Novel 3D Hydrogel System Enhances Regenerative Capacity of Stem Cells and Improves Tailorability of Biologics

Jacob Hodge[1,2] , A.J. Mellott[2] , Aisha Amari[3] and Alex Sim[3]

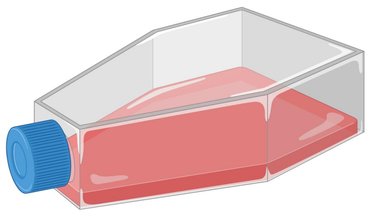
[1]Bioengineering Graduate Program, University of Kansas, Lawrence, KS

[2]Ronawk, Olathe, KS

[3]AMS Biotechnology (Europe) Ltd, UK

Technology & Methodology

Traditional 2D

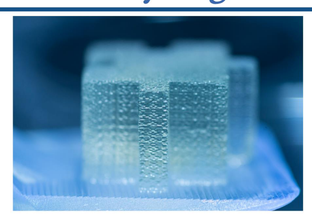


- 2D
- Rigid Plastic
- Unnatural Environment
- Overcrowding

→ Stemnss ↓, Health ↓, Total Number ↓, Regeneration ↓

Continuous Subculturing

3D Hydrogel



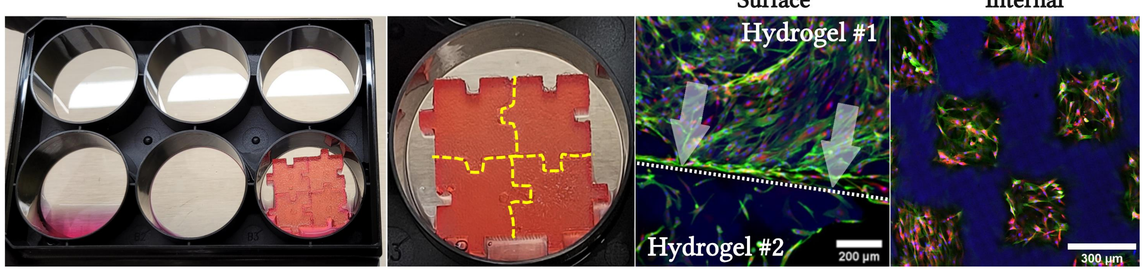
- 3D
- Soft/Flexible
- Tissue-Mimetic
- Modular
- More Efficent System
- Customizable

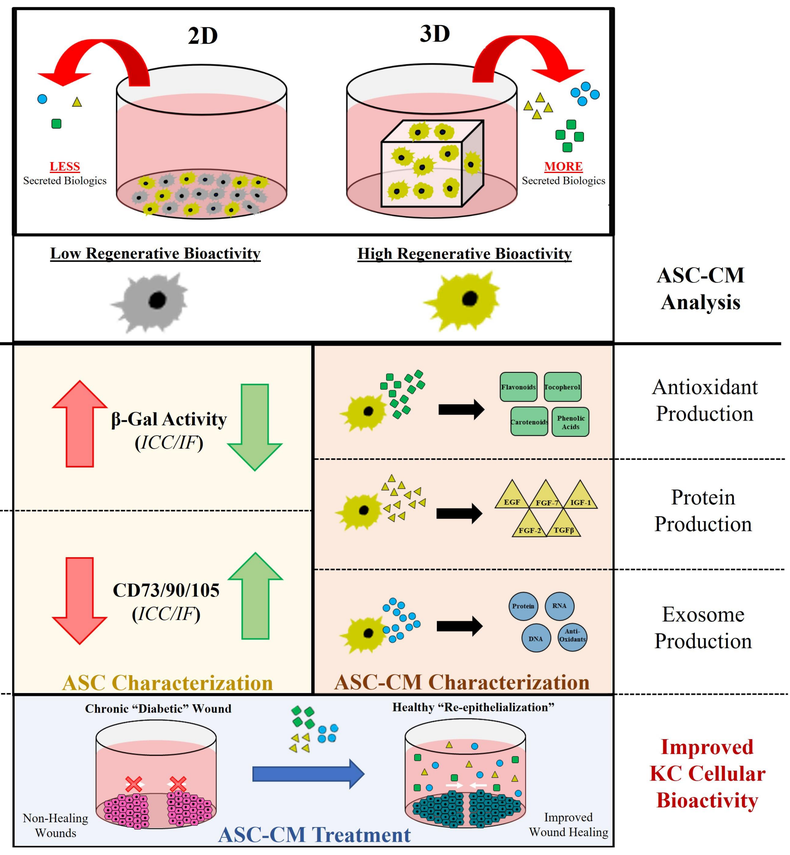
→ Stemnss ↑, Health ↑, Total Number ↑, Regeneration ↑, Efficiency ↑, Cost ↓

Eliminates Subculturing

Hypothesis

Expansion in novel 3D hydrogel system will result in enhanced secretion of therapeutic bioactive compounds from a more robust, "Stem-like" ASC population.





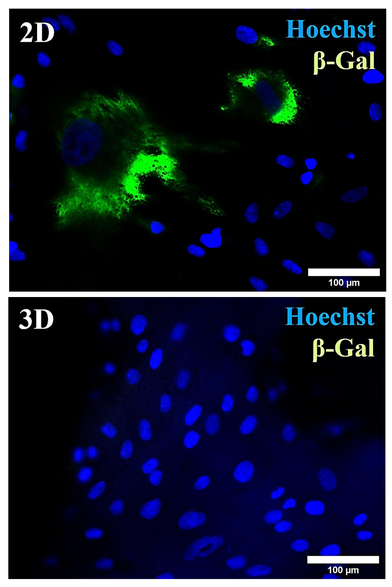
Decrease in **Senescence** and Increased Retainment of **"Stemness"** Phenotypic Markers

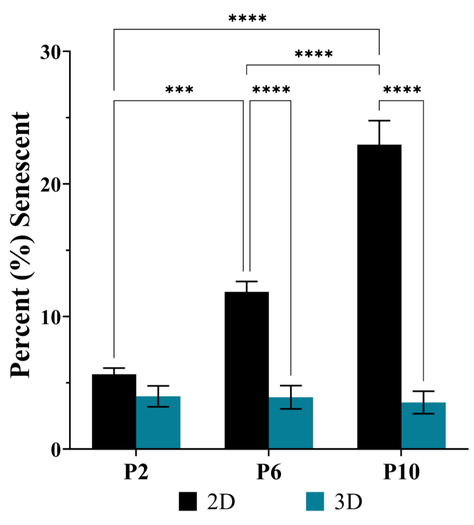
Graph

- Prevalence of **Senescence** overtime significantly **INCREASES** in 2D, with **NO CHANGE** in 3D.
- P2/6/10 indicate "passage" and/or "passage-equivalent" timepoint.

Representative Images

- Visual comparison of 2D and 3D expanded ASCs at "P5".
- "Green" indicates positive Senescent staining for β-Galactosidase.



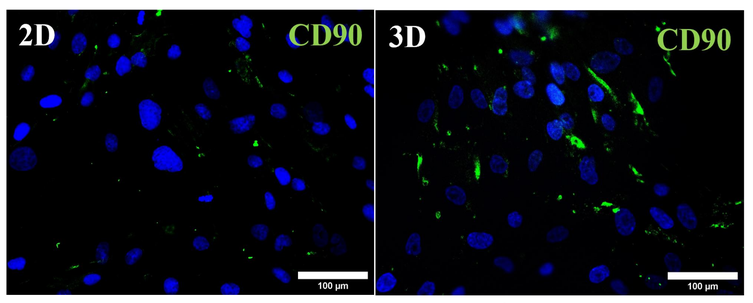


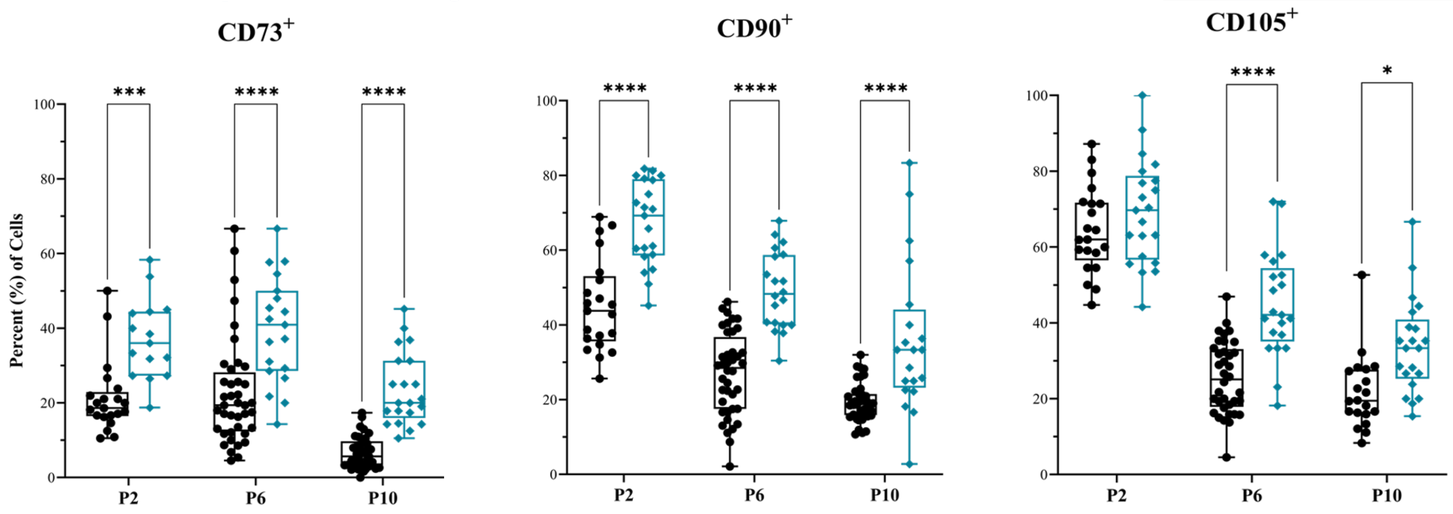
Graphs

- Significantly **INCREASED** retainment of "**Stemness**" markers overtime in 3D.
- P2/6/10 indicate "passage" and/or "passage-equivalent" timepoint.

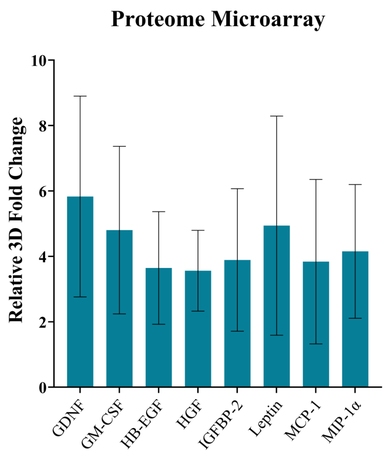
Representative Images

- Visual representation of "stemness" analysis for 2D and 3D expanded ASCs.
- "Green" indicates positive CD90 staining.

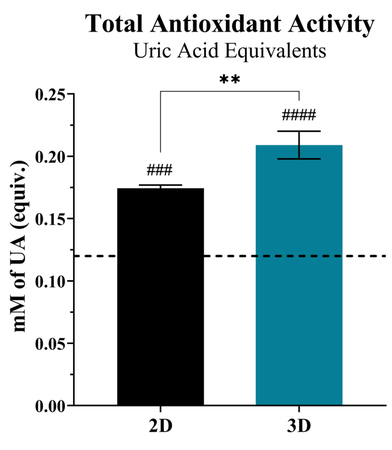




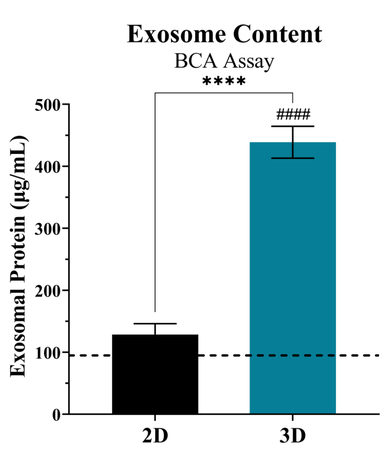
Augmented **Secretion of Regenerative Factors**



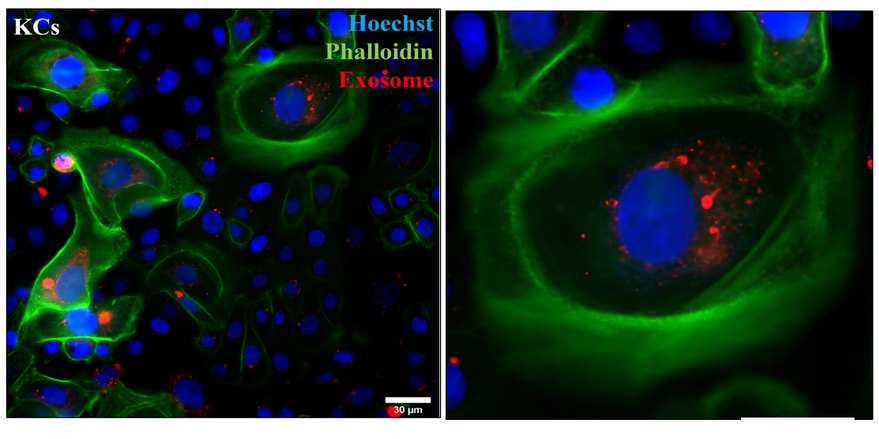
ASCs INCREASE Secretion of Soluble Proteins within ASC-CM in 3D System



ASCs INCREASE Secretion of Anti-Oxidant Factors within ASC-CM in 3D System

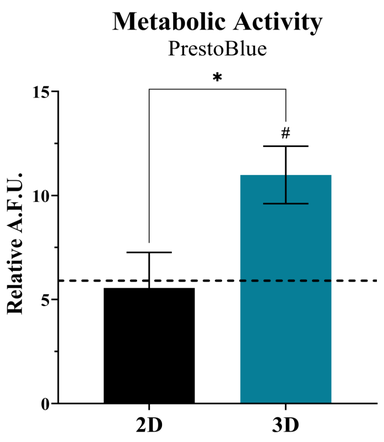


ASCs INCREASE Secretion of Exosomes within ASC-CM in 3D System

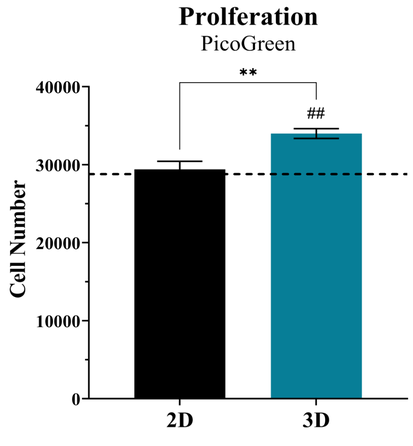


Keratinocytes (KCs) uptake of ASC-derived Exosomes in <18 hours and undergo Morphological Changes

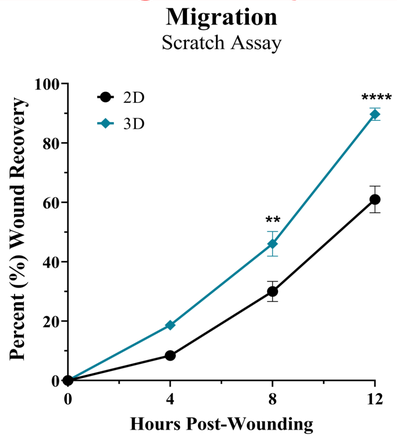
Enhanced **Wound Healing Activity** of Keratinocytes



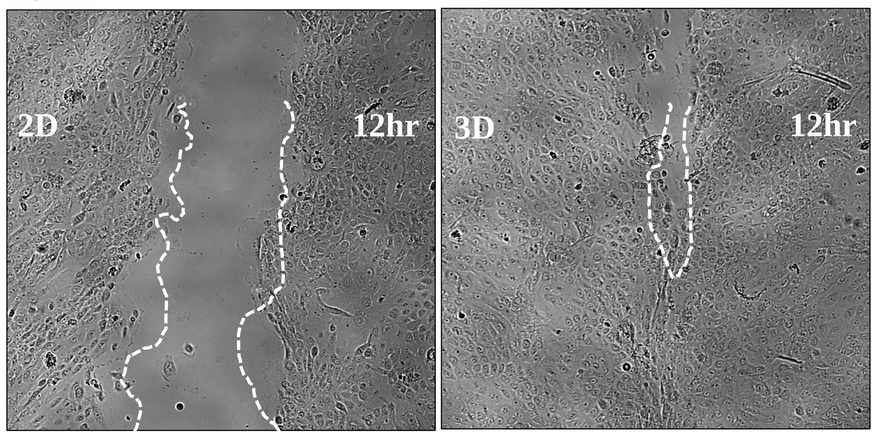
INCREASED Keratinocyte Metabolic Activity After Treatment with ASC-CM from 3D



INCREASED Keratinocyte Proliferative Activity After Treatment with ASC-CM from 3D



INCREASED Keratinocyte Migratory Activity After Treatment with ASC-CM from 3D



Performance of a Scratch Assay with Keratinocytes (KCs) treated with ASC-CM Treatment from 2D or 3D culture

www.amsbio.com