Research Briefs

Following are highlights from the lead faculty investigators of the original research articles that appeared in journals with the four highest impact factors among those represented by Penn Dental Medicine faculty scholarly activity over the past year.

FOXO1 PROMOTES WOUND HEALING THROUGH THE UP-REGULATION OF TGF-B1 AND PREVENTION OF OXIDATIVE STRESS

***Journal of Cell Biology* (10.8 Impact Factor)**

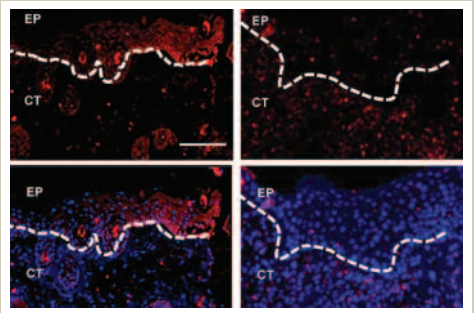
Ponugoti, B., Xu, F., Zhang, C., Tian, C., Pacios, S., Graves, D.T.

What were the key objectives of this study?

The events in wound healing are carefully orchestrated. The concept that we investigated was that a transcription factor would coordinate the regulation of events that are needed in order for wound healing to occur properly. Based on our previous work, we focused on the transcription factor FOXO1. Transcription factors are important in wound healing because they control the expression of other genes, which are needed for healing to occur.

What were some of the important techniques used in the study?

The most important aspect of the study from a technical standpoint was the deletion of FOXO1 in a single type of cell in vivo. These studies used a genetically modified mouse in which the FOXO1 transcription factor was deleted only in keratinocytes, an epithelial cell type that lines mucosal and skin surfaces. All of the other cell types were normal, allowing us to focus on the impact of deleting FOXO1 only in keratinocytes.



ABOVE: Numbers of migrating cells, shown in red, are greatly reduced when the molecule FOX01 is deleted (right column).

What results were most surprising or of particular clinical relevance?

When FOXO1 was deleted in keratinocytes normal wound healing was significantly delayed. We found that this was largely due to FOXO1 regulation of two very important aspects of wound healing. The first was that FOXO1 was needed for keratinocytes to migrate. The second was that FOXO1 protected the cells during wound healing from oxidative stress. When FOXO1 was deleted, keratinocytes did not migrate as well and suffered damage from oxidative stress.

What conclusions or other applications could the findings lead to?

The simple answer would be that increasing FOXO1 should help wound healing based on our result that FOXO1 is needed for normal healing to occur. However, it is more complicated than this since high levels of FOXO1 are just as detrimental as not having enough FOXO1.

IL-17-MEDIATED M1/M2 MACROPHAGE ALTERATION CONTRIBUTES TO PATHOGENESIS OF BISPHOSPHONATE-RELATED OSTEONECROSIS OF THE JAWS

***Clinical Cancer Research* (7.8 Impact Factor)**

Zhang Q, Atsuta I, Liu S, Chen C, Shi S, Shi S, Le AD.

What were the key objectives of this study?

Osteonecrosis of the jaw (ONJ) is a condition involving the necrosis of the jaw bone that is associated with certain anti-resorptive drugs to treat cancer or prevent bone loss in osteoporosis. The objective of this study is to explore the potential role of interleukin 17 (IL-17)-mediated regulation of macrophage function, specifically the subtype M1/M2 macrophage, in the pathogenesis of bisphosphonate-related osteonecrosis of the jaw (BRONJ).

What were some of the important techniques used in the study?

In this study, we developed a BRONJ-disease model in multiple myeloma (MM)-burdened mice. The important techniques include therapeutic approaches using adoptive transfer of ex vivo expanded M2 macrophages, or pharmacological blockage of IL-17 activity to treat BRONJ.

What results were most surprising or of particular clinical relevance?

This work reports that increased Th17 cells and IL-17 cytokine levels correlate with an increase in M1/M2 macrophages ratio at the non-healing extraction sockets of both BRONJ patients and murine models; adoptive transfer of ex vivo expanded M2 macrophages could reverse systemic increase of IL-17 and ONJ severity, and blocking IL-17 activity significantly decreased M1/M2 ratio and concomitantly suppressed BRONJ condition in mice. These findings have provided compelling evidence that IL-17-mediated M1/M2 macrophage alteration plays a critical role in the pathophysiology of BRONJ, a promising breakthrough for identifying novel biomarkers and therapeutic targets for this debilitating and painful disease.

What conclusions or other applications could the findings lead to?

The study concludes that IL-17-mediated M1/M2 macrophage alteration induced by zoledronate may be beneficial for cancer therapy, but might have contributed to an increased susceptibility to BRONJ development.

LOCALLY DELIVERED SALICYLIC ACID FROM A POLY(ANHYDRIDE-ESTER): IMPACT ON DIABETIC BONE REGENERATION

***Journal of Controlled Release* (7.6 Impact Factor)**

Wada, K., Yu, W., Elazizi, M., Barakat, S., Ouimet, M.A., Rosario-Meléndez, R., Fiorellini, J.P., Graves, D.T., Uhrich, K.E.

What were the key objectives of this study?

Diabetes mellitus (DM) involves metabolic changes that can impair bone repair, including a prolonged inflammatory response. A salicylic acid-based poly(anhydride-ester) (SA-PAE) provides controlled and sustained release of salicylic acid (SA) that locally resolves inflammation. This study investigates the effect of polymer-controlled SA release on bone regeneration in diabetic rats where enhanced inflammation is expected.

What were some of the important techniques used in the study?

This study is the first time that an SA-PAE has been applied to diabetic animals for bone regeneration purposes. The difficult part in this project was the surgical procedure and postoperative management. Because of the diabetic animals, the grafting procedure needed to be minimally invasive and postoperative monitoring has a significant impact on wound healing process.

What results were most surprising or of particular clinical relevance?

We found that treatment with SA-PAE enhances bone regeneration in diabetic rats. Plus, it can accelerate bone regeneration in normoglycemic (non-diabetic) animals. It could be possible for this polymer to combine with a bone graft in order to achieve more predictable bone formation for oral and maxillofacial reconstruction purposes.

What conclusions or other applications could the findings lead to?

The advantages of localized, controlled, and sustained SA release, our polymer system enables the incorporation of other bioactives (such as insulin) to further improve bone regeneration.

ORAL DELIVERY OF BIOENCAPSULATED PROTEINS ACROSS BLOOD-BRAIN AND BLOOD-RETINAL BARRIERS

***Molecular Therapy* (7.0 Impact Factor)**

Westerveld, D.R., Ayache, A.C., Verma, A., Shil, P., Prasad, T., Zhu, P., Chan, S.L., Li, Q., Daniell, H.

What were the key objectives of this study?

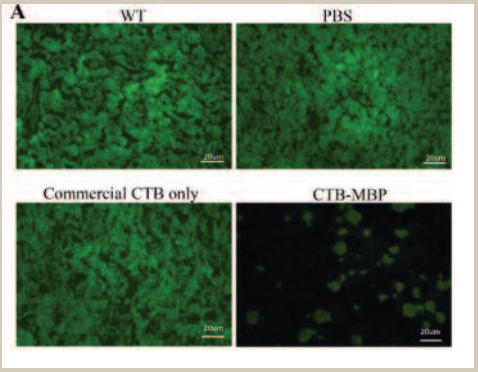
The goal of this study is to develop a low cost, orally deliverable drug for long-term treatment of Alzheimer's patients at home.

What were some of the important techniques used in the study?

The most important concept is bio-encapsulation of therapeutic proteins. Plant cells protect protein drugs from acids and enzymes in the digestive system. However, when plant cells carrying therapeutic proteins reach the gut intact, they are broken down by microbes colonizing the gut, thereby releasing the drugs in the gut for rapid absorption into the blood circulatory system.

What results were most surprising or of particular clinical relevance?

The most surprising observation is our ability to deliver therapeutic proteins across the blood-brain barrier, which tightly regulates the transport of molecules to the brain, blocking bio-therapeutics from reaching their site of action. Alzheimer's disease results in accumulation of the Beta amyloid plaques in the brain. Myelin basic protein (MBP) inhibits amyloid fibril formation through binding and degradation of amyloid by intrinsic protease activity. Oral delivery of MBP fused to a transmucosal carrier (CTB) that facilitates delivery of proteins to the circulatory system from the gut degraded amyloid plaques in post-mortem human Alzheimer's disease brains and in Alzheimer's mouse model. Another surprising finding was the observation of amyloid plaques in the retina and their clearance after oral delivery of CTB-MBP. This raises an interesting question —whether dementia and decrease in vision could both contribute to early symptoms of Alzheimer's disease (especially the inability to recognize close relatives).



ABOVE: Compared to controls, Dr. Daniell's strategy (lower right) reduced plaques in the brain tissue of Alzheimer's patients.

What conclusions or other applications could the findings lead to?

Alzheimer's disease is currently incurable. Right now, treatment is limited to management of symptoms and palliative care. This invention opens the possibility to treat Alzheimer's disease before or after early onset by elimination of plaques through oral delivery of CTB-MBP. This also opens the possibility of delivering drugs to the retina across the blood-retinal barrier.