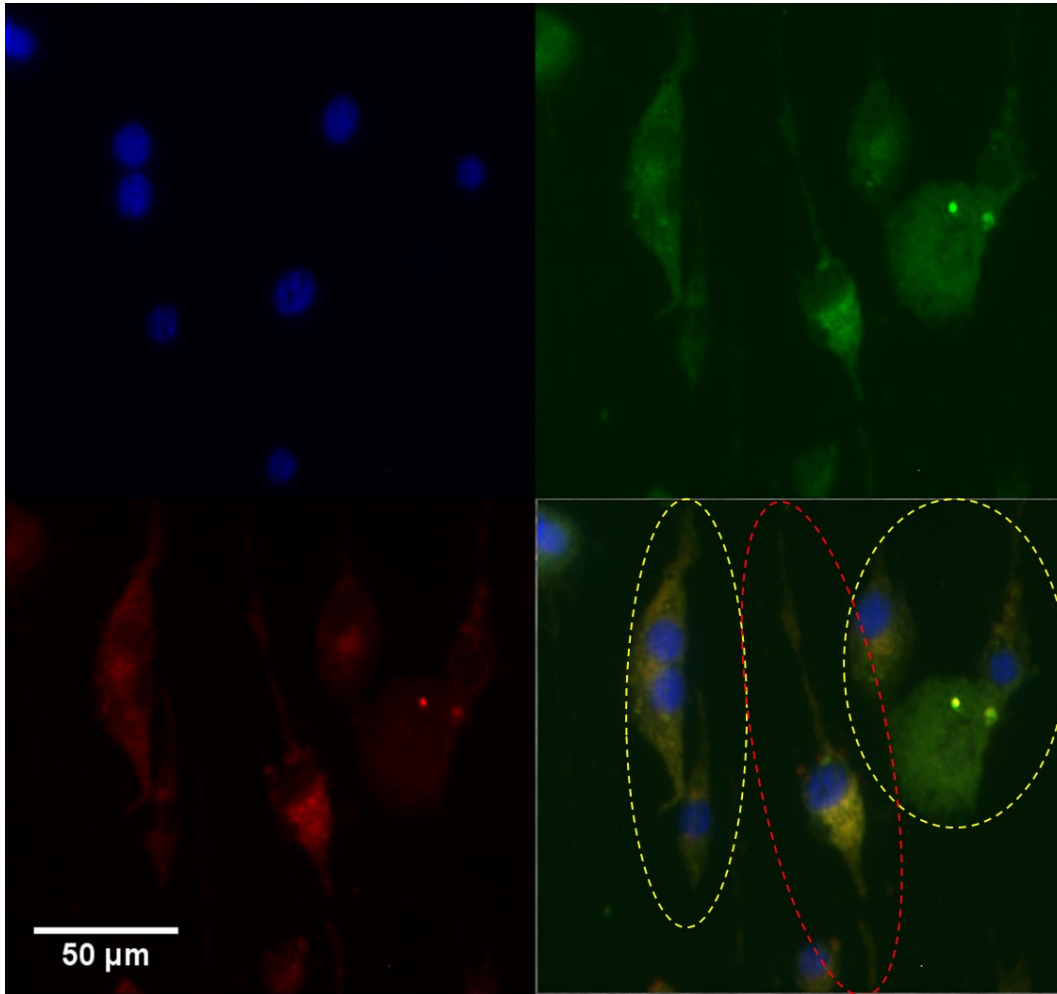


Figure S2 – A representative image showing the morphology of failed reprogramming and successful reprogramming. The dashed-yellow outlines indicate cells that expressed neuronal markers but maintained fibroblastic morphology and thus they were classified as failing to reprogram. The dashed-red outlines indicate a cell that was considered to be successfully reprogrammed. It expresses neuronal markers and has the distinct neuronal morphology of a round soma with protruding extensions.

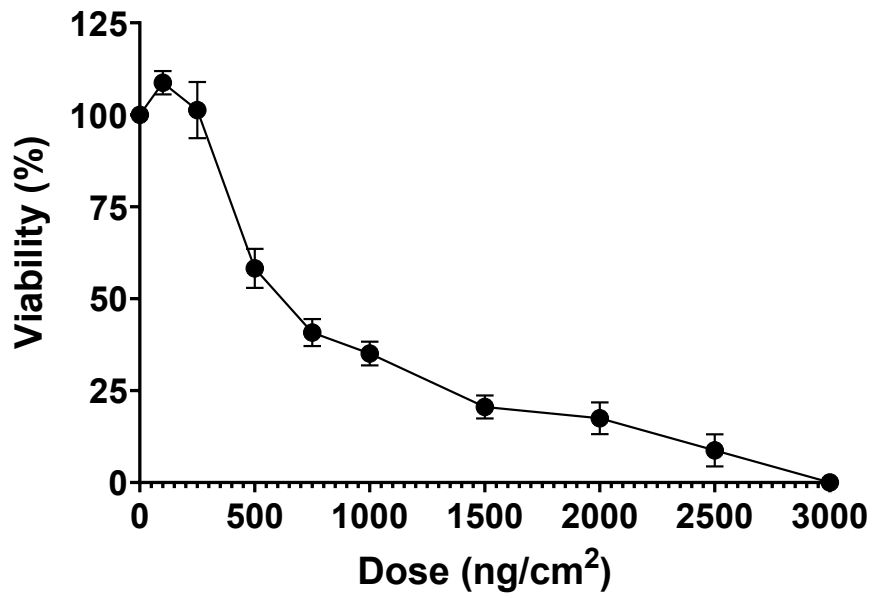


Figure S3 – Viability of primary mouse embryonic fibroblasts transfected with pABOL-pmax-GFP polyplexes at different dosage levels. Data shown as the average of n=6 technical replicates and standard error mean (SEM).

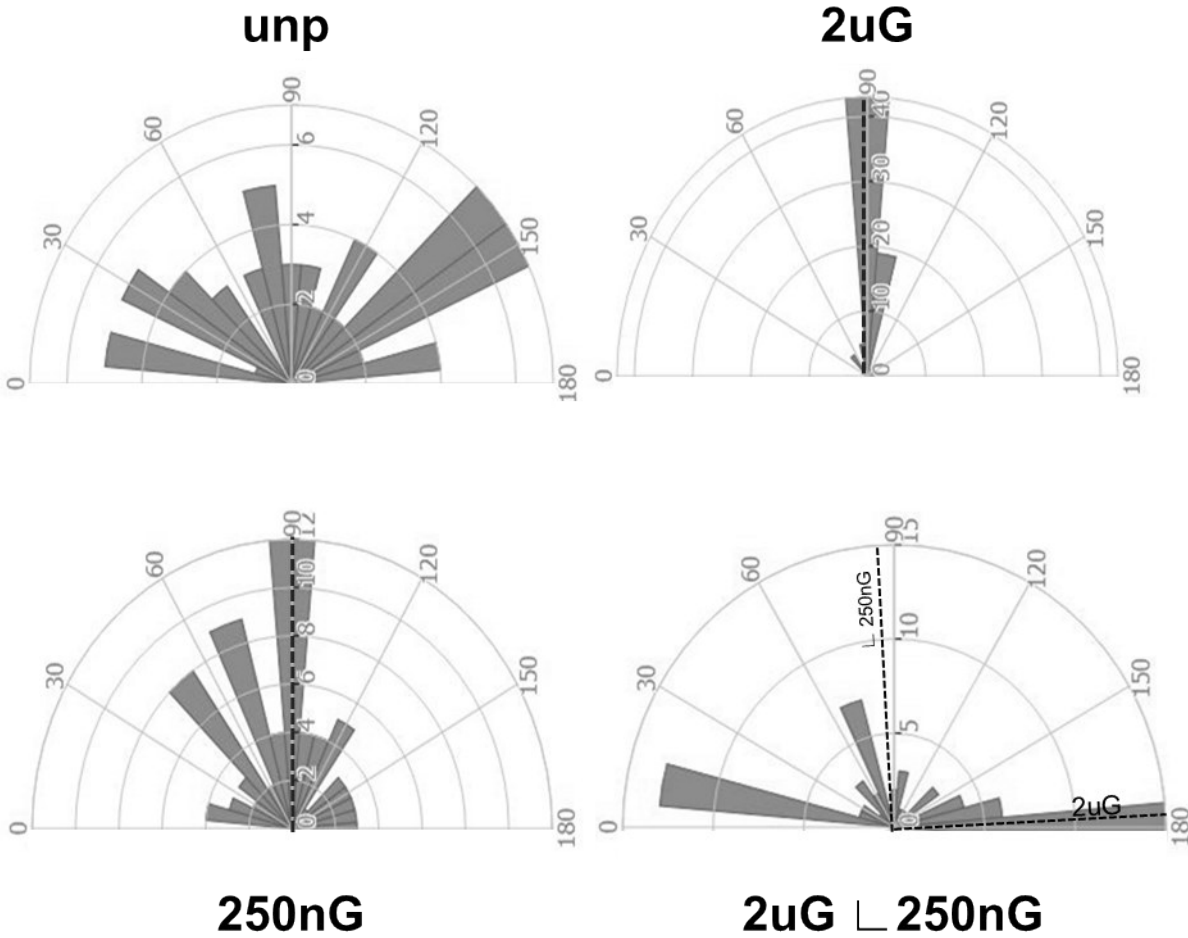


Figure S4 – Cell alignment during single pattern studies on the unpatterned control (unp), 2x2x2 μ m gratings (2 μ G), 250x250x250 nm gratings (250nG), and 2x2x2 μ G with hierarchical with perpendicular 250x250x250 nm gratings (2 μ G \perp 250nG). Dashed lines indicate pattern direction for the 2 μ G and 250nG plot. On the 2 μ G \perp 250nG plot the hierarchical nanoscale pattern, \perp 250nG, and microscale base pattern, 2 μ G, are both marked with respective dashed lines. Data is shown is from N=2 replicates. Approximately 30 - 70 cells were measured for each group. Counting varied depending on the number of cells available.

Reference

1. C. Lin, Z. Zhong, M. C. Lok, X. Jiang, W. E. Hennink, J. Feijen and J. F. J. Engbersen, *Bioconjugate Chemistry*, 2007, **18**, 138-145.
2. A. F. Adler, C. L. Grigsby, K. Kulangara, H. Wang, R. Yasuda and K. W. Leong, *Molecular therapy. Nucleic acids*, 2012, **1**, e32-e32.