MSCS AND EXOSOME/EV THERAPEUTICS

LESSONS LEARNED AND A PROMISING FUTURE- VITACYTE WEBINAR SERIES

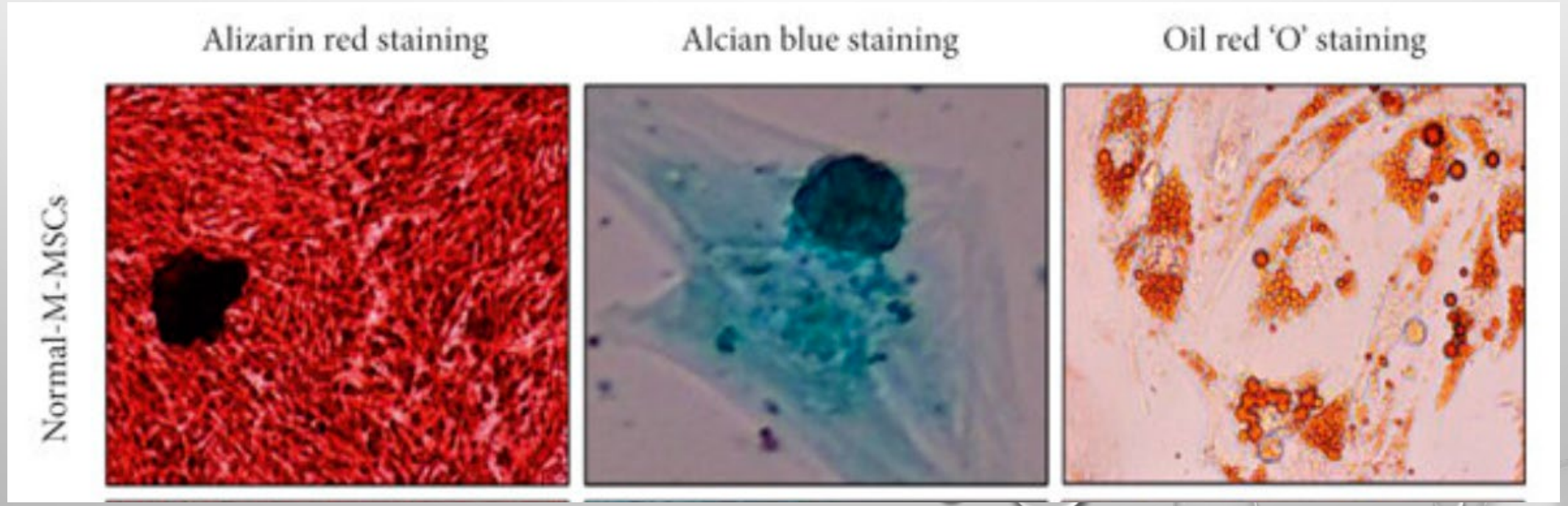
9/28/2023

DISCLOSURE STATEMENT

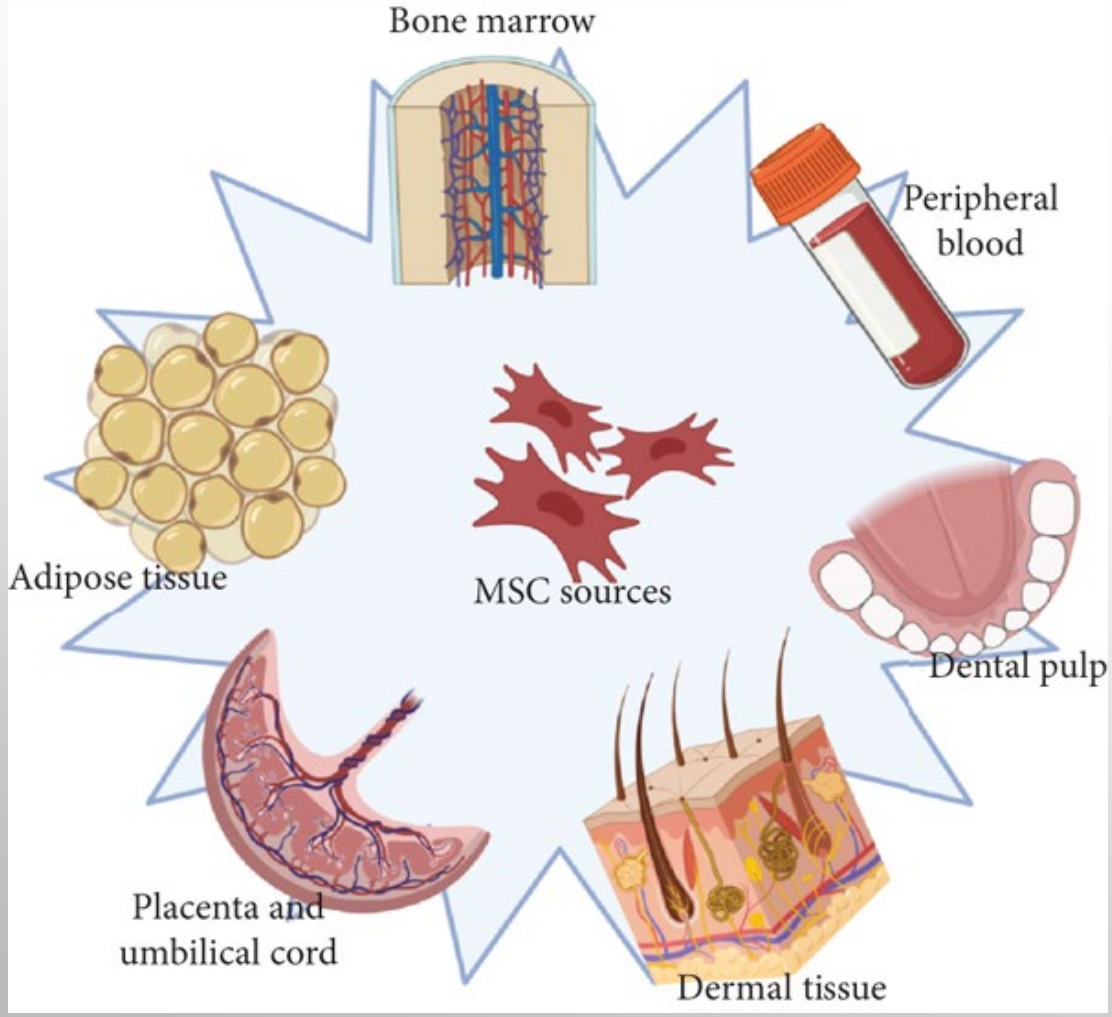
- I AM CURRENTLY CONSULTING BUT ALSO A COFOUNDER AND CHIEF SCIENCE OFFICER FOR A STEALTH START UP IN THE REGENERATIVE MEDICINE SPACE.
- THE PERSPECTIVES AND OPINIONS I SHARE IN THIS WEBINAR ARE MY OWN AND ARE NOT MEANT TO REPRESENT THOSE OF MY CURRENT OR PREVIOUS EMPLOYERS AND COLLEAGUES OR THE SPONSOR OF THIS WEBINAR.

MSCS-WHERE IT STARTED:

- FRIEDENSTEIN AND OWEN-UNIVERSITY OF MOSCOW- late 60s to late 80s-identified fibroblast like cells in the bone marrow referred to as “mesenchymal stromal cells” that supported hematopoiesis and could differentiate into osteoblasts, adipocytes and chondrocytes.
- Jim Burns and Arnold Caplan founded Osiris Therapeutics in Baltimore in 1992 to develop the first BM MSC therapies. Caplan coined and popularized the term “Mesenchymal Stem Cells”



SO WHERE ARE MSCS FOUND AND WHAT ARE THEY?



Most commonly located in the external vascular niche
Abundance varies from tissue to tissue-
Bone marrow 1:10,000 to 1:1,000,000

Adipose: 1% to 10% of total Non-adipocyte cells isolated

Umbilical Cord/WJ/Placenta 5-10% of isolated adherent cells

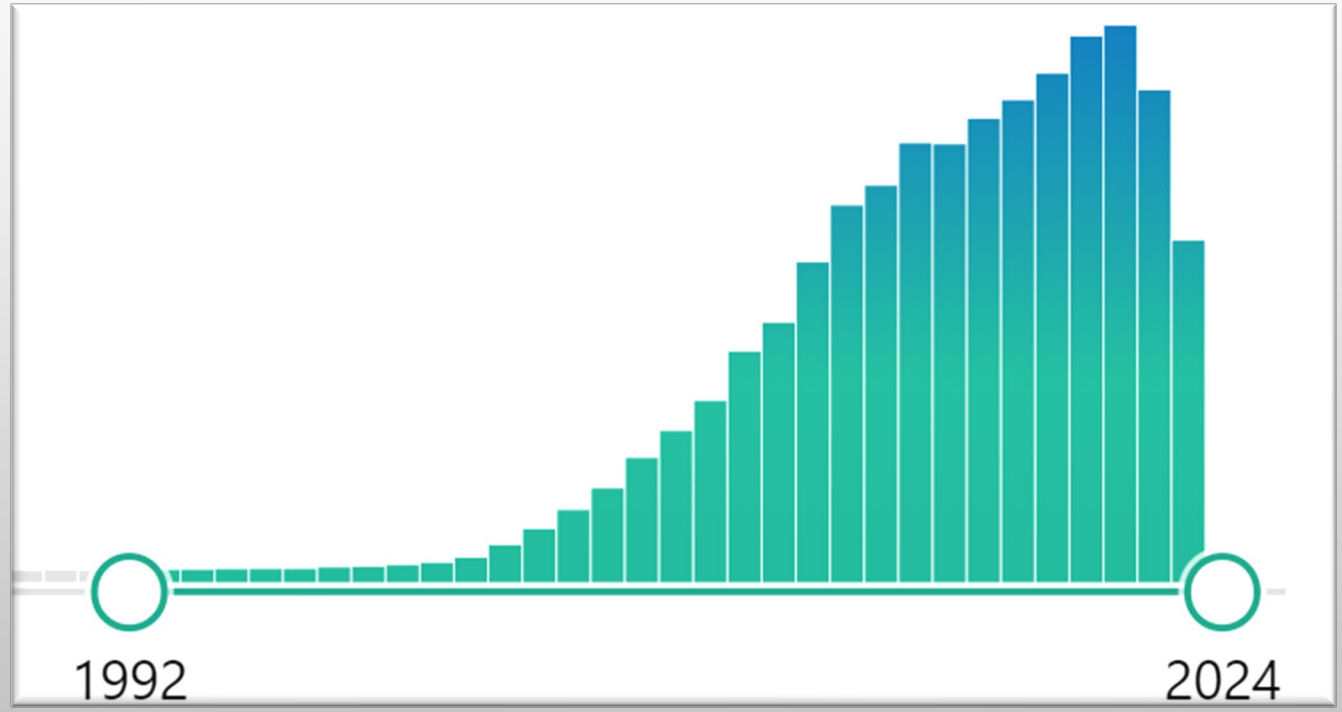
Peripheral blood concentration less than BM

Function to repair and remodel disrupted vasculature, ECM, and modulate immune cell activity, role in tissue turnover depends on cell phenotype.

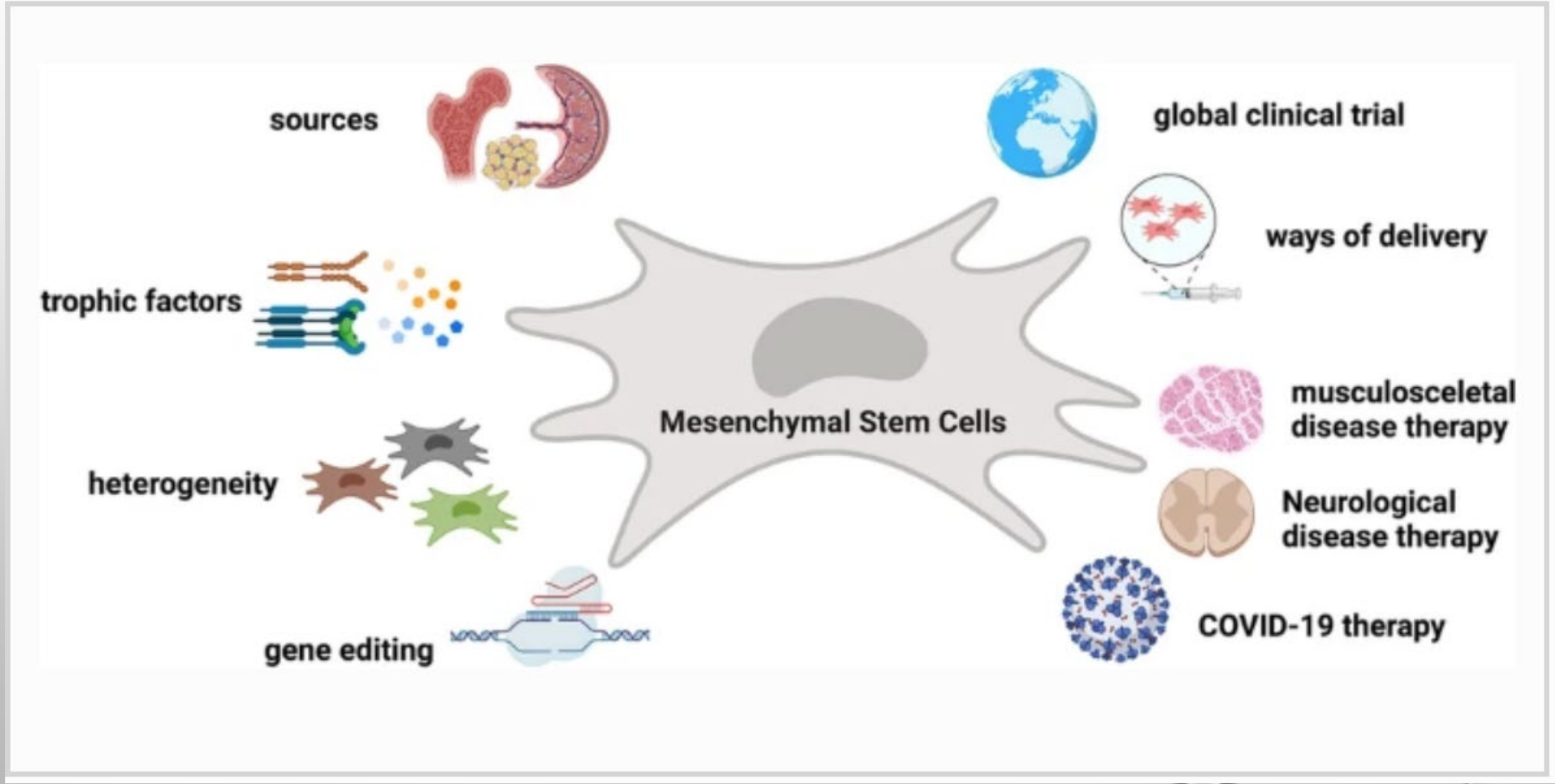
https://www.hindawi.com/journals/sci/2020/8892189/

THE BOOM OF THE MSC INDUSTRY

- GREATER THAN 88,000 PUBLICATIONS
- 2021 PEAK PUBLICATION YEAR: 8505



1014 CLINICAL TRIALS RECORDED AS OF 2021
10,000+ PRECLINICAL STUDIES



AFTER 30 YEARS-HOW FAR HAVE WE COME?

MSC Therapy, Company and Target Indication	Regulatory Status
Alofisel (Tigenix/Takeda) Crohn's Disease	EMA and Japan
Chondrocytes-T-Ortho-ACI-articular cartilage defects	Australian Register of Therapeutic Goods
Spherox-Relive Biotechnologies LTD-cartilage repair	EMA
Ossgrow-Regrow Biosciences Pvt Ltd- autologous osteoprogenitors Avascular osteonecrosis	India DCGI
Stempeucel-Stempeutics-BMSCs-Critical Limb Ischemia	India DCGI
Prochymal (remestemcel-L)- (Originally Osiris IP) Mesoblast-GVHD Temcell HS-Mesoblast-GVHD	Canada
	Japan
Neuronata-R-Corestem/ Autologous BMSCs for ALS	South Korea KFDA
Cupistem-Anterogen-Adipose MSCs for Crohn's Disease	South Korea KFDA
Cellgram-AMI-BioStar Rbio- MSCs to treat A Myocard Infarct	South Korea KFDA
NurOwn-BrainStorm BM-MSCs to treat ALS	BLA rejected-w/ refuse to file letter at

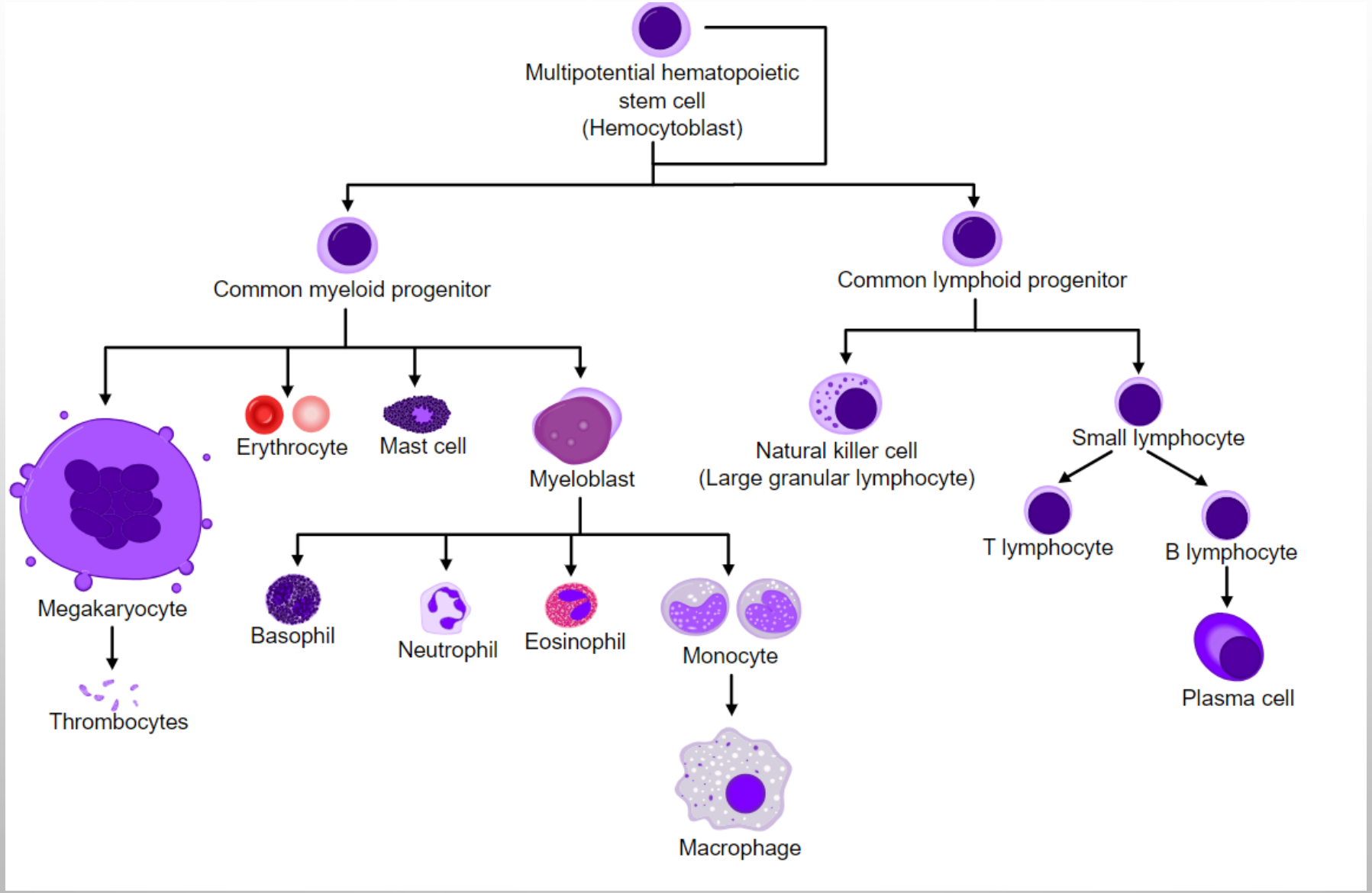
CHALLENGES TO GET ACROSS THE FINISH LINE

“LET THE DATA SPEAK TO YOU, DON’T SPEAK TO THE DATA”

DR. B. LAWRENCE RIGGS, MD, MAYO CLINIC

- WHAT MSCS ARE AND WHAT THEY ARE NOT-ASSUMPTIONS VERSUS EMPIRICAL DATA
- MULTIPLE MECHANISMS OF ACTION
- REGULATORY CHALLENGES: FITTING A SQUARE PEG INTO A ROUND HOLE
- POTENCY AND CLINICAL TRIAL DESIGN

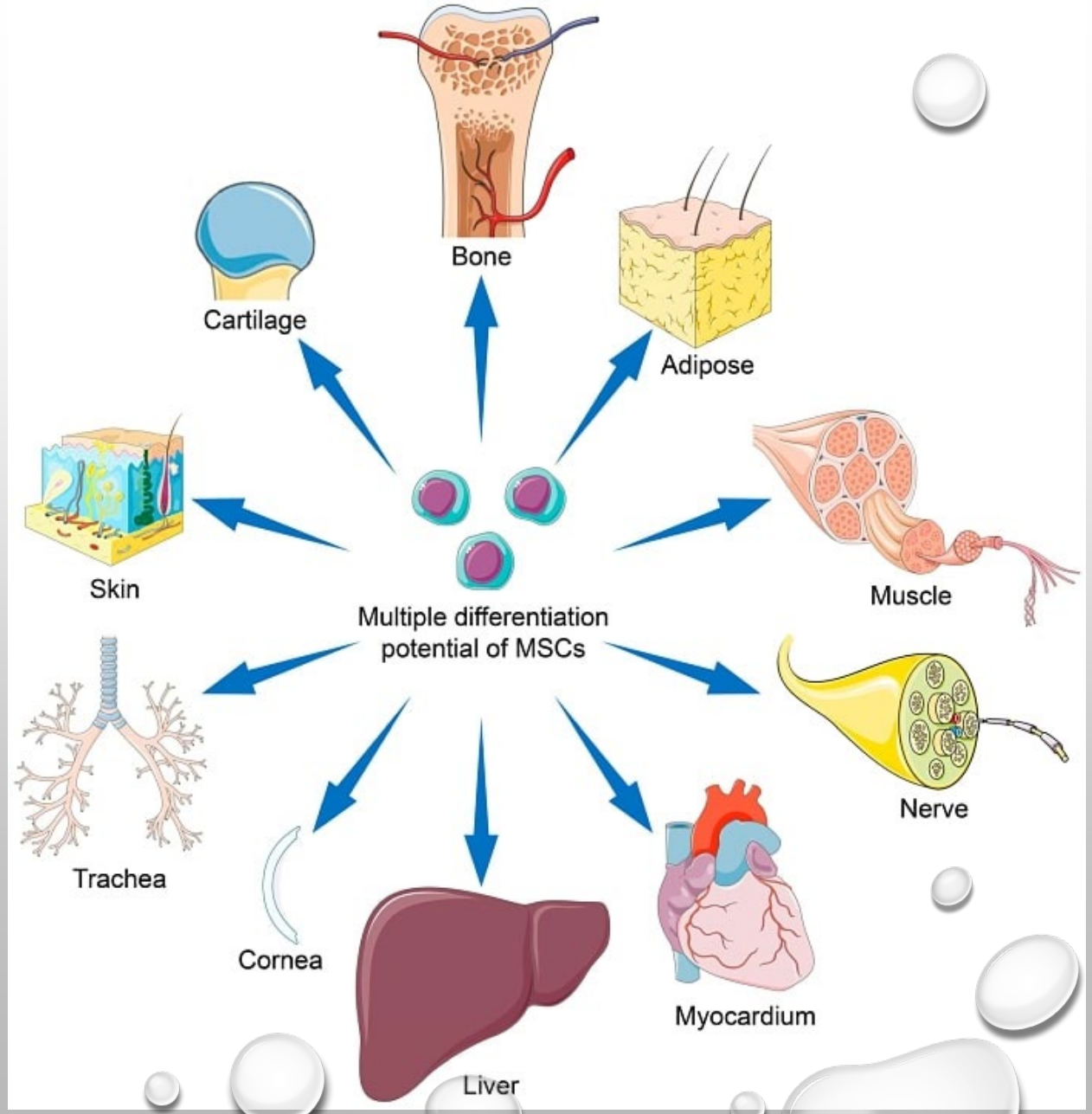
THE HSC ASSUMPTION



UNDERSTANDING WHAT MSCS ARE AND ARE NOT

- IN VITRO – UNDER THE RIGHT CULTURE CONDITIONS MSCS CAN BE COAXED TO DISPLAY MANY MATURE CELL PHENOTYPES

- IN VIVO- THE MICROENVIRONMENT DOES NOT TYPICALLY SUPPORT SIGNIFICANT CELL EXPANSION AND DIFFERENTIATION INTO MATURE NEW TISSUE



https://uploads-ssl.webflow.com/5d9667bad4a41e222995e15b/5df3e5bce2fb7e5258abc022_stem%20cells%20chart_opt.jpg

DIFFERENTIATION OF MSCS ESTABLISHED BUT IS IT CLINICALLY RELEVANT?

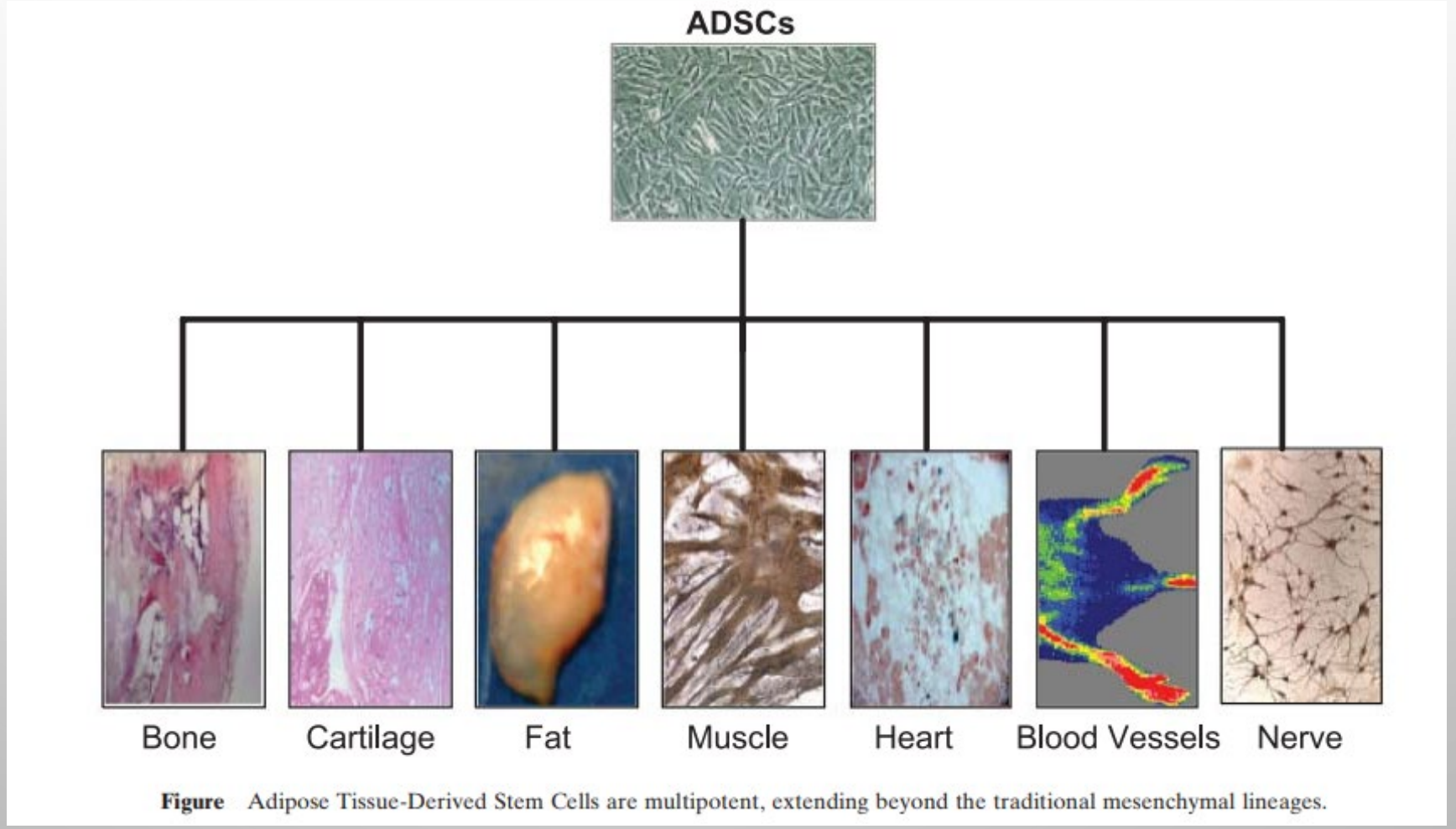


Figure Adipose Tissue-Derived Stem Cells are multipotent, extending beyond the traditional mesenchymal lineages.

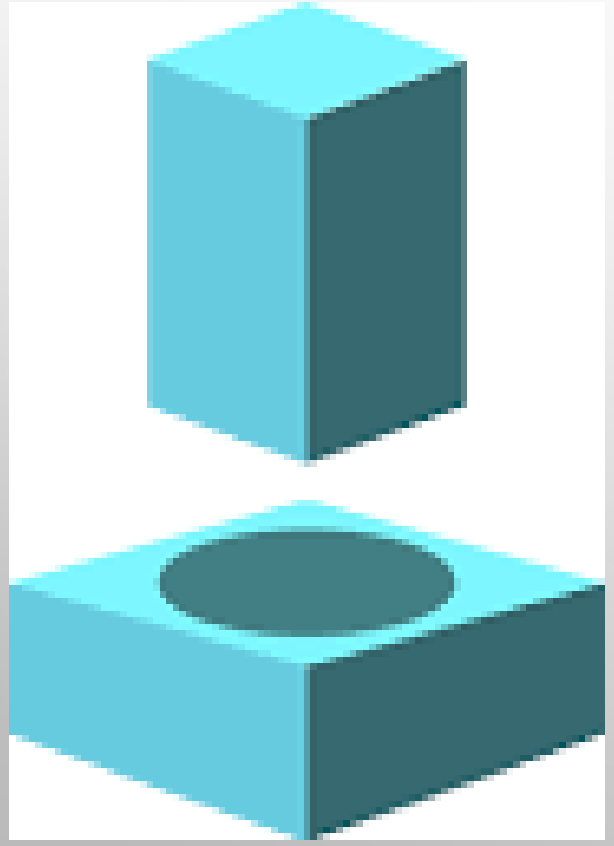
Brian M Strem, **Kevin C Hicok**, Min Zhu, Isabella Wulur, Zeni Alfonso, Ronda E Schreiber, John K Fraser, Marc H Hedrick, Multipotential differentiation of adipose tissue-derived stem cells, The Keio Journal of Medicine, 2005, Volume 54, Issue 3, Pages 132-141, Released on J-STAGE June 20, 2006, Online ISSN 1880-1293, Print ISSN 0022-9717, https://doi.org/10.2302/kjm.54.13

EVIDENCE OF β-ISLET CELL DIFFERENTIATION

- 2004, Chen et al reported in vitro differentiation of rat MSCs to a cell phenotype that expressed increased insulin by RT-PCR and RIA. In a small pilot study, a trend toward improved glucose regulation in STZ diabetic rat model where these differentiated cells were transplanted.
  - Advent of IPSC discovery led to shift toward developing B-cell transplants using them as source
  - MSCs still found to have a roll to play.
  - Encapsulation- provides immunoprotection
  - Promotes vascularization in/around transplanted islets

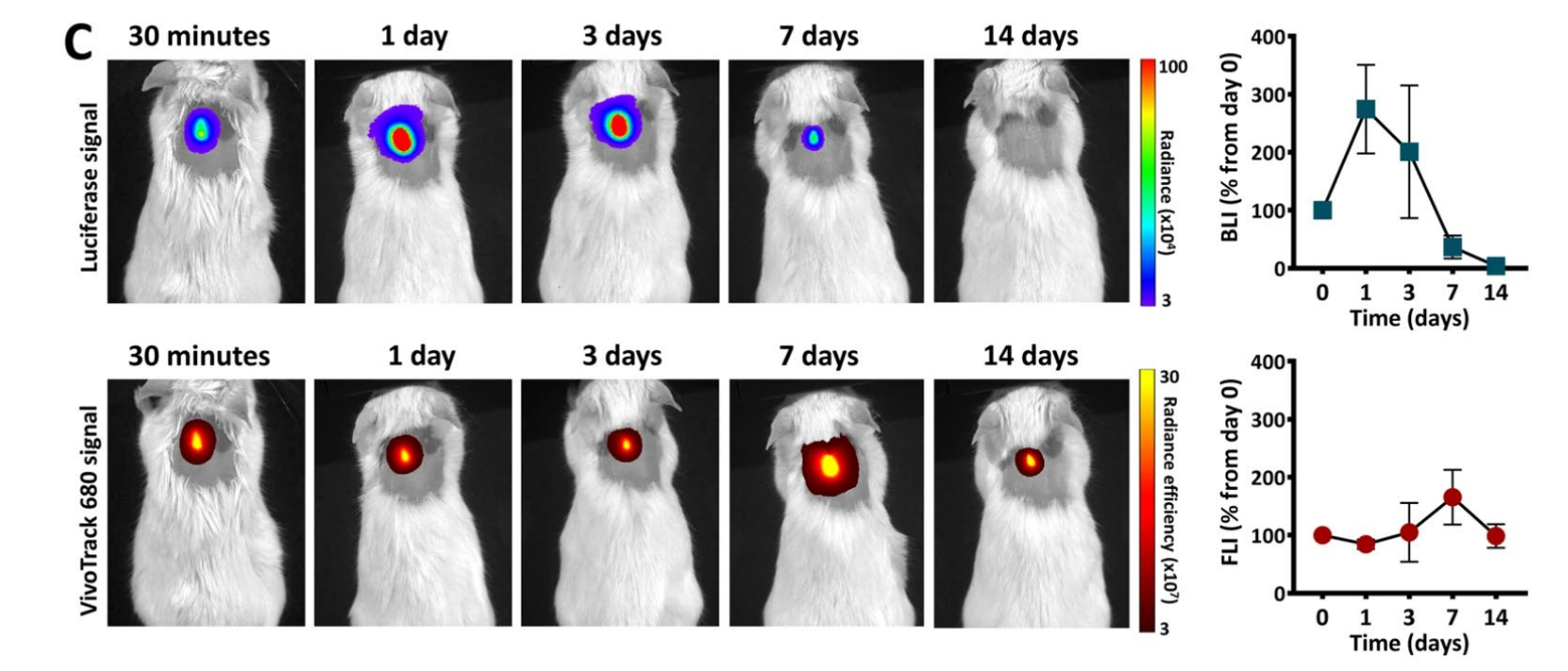
Chen LB, Jiang XB, Yang L. Differentiation of rat marrow mesenchymal stem cells into pancreatic islet beta-cells. World J Gastroenterol. 2004 Oct 15;10(20):3016-20. doi: 10.3748/wjg.v10.i20.3016. PMID: 15378785; PMCID: PMC4576264.

THE REGULATORY HURDLE, CHALLENGES OF EVOLUTIONARY ENFORCEMENT ON REVOLUTIONARY TECHNOLOGIES



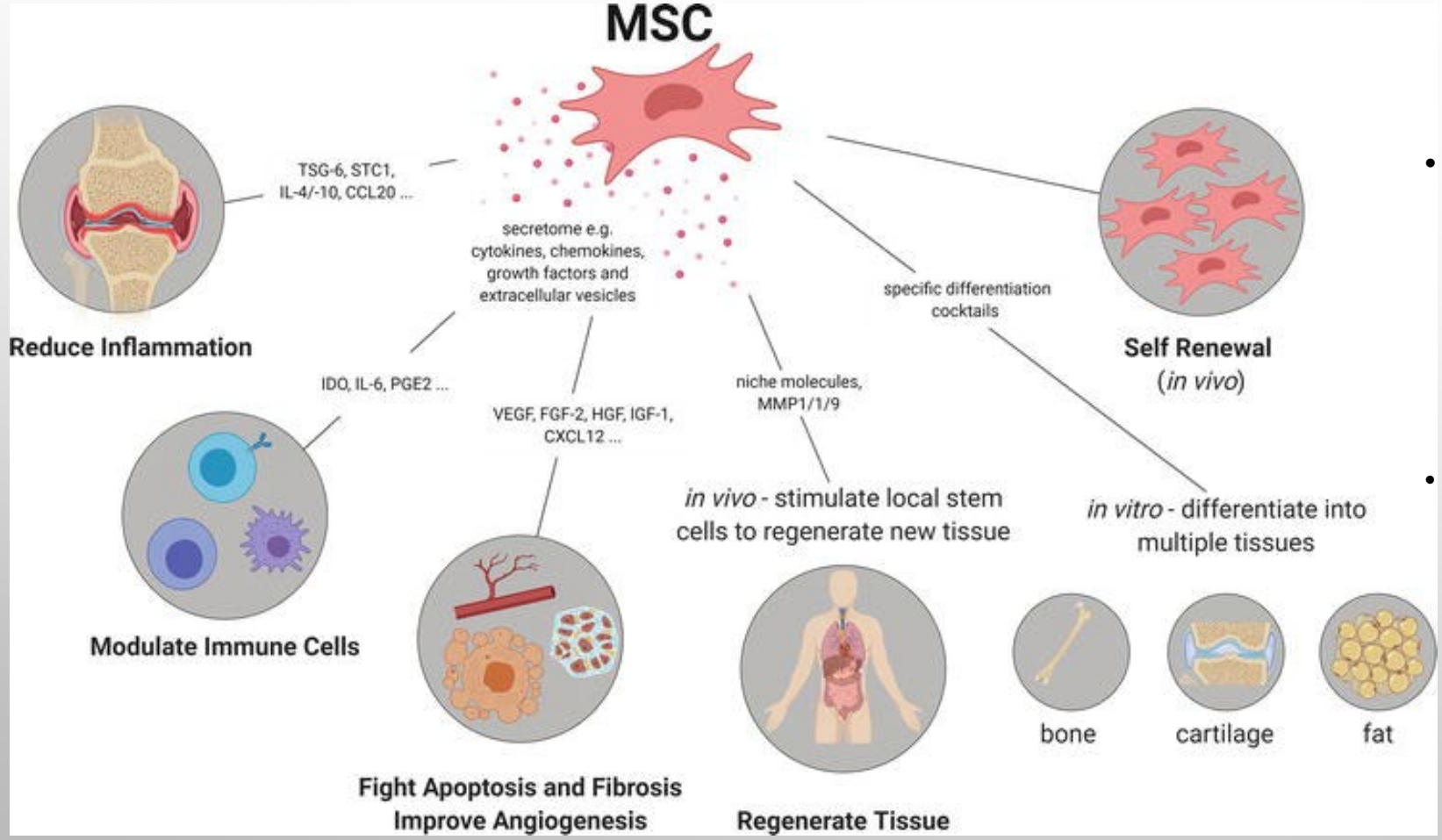
- THE SMALL MOLECULE PARADIGM IS DIDN'T ALIGN WITH COMPLEX CELL SOLUTIONS
- SAFETY PARAMETERS ARE SIMILAR
- PURITY AND POTENCY REQUIREMENTS FOR CELL THERAPIES INVOLVING 100S OR 1OOOS OF MOLECULAR INTERACTIONS POSE A SIGNIFICANT CHALLENGE EVEN WITH CURRENT CGT GUIDANCES

MOST MSCS DON'T STICK AROUND FOR VERY LONG BUT A LONG-LASTING EFFECT IS OBSERVED



Preda, M.B., Neculachi, C.A., Fenyo, I.M. *et al.* Short lifespan of syngeneic transplanted MSC is a consequence of in vivo apoptosis and immune cell recruitment in mice. *Cell Death Dis* **12**, 566 (2021). https://doi.org/10.1038/s41419-021-03839-w

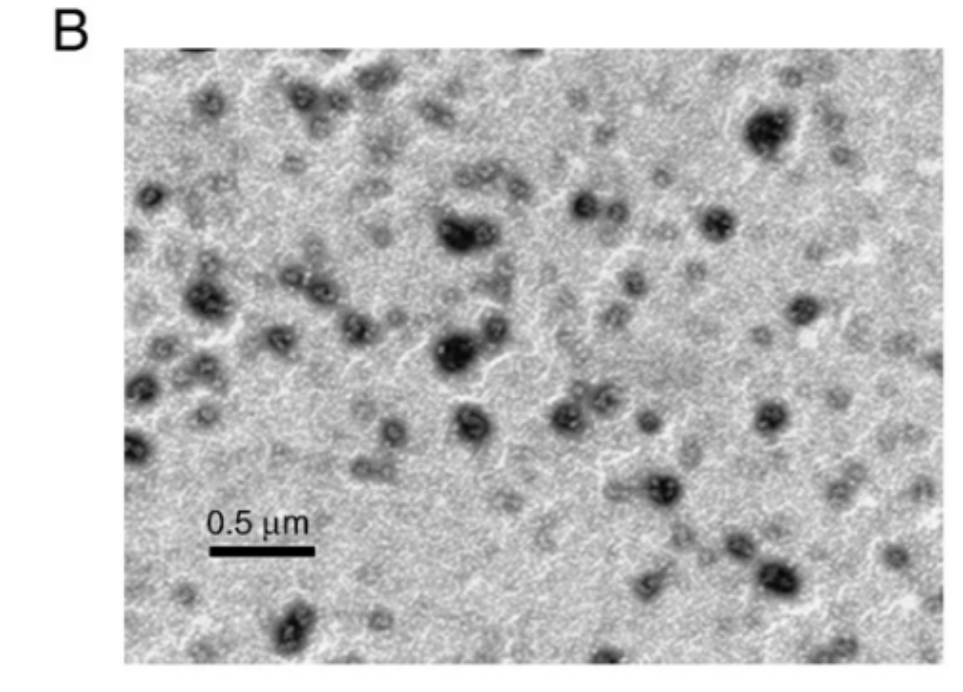
EFFICACY IN PRECLINICAL MODELS OBSERVED USING THE CONDITIONED MEDIUM ALONE

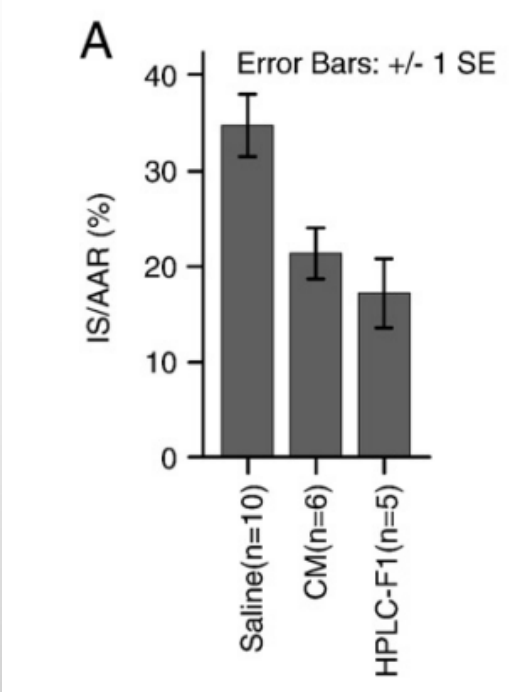


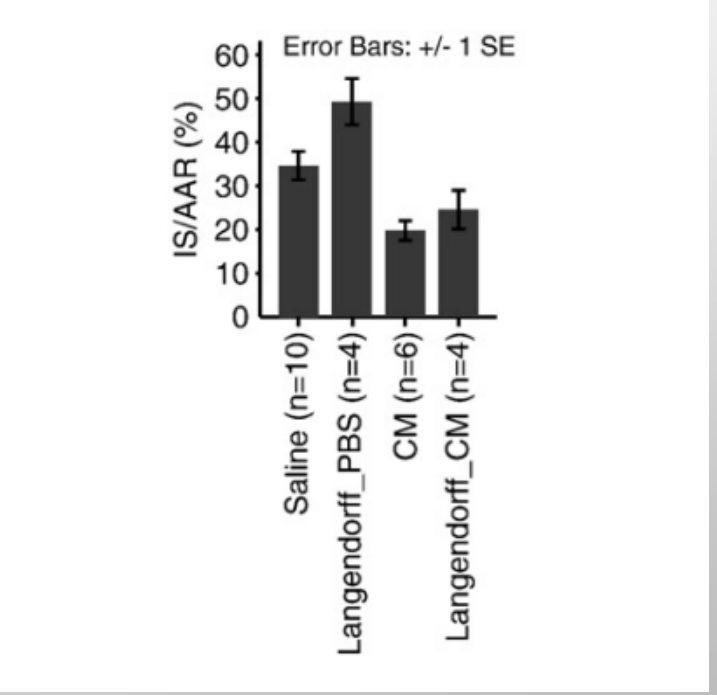
https://www.intechopen.com/media/chapter/69881/media/F1.png

- Rasmusson I, Ringdén O, Sundberg B, Le Blanc K. Mesenchymal stem cells inhibit the formation of cytotoxic T lymphocytes, but not activated cytotoxic T lymphocytes or natural killer cells. Transplantation. 2003 Oct 27;76(8):1208-13. doi: 10.1097/01.TP.0000082540.43730.80. PMID: 14578755.
- Rehman J, Traktuev D, Li J, Merfeld-Clauss S, Temm-Grove CJ, Bovenkerk JE, Pell CL, Johnstone BH, Considine RV, March KL. Secretion of angiogenic and antiapoptotic factors by human adipose stromal cells. Circulation. 2004 Mar 16;109(10):1292-8. doi: 10.1161/01.CIR.0000121425.42966.F1. Epub 2004 Mar 1. PMID: 14993122.

DR. SK LIM TEAM; 2010 DEMONSTRATED EXOSOME ROLE IN MSC MOA IN HEART INJURY MODELS

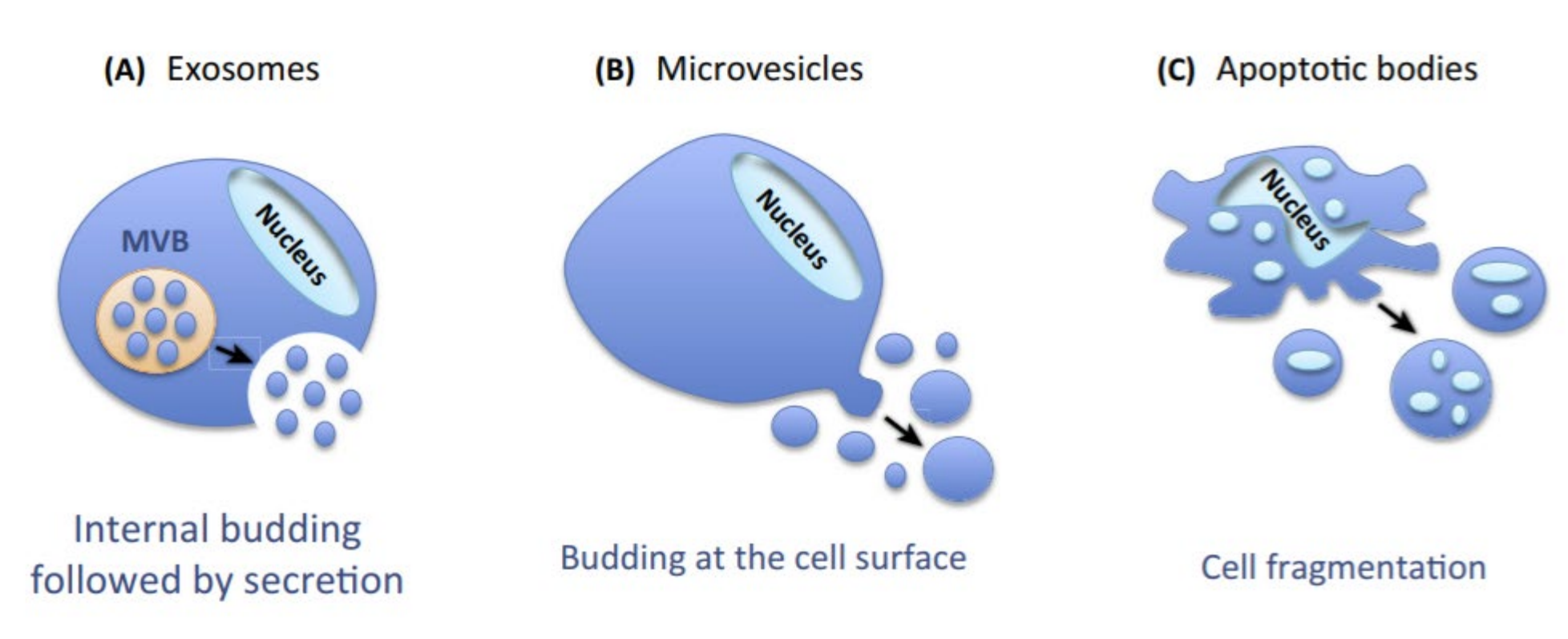






Lai, R. C., Arslan, F., Lee, M. M., Sze, N. S. K., Choo, A., Chen, T. S., … Lim, S. K. (2010). Exosome secreted by MSC reduces myocardial ischemia/reperfusion injury. Stem Cell Research, 4(3), 214–222. doi:10.1016/j.scr.2009.12.003

DIFFERENT TYPES OF EXTRACELLULAR VESICLES

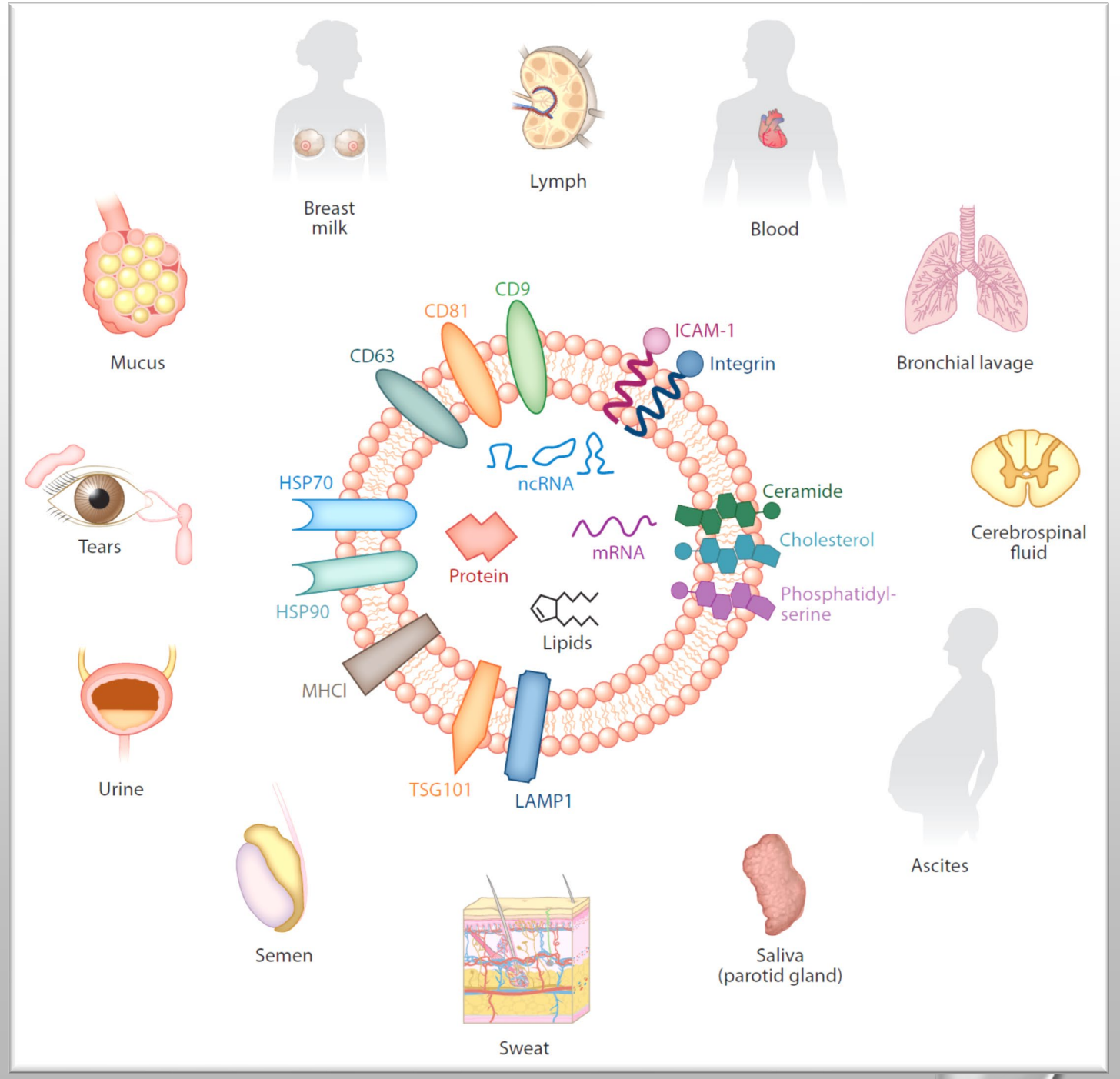


(30-150 nm)

(100-1000 nm)

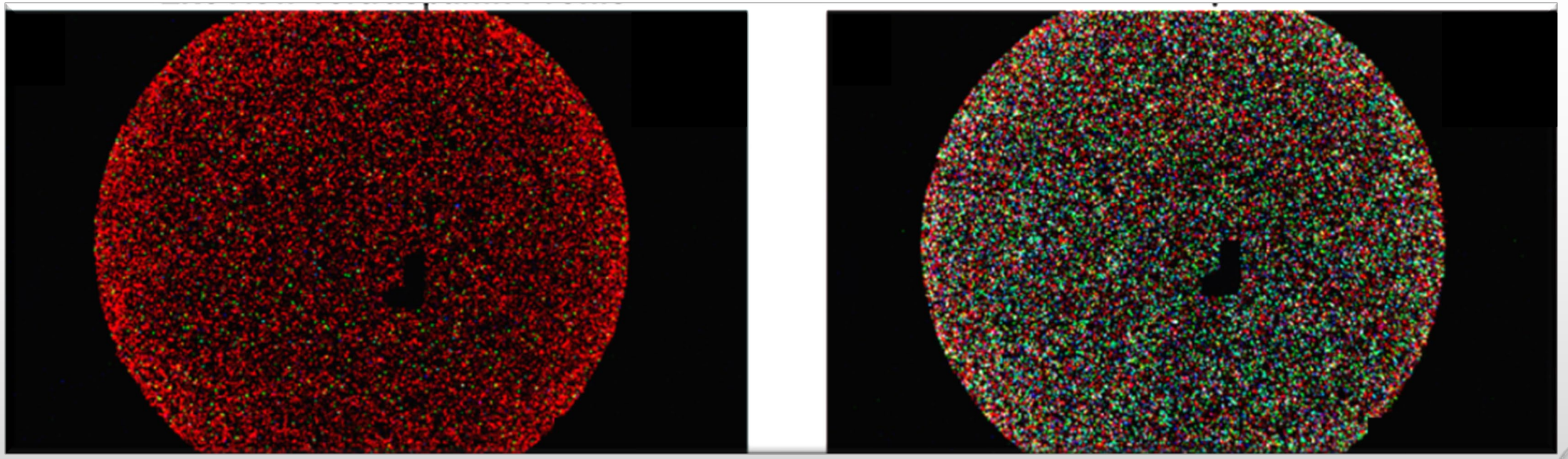
(100-5000 nm)

EXOSOMES ARE SECRETED BY ESSENTIALLY EVERY CELL IN YOUR BODY

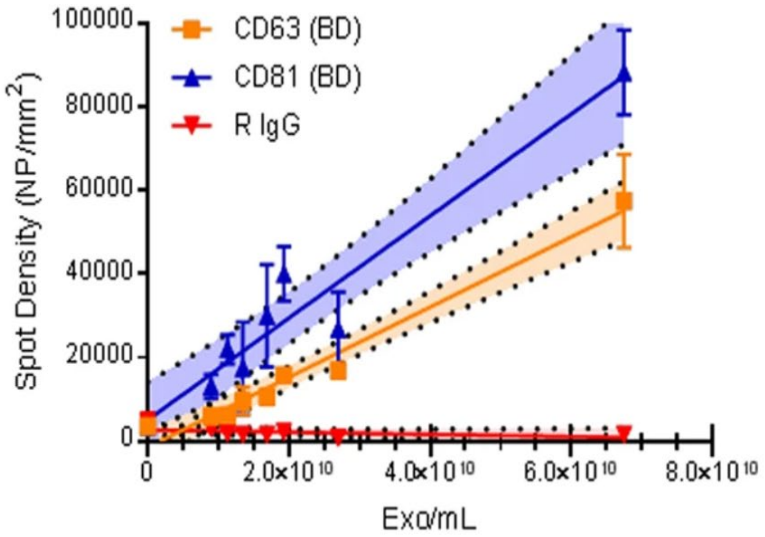


Ibrahim and Marbán. Exosomes: Fundamental Biology and Roles in Cardiovascular Physiology. *Annual Reviews of Physiology* 2016. 78:67–83.

Interferometric Imaging of EVs



From: Digital Detection of Exosomes by Interferometric Imaging



Dilution curve of exosomes purified from HEK cell line and detected with SP-IRIS.

DIFFERENT CELL TYPES AND MANUFACTURING CONDITIONS AFFECT MOLECULAR CONTENT OF EVS

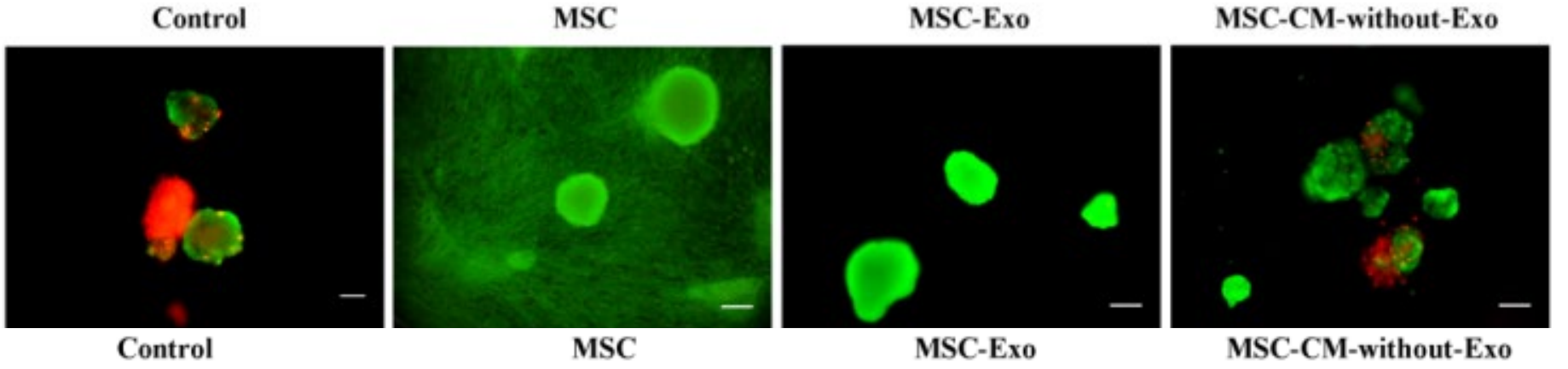
EXCLI Journal 2020;19:1064-1080 – ISSN 1611-2156
Received: May 23, 2020, accepted: July 20, 2020, published: August 03, 2020

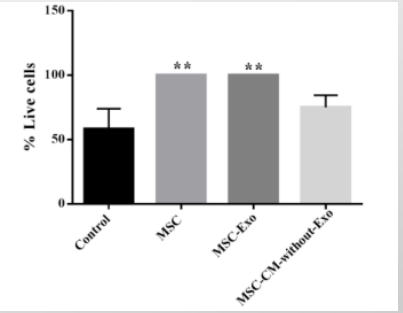
Original article:

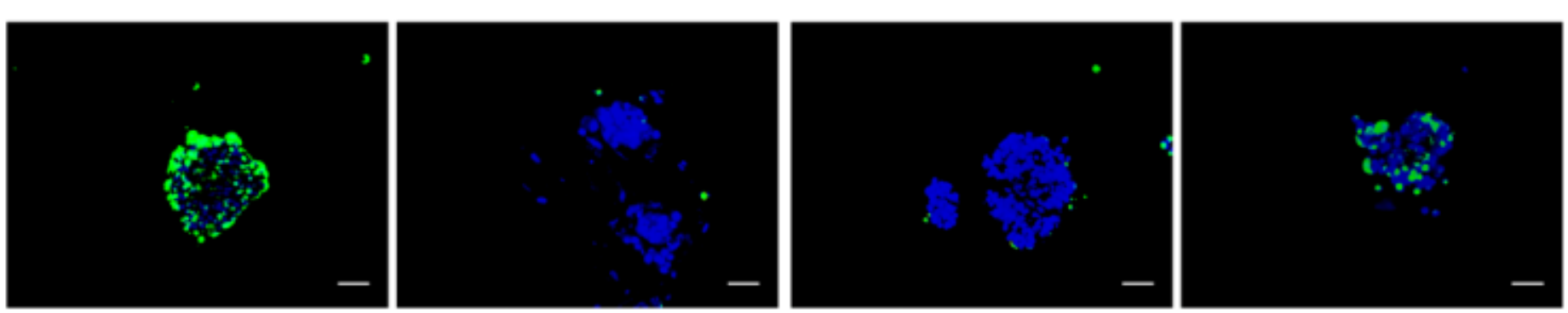
EXOSOMES DERIVED FROM HUMAN MESENCHYMAL STEM CELLS PRESERVE MOUSE ISLET SURVIVAL AND INSULIN SECRETION FUNCTION

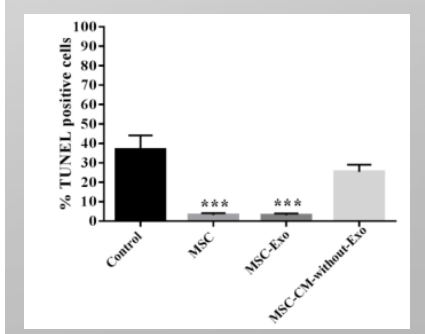
Somayeh Keshtkar[1,2], Maryam Kaviani[2], Fatemeh Sabet Sarvestani[2], Mohammad Hossein Ghahremani[1], Mahdokht Hossein Aghdaei[2], Ismail H. Al-Abdullah[3], Negar Azarpira[2*]

- MSC-DERIVED EXOSOMES ARE AS EFFICIENT AS PARENT MSCS FOR MITIGATING CELL DEATH AND IMPROVING ISLET SURVIVAL AND FUNCTION









OPTIMIZATION FOR MSC-BASED THERAPY FUTURE

- THE EXTRACELLULAR VESICLE/EXOSOME APPROACH ADDRESSES SOME THE MAJOR CHALLENGES OF MSC THERAPEUTICS.
  - NOT AFFECTED BY POTENTIALLY HOSTILE MICROENVIRONMENTS
  - COLD SUPPLY CHAIN AND STABILITY ISSUES ADDRESSED
  - BETTER MANUFACTURING SCALABILITY
  - BIOLOGIC REGULATORY STRATEGIES ARE EASIER TO TRANSLATE THAN THE CELLS ALONE

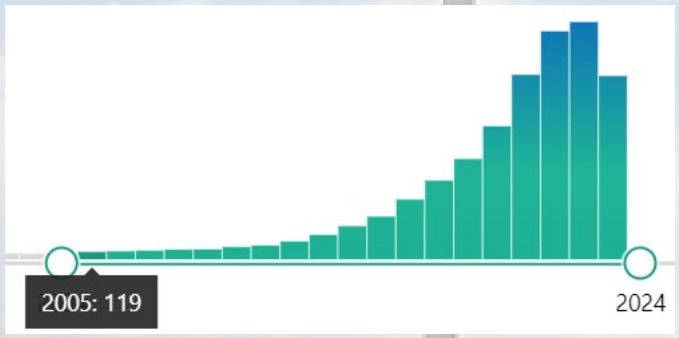
THE FINISH LINE IS IN SIGHT!

AEGLE THERAPEUTICS

First Patient Dosed in Phase I/2a Clinical Trial to Treat Burn Wounds

DIRECT BIOLOGICS™

Direct Biologics Announces FDA Authorization to Expand Ongoing Phase 3 Clinical Study of ExoFlo™ to All-Cause Moderate-to-Severe ARDS



- THE GLOBAL EXOSOMES MARKET SIZE WAS VALUED AT USD 112.25 MILLION IN 2022 AND IS ANTICIPATED TO EXPAND AT A COMPOUND ANNUAL GROWTH RATE (CAGR) OF 32.75% BY 2030.

https://www.grandviewresearch.com/industry-analysis/exosomes-market#:~:text=Report%20Overview,within%20a%20single%20outer%20membrane.

THANK YOU!

KEVHICOK3@OUTLOOK.COM