BIOGRAPHICAL SKETCH

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NAME: Dmitriy Sheyn

eRA COMMONS USER NAME (credential, e.g., agency login): SHEYND3

POSITION TITLE: Project Scientist

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
Hebrew University of Jerusalem, Faculty of Medicine	BSc	2004	Basic Medical Sciences
Hebrew University of Jerusalem, Faculty of Dental Medicine	MSc	2005	Skeletal Tissue Engineering
Hebrew University of Jerusalem, Faculty of Dental Medicine	PhD	2011	Skeletal Tissue Engineering
Cedars-Sinai Medical Centre, Department of Surgery, Regenerative Medicine Institute	Postdoc	2013	Stem Cell-Based Skeletal Regeneration

A. Personal Statement

In the last decade devoted to my research I'm please to say I have achieved several important discoveries and advancements in the field of gene-and-stem cell therapy. This field holds a great therapeutic potential. I have developed an efficient and easily reproducible method to modify stem cells and to regenerate bone defects and generate new bone tissue for spinal fusion. The cell therapy approach was tested and found biomechanically valid and comparable to the native bone. This is of enormous importance since the current clinical practice mostly involves artificial materials that are extremely far from the native tissues by their biomechanical properties, which often causes rejection and failure. Additionally I developed a novel gene delivery method for bone formation and regeneration and currently prospectively validating data that may contradict current thinking about the direct gene delivery strategy. My colleagues and me developed a novel method to deliver genes into injured tissue using ultrasound. The study involves targeting endogenous stem cells and induction of their differentiation. This method is specifically appealing for long bone superficial non-union fractures. In recognition of the importance of this discovery, the California Institute of Regenerative Medicine recently funded a further study with the prestigious Early Translational Award to develop a preclinical model for such treatment for non-union fractures in long bones. The outcome of this study will generate a simple, affordable and safe solution for non-union fractures. In another project we discovered a novel method to generate brown adipose tissue de novo using stem cell and genetic engineering. Brown adipose tissue plays critical role in several physiological processes like thermogenesis, energy expenditure and specifically glucose metabolism. The new way to generate brown adipose tissue opens new horizon for developing new treatments for disorders of the 21st century like obesity and diabetes.

Currently I'm developing new applications for employing induced pluripotent stem cells in the filed of skeletal tissue regeneration, particularly focusing on bone, intervertebral disc and adipose tissues. Applying my previous discoveries on the novel induced pluripotent stem cells will bring the future of personalized medicine to the clinical application much sooner than we ever imagined. Pluripotent stem cells can be derived from skin biopsy and recently even from simple blood sample. But it is not enough to isolate the cells, we need to know how to manipulate them, keep under control, track in vivo and induce their differentiation to the desired tissue. My vision is that when we would translate the knowledge we gained using adult stem cells and embryonic stem

cells over the years and will apply it to the easily available and expandable pluripotent stem cells, we can actually bring the stem cell therapy from the lab to the clinic in almost all kinds of human disorders, including skeletal. To bring this vision to reality I plan to apply all the discoveries I was able to make so far on the novel stem cell population. Development of personalized medicine will enable treat many as of today untreatable conditions, but also will be able to adjust the treatment for every patient reducing costs, side effects and increasing the success rate.

B. Positions and Honors

Positions and Employment

2013- 2015 Project Scientist in Skeletal Regeneration and Stem Cell Therapy Lab, Department of Surgery, Regenerative Medicine Institute, Cedars-Sinai Medical Center, Los Angeles, CA.

2015- present Research Scientist in Skeletal Program, Department of Surgery, BOG Regenerative Medicine Institute, Cedars-Sinai Medical Center, Los Angeles, CA.

Other Experience and Professional Memberships

Tissue Engineering and Regenerative Medicine (TERMIS)
Orthopaedic Research Society (ORS)
North American Spine Society (NASS)
International Society for Stem Cell Research (ISSCR)

Manuscript reviewer:

Molecular Biotechnology
Journal of Biomedical Materials Research: Part A
Tissue Engineering
PLOS One

Honors

2007 – 2010	Rector's Fellowship for outstanding doctoral students
2007 – 2010	Levy Eshkol doctoral fellowship, provided by Israeli Ministry of Science
2007	The Kaye Innovation Award at Hebrew University of Jerusalem
2008	Rusk Foundation Travel Award for excellent graduate students
2008	Best Basic Science Paper – 16th Ann Scientific Mtg of the Int'l Spine Intervention Society
2010 – 2012	Regenerative Medicine Institute fellowship for postdoctoral researcher
2013	Best Oral Presentation Award at Regenerative Medicine Institute Annual Retreat

C. Contribution to Science

1) **The intervertebral disc (IVD)** possesses a minimal capability for self-repair and regeneration. Changes in the differentiation of resident progenitor cells can represent diminished tissue regeneration and a loss of homeostasis. Degenerative intervertebral disc (IVD) disease and associated chronic lower back pain constitute a major health problem with estimated costs in the U.S. of up to $50 billion yearly. Despite decades of research, no fundamental multidisciplinary understanding of the mechanism(s) of IVD degeneration has surfaced and consequently, clinical therapies are still in the earliest stages of development. Recently the main focus of my work and this proposal is on the regeneration of the IVD. I strongly believe that our recent study (a) facilitated a deeper understanding of the IVD degeneration process and triggered further studies that will contribute to development of novel therapies for IVD degeneration. Now the main challenge is to develop new stem cell therapies for IVD and use appropriate animal models that can lead to future clinical practice.
    a) Mizrahi O#, Sheyn D# *(equal contributor),* Tawackoli W, Ben-David S, Su A, Li N, Oh A, Bae H, G, Gazit D and Gazit Z; Nucleus pulposus degeneration alters properties of resident progenitor cells; Spine J. 2013 Apr 9.

2) **Genetically engineered stem cells are a tool for tissue engineering and regenerative medicine**, albeit a tool whose development is fraught with difficulties. Gene-and-cell therapy offers solutions to severe problems faced by modern medicine, but several impediments obstruct the path of such treatments as they

move from the laboratory toward the clinical setting (a). One of the main focuses of my research was to develop gene-and-stem cell therapy for bone tissue related disorders. My colleagues and me have developed an efficient and easily reproducible method to modify stem cells to regenerate bone fractures and generate new bone tissue and repair vertebral compression fractures. Our studies in this area have demonstrated that the adipose-derived stem cells overexpressing osteogenic factors can restore significant defects and injuries in the vertebral body otherwise incurable (b-d). Our data provide the platform for new stem cell-based approaches for treating vertebral fractures especially in osteoporotic patients (d). This study lead to substantial funding from the California Institute of Regenerative Medicine (Early Translational Award II) in which we developed a systemic stem cell therapy for osteoporosis related spine injuries (e).

a) Sheyn D, Mizrahi O, Benjamin S, Pelled G and Gazit D, Genetically modified cells in regenerative medicine and tissue engineering, Advanced Drug Delivery Reviews, 2010 Jun 15;62(7-8):683-98.
b) Sheyn D, Pelled G, Zilberman Y, Talasazan F, Frank JM, Gazit D and Gazit Z, Nonvirally Engineered Porcine Adipose Tissue–Derived Stem Cells: Use in Posterior Spinal Fusion, Stem Cells, 2008; Apr;26(4):1056-64
c) Sheyn D#, Kallai I#, Tawackoli W, Cohn Yakubovich D, Oh A, Su S, Da X, Lavi A, Kimelman-Bleich N, Zilberman Y, Li N, Bae H, Gazit Z, Pelled G, Gazit D. Gene-Modified Adult Stem Cells Regenerate Vertebral Bone Defect in a Rat Model. Mol Pharm. 2011 Oct 3;8(5):1592-601.
d) Mizrahi O#, Sheyn D#, Tawackoli W, Kallai I, Oh A, Su A, Zarrini P, Cook-Wiens G, Gazit D and Gazit Z; BMP-6 is more efficient in bone formation than BMP-2 when overexpressed in mesenchymal stem cells, Gene Therapy, 2012, Jun 21. doi: 10.1038/gt.2012.45.
e) **Sheyn, D.,** Shapiro, G., Tawackoli, W., Jun, D.S., Koh, Y., Su, S., Da, X., Ben-David, S., Bez, M., Yalon, E., Antebi, B., Avalos, P., Stern, T., Zelzer, E., Schwarz, E.M., Gazit, Z., Pelled, G., Bae, H.M., Gazit, D., "PTH induces systemically administered mesenchymal stem cells to home to and regenerate osteoporotic spine injuries" Molecular Therapy (2015)

3) **Stem cell therapy for bone regeneration was tested and found biomechanically valid and comparable to the native bone.** In this part of my research, my colleagues and I have asked the question what is the biomechanical property of the newly formed stem cell-mediated bone tissue. The question is enormously important as current clinical practice mostly involves artificial materials that are extremely far from the native tissues in their biomechanical properties, which often causes rejection and failure. We tested the new bone from two different aspects: the nanobiomechanical properties (a,b) and structural mechanical properties of the new bone (c) and in both aspect the gene-and-cell-engineered bone tissue was found similar in its properties to natively form bone tissue.

   a) Pelled G, Tai T, Sheyn D, Zilberman Y, Kumbar S, Nair LS, Laurencin CT, Gazit D, Ortiz C. Structural and nanoindentation studies of stem cell-based tissue engineered bone. J. of Biomechanics. 2007;40(2):399-411.
   b) Tai K, Pelled G, Sheyn D, Bershteyn A, Kallai I, Zilberman Y, Ortiz C and Gazit D, Nanobiomechanics of Repair Bone Regenerated by Genetically Modified Mesenchymal Stem Cells, 2008, Tissue Eng Part A. 2008 Oct;14(10):1709-20.
   c) Sheyn D, Rüthemann M, Mizrahi O, Kallai I, Zilberman Y, Tawackoli W, Kanim LEA, Zhao L, Bae H, Pelled G, Snedeker JG and Gazit D. Genetically Modified Mesenchymal Stem Cells Induce Mechanically Stable Posterior Spine Fusion. Tissue Engineering Part A, 2010, Dec;16(12):3679-86

4) **Novel non-viral gene delivery methods for bone formation and regeneration.** To enhance the efficiency of non-viral gene delivery, methods have been developed that rely on a short pulse of energy to optimize gene delivery. These methods induce the formation of transient nano-sized pores in the membranes of cells, enabling the uptake of DNA, which leads to cell transfection. After developing a method for ex vivo electroporation-based gene delivery method (a), my colleagues and I developed a novel gene delivery method for bone formation and regeneration and currently prospectively validating data that may contradict current thinking about the direct gene delivery strategy (b, c). In this regard, it is truly astounding that ultrasound based gene delivery system that resulted in in vivo bone formation de novo (b). This method is specifically appealing for long bone superficial non-union fractures. The study involves targeting endogenous stem cells and induction of their differentiation. And in recognition of the importance of this discovery, the California Institute of Regenerative Medicine recently funded a further study with the

prestigious Early Translational Award III to develop a preclinical model for such treatment for non-union fractures in long bones. The outcome of this study will generate a simple, affordable and safe solution for non-union fractures.

a) Aslan H, Zilberman Y, Arbeli V, Sheyn D, Matan Y, Liebergall M, Li JZ, Helm GA, Gazit D, Gazit Z. Nucleofection-based ex vivo nonviral gene delivery to human stem cells as a platform for tissue regeneration. Tissue Engineering. 2006;12(4):877-89.
b) Sheyn D, Kimelman-Bliech N, Pelled G, Zilberman Y, Gazit D and Gazit Z, Ultrasound-based Nonviral Gene Delivery Induces Bone Formation In Vivo, Gene Therapy, 2008, Feb;15(4):257-66
c) Shapiro G, Kallai I, Sheyn D, Tawackoli W, Koh YD, Bae H, Trietel H, Goldbart Riki, Kost J, Gazit Z , Gazit D and Pelled G; Ultrasound-mediated transgene expression in endogenous stem cells recruited to bone injury sites. Polym. Adv. Technol. 2014, 25 525–53; DOI: 10.1002/pat.3297

5) **Oxygen carriers improve stem cell survival and differentiation after implantation**. Although mesenchymal stem cells (MSCs) are known as propitious candidates for cell therapies in regenerative medicine, many of the cells die or lose function when implanted in vivo. One reason for this phenomenon could be the microenvironmental hypoxia at the implantation site, especially in the process of bone formation. It is conceivable that the distance from host blood vessels prevents the transport of oxygen and nutrients, leading to considerable cell death shortly after implantation. We have recently discovered and validated new method to implant cells using oxygenated scaffold and thus increase cell graft survival and differentiation in vivo (a). Little is known about the mechanism by which hypoxia affects osteogenesis and chondrogenesis, and specifically on the direct pathways between hypoxia related genes and osteogenic genes. In another study we have shown that by adding artificial oxygen carriers to hydrogel containing MSCs, and thus increasing the oxygen level available for the cells although for a short term, so we can improve MSC survival and metabolism, and moreover to promote osteogenesis over chondrogenesis (b). Synthetic oxygen carriers succeeded to prevent the hypoxia-related cell death of the implanted cells for several days after the implantation. This has enormous importance for any cell therapy application.

a) Kimelman-Bleich N, Pelled G, Sheyn D, Kallai I, Zilberman Y, Mizrahi O, Tal Y, Tawackoli W, Gazit Z, Gazit D. The use of a synthetic oxygen carrier-enriched hydrogel to enhance mesenchymal stem cell-based bone formation in vivo. Biomaterials 2009; Sep;30(27):4639-48.
b) Benjamin S#, Sheyn D# *(equal contributor),* Ben-David S, Oh A, Pelled G, Gazit D and Gazit Z. Oxygenated environment enhance both, stem cell survival and osteogenic differentiation; Tissue Engineering 2013 Mar;19(5-6):748-58

Complete List of Published Work in MyBibliography:
http://www.ncbi.nlm.nih.gov/sites/myncbi/1BULvGNgCBL/bibliography/41037283/public/?sort=date&direction=ascending

Additional publications:
http://onlinelibrary.wiley.com/doi/10.1002/pat.3297/abstract

Books chapters:

1. **Sheyn D**, Pelled G, Gazit D. “Nanotechnologies in Adult Stem Cell Research”, in Nanotechnology in Biology and Medicine: Methods, Devices, and Applications, Vo-Dinh T. Ed. 2007, CRC Press LLC.
2. Gazit Z, Pelled G, **Sheyn D**, Kimelman-Bleich N, Gazit D. Mesenchymal stem cells. Chapter 26 in “Regenerative Medicine and Tissue Engineering” textbook, Second edition, 2010.
3. Kimelman N, Kallai I, **Sheyn D**, Tawackoli W, Gazit Z, Pelled G, Gazit D. Real-time bioluminescence functional imaging for monitoring tissue formation and regeneration. *Methods Mol Biol*. 2013;1048:181-93. doi: 10.1007/978-1-62703-556-9_14.

D. Research Support

Ongoing Research Support

CIRM TR4-06713 Gazit, D. (PI) 12/2013 – 12/2016
Gene Targeting to Endogenous Stem Cells for Segmental Bone Fracture Healing
The objective of the proposed research is to achieve a development candidate consisting of ultrasound-mediated gene targeting to endogenous MSCs for bone repair. Milestones are to determine a protocol for ultrasound-mediated gene delivery to a micropig segmental bone defect, to achieve preclinical proof of concept using defined protocol of ultrasound-mediated BMP gene delivery to endogenous stem cells, to show reproducibility of segmental bone defect repair using ultrasound-mediated delivery of a GMP compatible plasmid and finally develop a clinical plan.
Role: Co-Investigator

NASS Young investigator award Juliane Gleaser (PI) 01/2016-12/2016
Treatment with the NFκB inhibitor NEMO-binding domain peptide to inhibit disc degeneration
Purpose: to investigate the efficacy of an NBD peptide treatment to reduce IVD degeneration by inhibiting cytokine/metalloprotease levels under detrimental mechanical conditions in vitro and in full biological complexity in vivo.
Role: Co-Investigator

Completed Research Support

Cedars-Sinai Translational Research Pipeline Gazit, Z. (PI) 06/2013 – 12/2014
A Combined Treatment of Gene Modified Stem Cells and Oxygenated Scaffolds for Scaphoid Nonunion with Avascular Necrosis
The overall objectives focused on scaphoid nonunion to develop a tractable, clinically relevant model to augment the limited blood supply.
Role: Co-Investigator

CIRM TR2-01780 Gazit, D. (PI) 03/2011- 02/2014
Systemic Adult Stem Cell Therapy for Osteoporosis-Related Vertebral Compression Fractures.
The goals of the project were to develop a stem cell-based therapy for osteoporotic vertebral fractures.
Role: Project Scientist.

NIH R01DE19902 Gazit, D. (PI) 6/1/09- 5/31/14
PTH Effects on Craniofacial Allografting.
The goal of the project was to define the effects of PTH on bone healing using allografts with specific emphasis on scar tissue formation and inflammation.
Role: Project Scientist.

NIH R01DE19902 S1 Gazit, D. (PI) 8/1/12- 5/31/14
PTH Effects on Craniofacial Allografting.
The aim was to integrate state-of-the-art functional imaging technologies in our studies in order to promote novel insights on the effect of parathyroid hormone (PTH) on craniofacial allografts.
Role: Project Scientist.