UNIVERSITY of VIRGINIA

ORTHOPAEDIC SURGERY

Mesenchymal Stem Cells (MSCs) for Bone Regeneration

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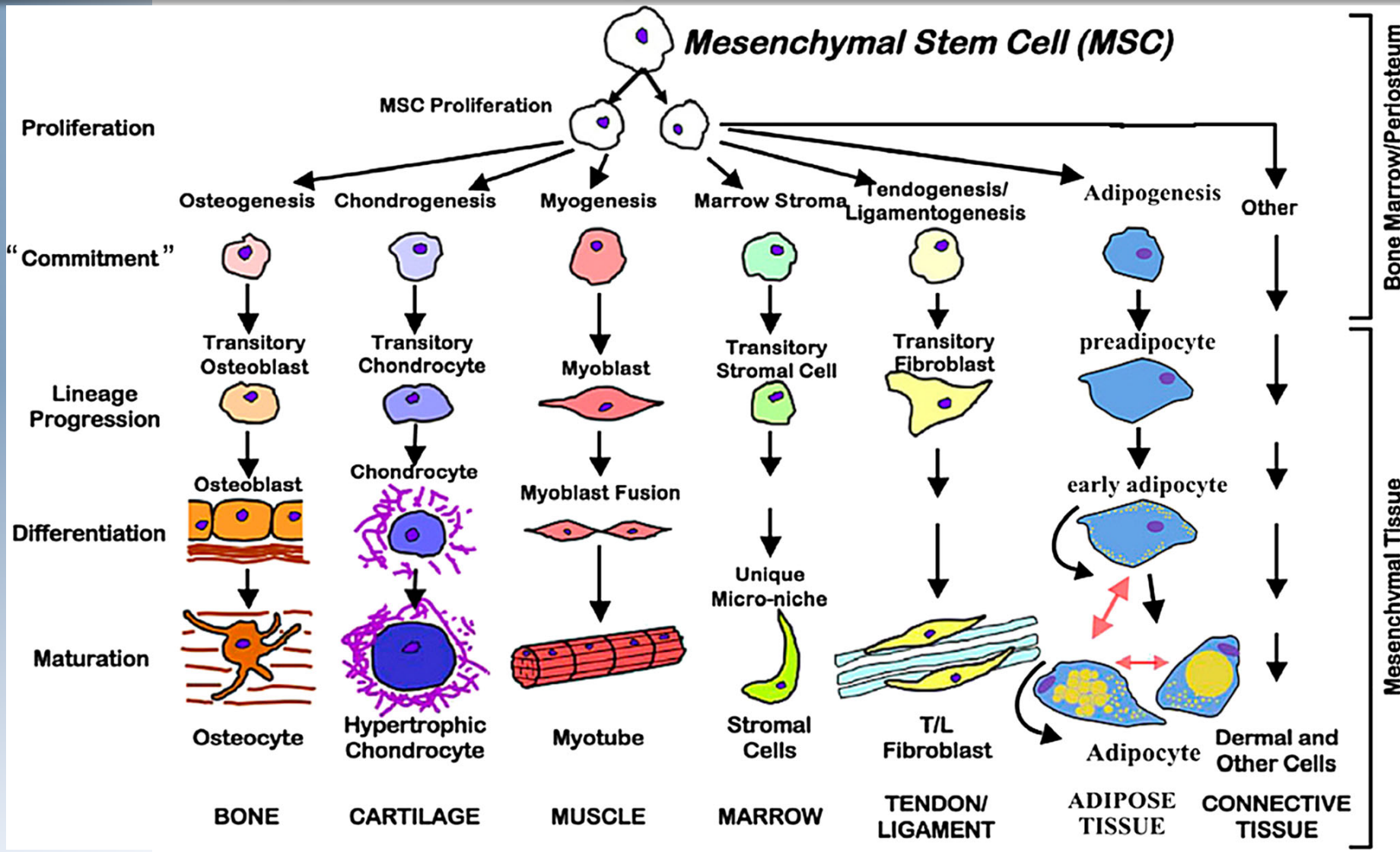
Research interest: Regenerative potential of MSCs



Research interests

- **Stem cells signaling pathways that dictate differentiation into osteogenic lineage**

J Cell Physiol. 2015. Mar 9. [Epub ahead of print]. Impact factor = 3.87

PLoS One. 2014. 9:e103060. Impact factor = 3.534

Growth Factors. 2012. 30:333-343. Impact factor = 3.088

J Orthop Res. 2012. 30:1051-1057. Impact factor = 2.972

- **Stem cells surface markers, their functions, use of stem cells surface markers for selection of osteogenic stem cells**

J Orthop Res. 2015. 33(5):625-632. Impact factor = 2.972

- **Bone tissue engineering using gene therapy and stem cells**

Cell and tissue research. 2015. Accepted for publication. Impact factor = 3.333

Bone Marrow Res. 2013. 2013:737580. Impact factor = Open access journal

Curr Pharm Des. 2013. 19:3374-3383. Impact factor = 3.55

Growth Factors. 2011. 29:36-48. Impact factor = 3.088

Growth Factors. 2010. 28:306-317. Impact factor = 3.088

- **Interaction between immune cells and stem cells during osteogenesis : use of allogeneic stem cells for bone regeneration**

Journal of Immunology Research. 2015. 2015:ID 752510. Impact factor = 2.934

Journal of Immunology Research. 2015. 2015:ID 192415. Impact factor = 2.934

J Orthop Res. 2013. 31:227-234. Impact factor = 2.972

Why study MSCs?

- **Deficiencies associated with fracture non-unions**

•**Defective Mesenchymal stem cells (MSCs):**

-Decreased osteogenesis, increased cell senescence and elevated Dickkopf-1 (Dkk-1) secretion.

-Decreased number in bone marrow, decreased proliferation.

•**Defective Bone morphogenetic proteins (BMPs) signaling in MSCs:**

-Downregulation of gene expression of BMPs-2,3,4,6,7; noggin, germlin, sclerostin, BAMBI in experimental atrophic nonunions in rats.

-Downregulation of gene expression of BMP-7

-Balance between local presence of BMPs and BMP-inhibitors; total absence of pSmads 1/5/8 in cartilage.

•**Defective induction of bone formation by MSCs in presence of immune cells – new concept**

-**T cells inhibit bone formation induced by MSCs**

Why study MSCs? Clinically speaking -

Bone Graft	Structural Strength	Osteo-Conduction	Osteo-Induction	Osteogenesis
Autograft				
Cancellous	No	+++	+++	+++
Cortical	+++	++	++	++
Allograft				
Cancellous				
Frozen	No	++	+	No
Freeze-Dry	No	++	+	No
Cortical				
Frozen	+++	+	No	No
Freeze-Dry	+	+	No	No
Demineralized Allogeneic Cancellous Chips	No	+	++	No

Figure 3: *Comparative properties of bone grafts*

Why study MSCs? Clinically speaking -

A REALITY CHECK

It is estimated that more than 500,000 bone-grafting procedures are performed annually in the United States, with approximately half of these procedures related to spine fusion. These numbers easily double on a global basis and indicate a shortage in the availability of musculoskeletal donor tissue traditionally used in these reconstructions. (Figure 1)

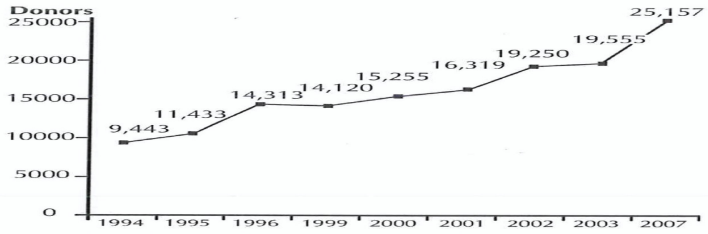


Figure 1: *U.S. trends in musculoskeletal tissue donors. Source: AATB Annual Survey*

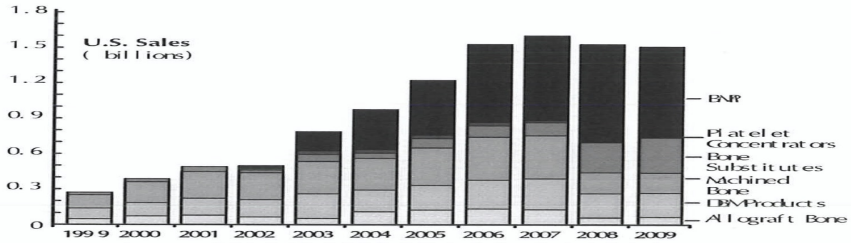


Figure 2: *U.S. sales of bone graft and bone-graft substitutes Source: Orthopedic Network News*

This reality has stimulated a proliferation of corporate interest in supplying what is seen as a growing market in bone replacement materials. (Figure 2) These graft alternatives are subjected to varying degrees of regulatory scrutiny, and thus their true effectiveness in patients may not be known prior to their use by orthopaedic surgeons. It is important to gain insight into this emerging class of bone-graft alternatives.

THE PHYSIOLOGY OF BONE GRAFTING

The biology of bone grafts and their substitutes is appreciated from an understanding of the bone formation processes of Osteogenesis, Osteoinduction and Osteoconduction.

Graft Osteogenesis: The cellular elements within a donor graft, which survive transplantation and synthesize new bone at the recipient site.

Graft Osteoinduction: New bone realized through the active recruitment of host mesenchymal stem cells from the surrounding tissue, which differentiate into bone-forming osteoblasts. This process is facilitated by the presence of growth factors within the graft, principally bone morphogenic proteins (BMPs).

Graft Osteoconduction: The facilitation of a bone healing process into a defined passive trellis structure.

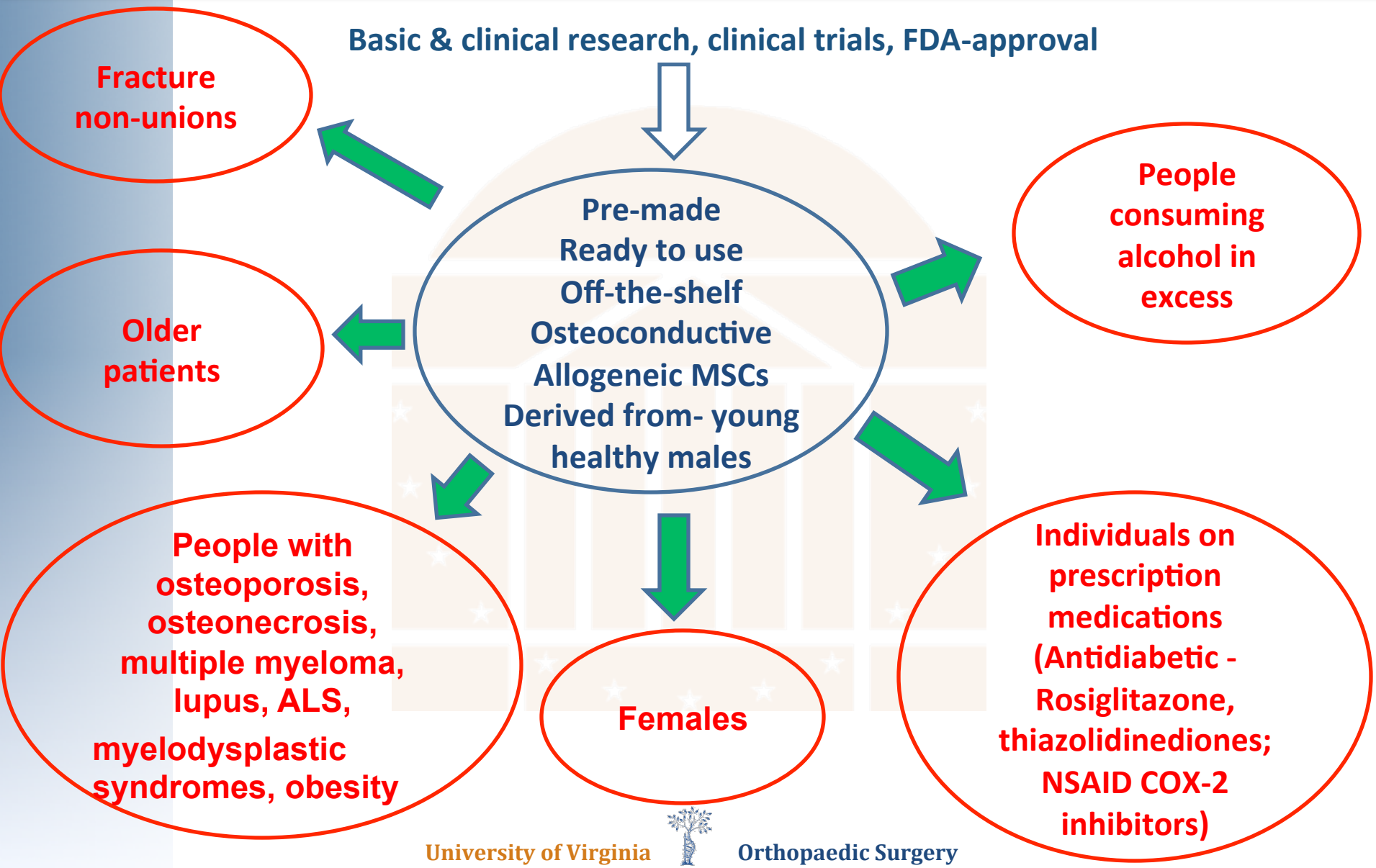
All bone graft and bone-graft substitute materials can be described through these processes.

While fresh autologous graft has the capability of supporting new bone growth by all three means, it may not be necessary for a bone graft replacement to inherently have all three properties in order to be clinically effective. When inductive molecules are locally delivered on a scaffold, mesenchymal stem cells are ultimately attracted to the site and are capable of reproducibly inducing new bone formation, provided minimal concentration and dose thresholds are met. In some clinical studies, osteoinductive agents have been shown to potentially perform equivalent or superiorly to autograft demonstrating efficacy as an autograft replacement.

However, bone marrow aspirate applied to osteoconductive scaffolds are still reliant on the local mechanical and biological signals in order to ultimately form bone. For this reason, these materials are typically used as an adjunct in order to retain efficacy equivalent to autograft.

Similarly, osteoconductive materials work well when filling non-critical size defects that would normally heal easily. However, in more challenging critical size defects, either fresh autologous bone graft or osteoinductive agents appear necessary for healing.

Potential need for allogeneic MSCs



Currently available FDA-approved MSCs-based cellular bone matrices (CBMs) (all data is shown exactly as reported by the individual companies; no extrapolations have been made)

Product	Osteocel Plus	Trinity Evolution	Cellentra VCBM	AlloStem	Ovation
Manufacturer	NuVasive, Inc. (San Diego, CA, USA)	Orthofix (Lewisville, TX, USA)	Biomet (Warsaw, IN, USA)	AlloSource (Centennial, CO, USA)	Osiris Therapeutics, Inc. (Columbia, MD, USA)
Source of MSCs	Cadaveric bone	Cadaveric bone	Cadaveric bone	Cadaveric adipose tissue	Live donor placenta chorion layer
Osteoinductive cytokines	Naturally occurring in bone	Naturally occurring in bone	BMP-2, 4, 7; VEGF; TGF-β; PDGF; IGF-1; FGF	Naturally occurring in bone	BMP-2, 7; PDGF; VEGF; FGF; IGF-1; TGF-β; PIGF
Osteoconductive carrier	Cancellous bone chips	Demineralized bone	Cancellous bone matrix	Demineralized bone	None (product can be added to any carrier)
Price for 1 cc	$460.00	$540.00	$620.00	$540.00	$7,150

Caveats of current MSCs-based CBMs

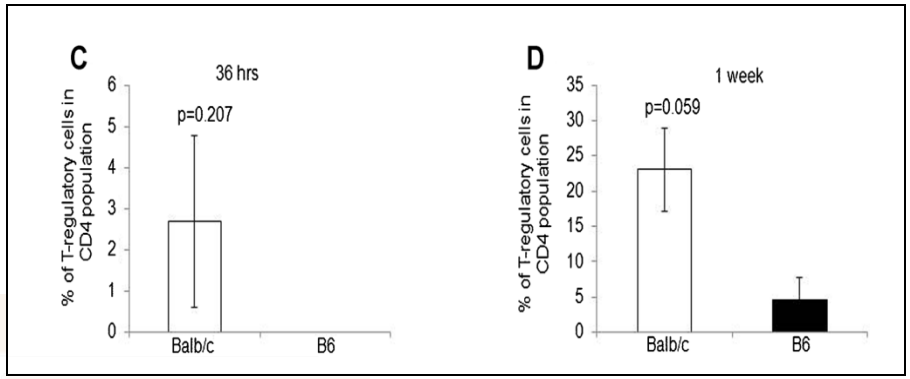
- The manufacturers claimed that efficacy of the product did not depend on MSCs – probably only to avoid FDA premarket application (PMA)

- Since the currently marketed CBMs bypassed FDA premarket review, they did not undergo clinical trials - safety and efficacy is elusive.

- All are allogeneic MSCs-based – effectiveness of allogeneic MSCs is controversial

The most expensive and only CBM using live source received warning from FDA in 2013 that Ovation does not meet 21 CFR Part 1271.10 – the production will stop in 1-2 years based on reports of FDA this year.

Allogeneic MSCs do not work



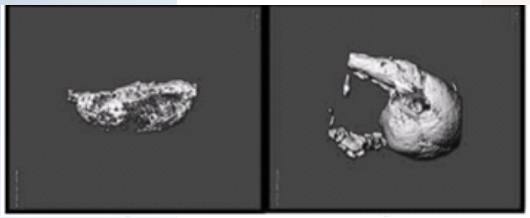
Mesenchymal stem cells

Syngeneic

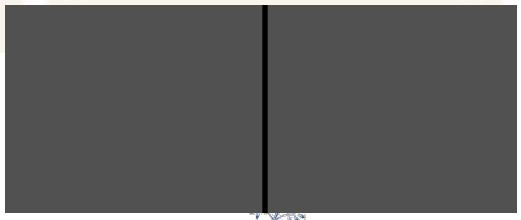
Allogeneic

Allogeneic

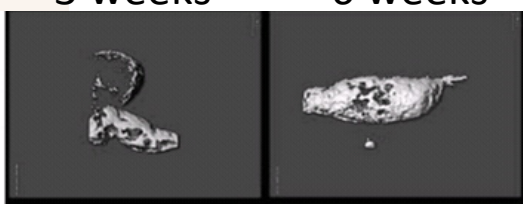
3 weeks 6 weeks



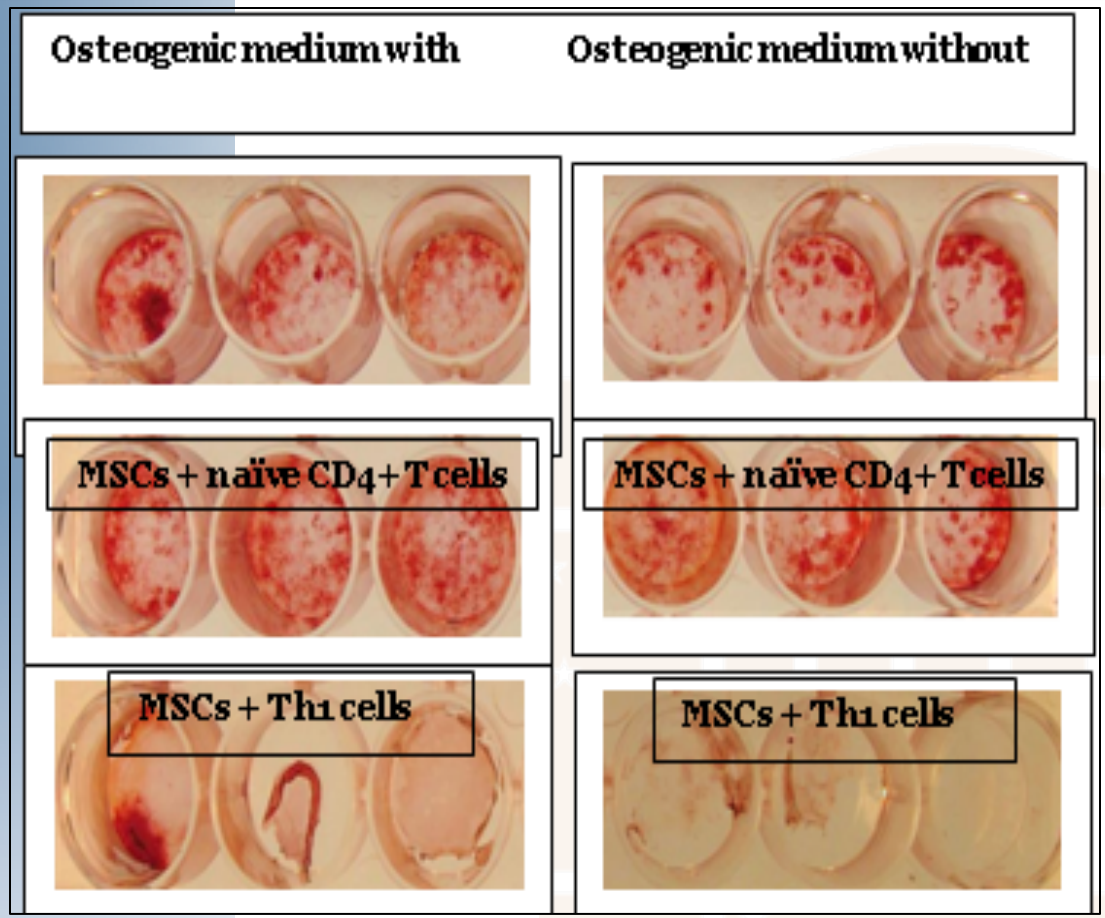
3 weeks 6 weeks

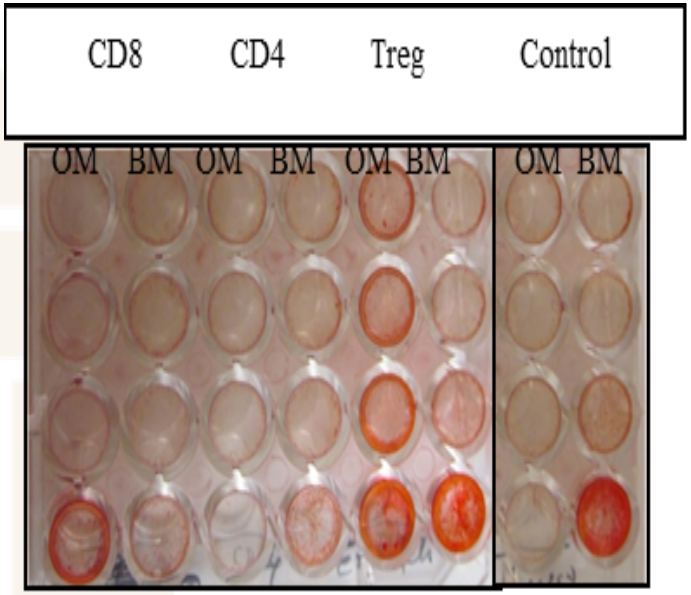


3 weeks 6 weeks



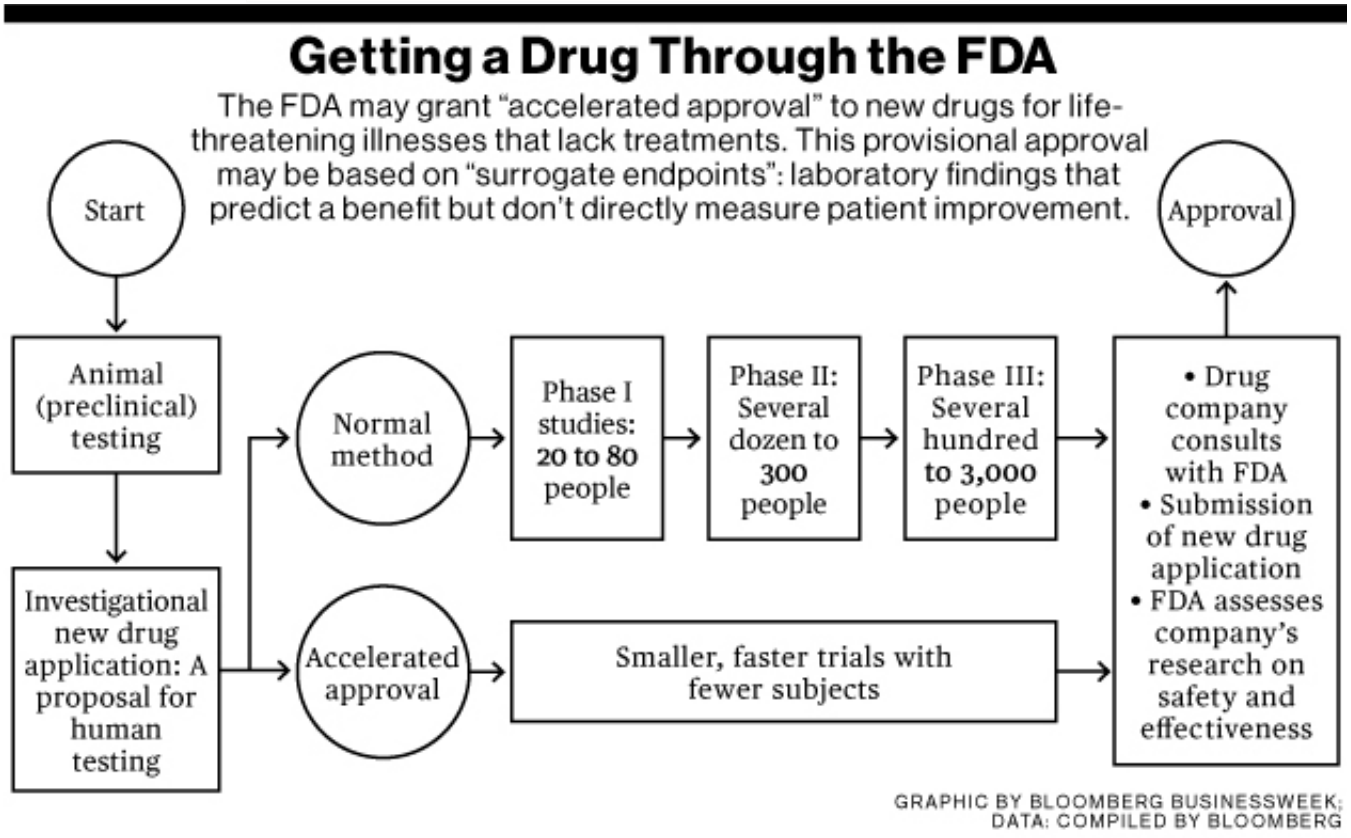
T cells inhibit osteogenesis
Treg cells promote osteogenesis





Goals

- **Our long term goal is to produce an optimum (osteoconductive, osteoinductive, T cells-inhibiting, providing structural strength) deliverable, FDA-approved bone graft substitute for bone regeneration**



Thank You