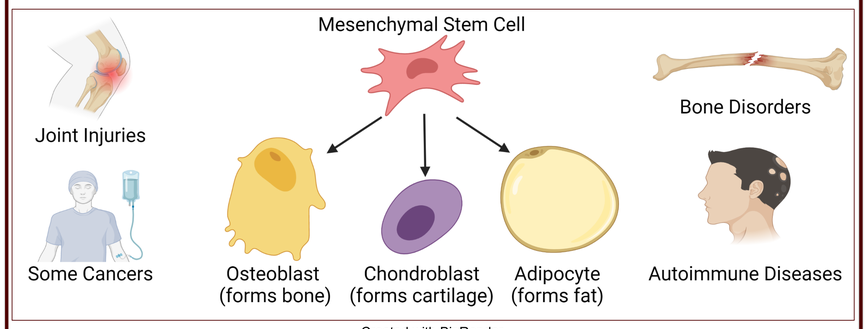
AFFORDABLE MANUFACTURE OF POWERFUL STEM CELL THERAPEUTICS

Andrew Haskell '22, Ph.D., PO1, USN (Ret.) | Ryang-Hwa Lee, Ph.D. | Roland Kaunas, Ph.D. | Carl Gregory, Ph.D.
Texas A&M School of Medicine

TEXAS A&M UNIVERSITY
School of Medicine

Mesenchymal stem cells (MSCs) exhibit characteristics that make them ideal to treat a variety of disorders



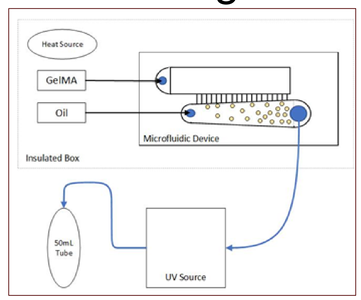
Created with BioRender

Current culture methods are too expensive and variable to safely manufacture enough cells for clinical use.

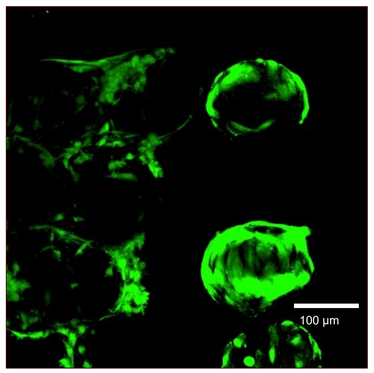
Hypothesis: MSCs can be manufactured in large bioreactors by fabricating spherical microcarriers made of gelatin methacryloyl (GelMA) to **create cost-effective therapeutics**.

GelMA microcarriers can be made without complex equipment or training

Aqueous GelMA forms tiny spheres in oil. They solidify when exposed to UV, creating microcarriers. Cells attach to the microcarriers as they move in bioreactors.



Microcarrier fabrication block diagram



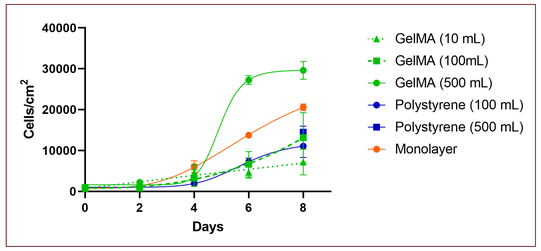
iPS-MSCs on GelMA microcarriers, green represents live cells.

Benefits:

- Permit cell growth in large bioreactors
- Compatible with standard culture protocols
- Batch customizable
- Degradable
- Easy to image under a microscope

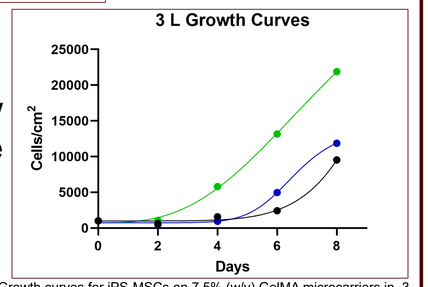
MSC culture on GelMA microcarriers is scalable to clinically-relevant quantities

Cell proliferation improves with bioreactor scale and exceeds commercial polystyrene (plastic) products.



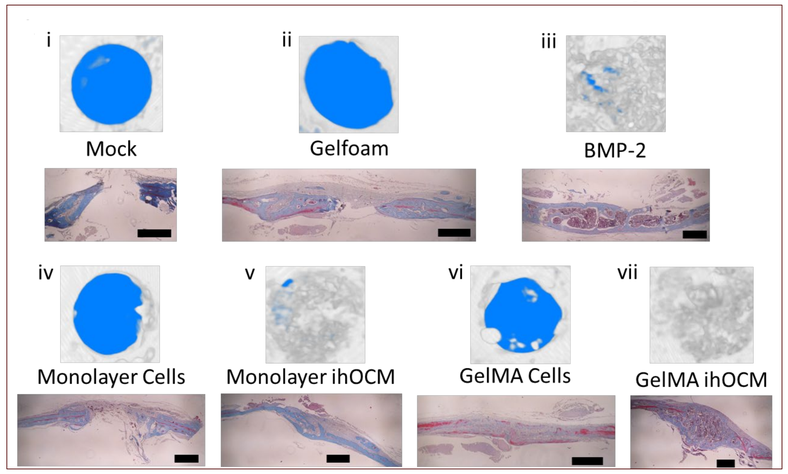
Growth curves of MSC cultures on 7.5% (w/v) GelMA microcarriers in 10 mL rotating wall vessel bioreactors, 100 mL vertical wheel bioreactors, or 500 mL VWBs compared to Pall SoloHill polystyrene microcarriers cultured in similar conditions and monolayer cultures on 10 cm culture plates (n = 3). 1% of culture volume was removed and counted every two days for bioreactor growth curves. 3 monolayer cultures were harvested on each time point. 500 mL polystyrene culture was endpoint only.

Large scale bioreactors allow for culture parameters to be optimized (i.e. pH and CO2).

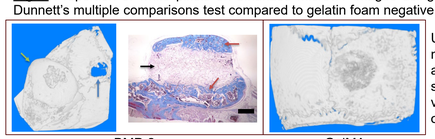


Growth curves for iPS-MSCs on 7.5% (w/v) GelMA microcarriers in 3 L vertical wheel bioreactors. Colors correspond to individual cultures.

Powerful bone regeneration without adverse effects of current therapeutics

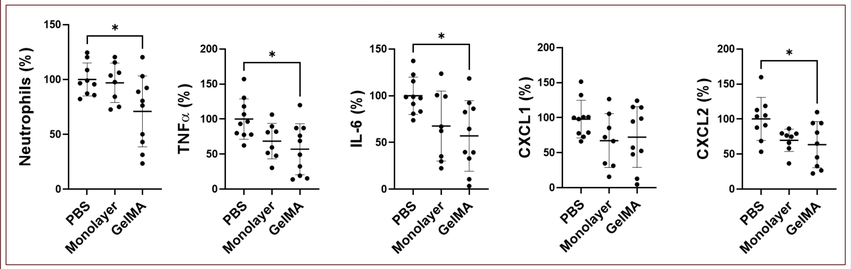


Left: Representative microCT (top) images of calvarial defect after 4 weeks of healing with Masson's trichrome stain (bottom). n = 4 for GelMA groups and monolayer osteogenic cell matrix (ihOCM), n = 5 for remaining groups. White represents bone, blue represents a void. Right: Proportion of white pixels on binarized microCT images using standardized volume of interest from mock defect. ANOVA with Dunnett's multiple comparisons test compared to gelatin foam negative control. Mock excluded from ANOVA.



Uncropped image of a BMP-2 specimen showing a large bony mass (green arrow) with no healing at the site of the defect (blue arrow) and histology revealing porous bone (red arrows) surrounding immature bone (black arrow). Uncropped image from vii displayed for comparison showing no bony masses or off-target osteogenesis.

Enhances inflammation suppression while minimizing production cost



Peritonitis induced in mice using Zymosan A followed by injections of phosphate buffered saline (PBS) or 2.5x10^6 cells. Lavage of the peritoneal cavity conducted after 5 hours. Immunomodulation determined by flow cytometry for CD-11b and Ly6G to determine neutrophil invasionand ELISAs for inflammatory cytokines. ANOVA with Dunnett's multiple comparisons test compared to PBS. n= 5. GelMA culture corresponds to blue curve in 3 L growth curve shown above with default bioreactor culture parameters.

iPS-MSC Culture Cost

	Per Culture	Per 1x10^6 Cells	Per Dose	Difference
Monolayer	$274.29	$203.18	$13,816.29	$0.00
100 mL	$1,317.03	$292.67	$19,901.82	$6,085.54
500 mL	$3,188.59	$141.72	$9,636.64	-$4,179.65
3 L (Polystyrene)	$18,667.26	$138.28	$9,402.77	-$4,413.52
3 L	$17,827.34	$132.05	$8,979.70	**-$4,836.59**

Prices of liquid components and culture vessels as of October, 2022. Does not account for common lab consumables, overhead, startup costs, or personnel salaries. Savings from GelMA microcarriers expected to *increase* when considering these factors with continuous production due to reduced time required from personnel and ability to work on multiple cultures concurrently.

Conclusion

MSC culture on GelMA microcarriers is scalable, enhances therapeutic potential, and can save patients over $4800 per dose.

Funding

Funded in part by grants from the **President's Excellence in Research Grant** (X-Grant) by Texas A&M University and the **National Institutes of Health** (R01AR066033-01). Access to BioRender provided by the Texas A&M Institute for Genome Sciences and Society to all faculty, staff, and students.

The presenter was additionally supported by scholarships from the Texas A&M University **Association of Former Students** and the **Knauss Family AggieVet Freedom Fund**.

View my 2-minute presentation:
tx.ag/VRS23hask