

Beta Adrenergic Receptor (β AR) Signaling as a novel target for optimizing skin wound healing

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Partnering goals (summary)

- To utilize our full spectrum panel of wound healing assays for development of novel therapies for chronic wounds (diabetic foot ulcers, pressure sores) based on inhibition of β 2AR-pathway.
- Proof of concept clinical trial for venous stasis ulcers using an FDA approved β AR antagonist (i.e. Timolol®).

Introduction/Business Opportunity

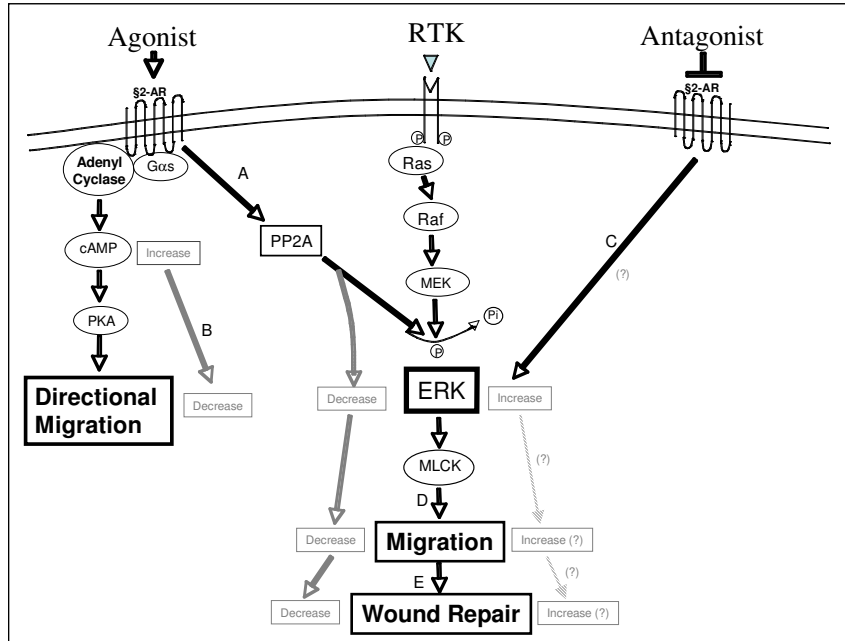
Chronic skin ulcers (venous stasis, diabetic foot and pressure ulcers being most common) present an immense burden to patients, payers, and society. Venous stasis ulcers, which account for 80-90% of all leg ulcers, affect 0.5-1.0% of men and 1.0 to 1.5% of women with increasing incidence in people over 60. The U.S. market is projected to consist of at least 700,000 patients annually, with the cost of therapy ranging from \$20K to \$30K per patient. It has also been estimated that in the United States, nearly a billion dollars are spent annually treating venous ulcers. The current standard of care for venous ulcers is compression dressing and leg elevation, and adjunctive therapies such as pentoxifylline, vein surgery and skin grafting may also improve outcome. Upon rigorous examination of the clinical evidence, multiple other dressing types, including alginates, silver impregnated dressings, collagen dressings and synthetic skin substitutes, have not demonstrated significant improvement in healing. It is not understood why chronic wounds do not heal—a persistent inflammatory component is thought to play a role, as well as aberrant intracellular signaling. This area is being actively investigated.

Keratinocyte migration is a key component in wound healing – it is required for skin re-epithelization and complete wound repair. No matter how well the dermis is repaired, without a durable epithelium, the skin cannot be considered to be “healed.” Thus, numerous laboratories have focused their efforts on improving keratinocyte migration and wound epithelialization with the ultimate goal of more rapid wound healing *in vivo*. We concentrated on the role of one of several classes of beta-adrenergic receptors, the beta 2 adrenergic receptor (b2AR) in this process, expressed primarily on human keratinocytes. Our work demonstrates that blocking of b2AR pathway using pharmacological antagonists (i.e. “beta-blockers”) promotes keratinocyte migration and re-epithelization *in vitro*, in human skin, in an *ex-vivo* skin wound model, and *in vivo* in animal preclinical studies.

Core Technology

The β_2 AR is a G-protein coupled receptor. β_2 AR activation results in signaling through different cellular pathways depending on the cell type. Activation of β_2 AR on keratinocytes affects several cAMP-dependent and -independent pathways that modulate the process of migration. Polymorphisms in β_2 AR may be also important in modifying the pharmacologic response to β AR-active drugs.

Our previous work demonstrated that activation β_2 AR on keratinocytes using a receptor agonist (i.e. isoproterenol, approved for use in bronchospasm, cardiac arrest or heart block) significantly inhibits their migration, and affects the rate of wound healing *in vitro* and *in vivo*. Catecholamines, such as epinephrine and norepinephrine, are endogenously generated agonists, and these are also generated by the wound epithelium. Therefore it is possible that one pathogenic mechanism contributing to non-healing skin wounds is the chronic activation of β_2 AR by endogenously generated catecholamines.



Isoproterenol inhibited migration of cultured human keratinocytes isolated from neonatal foreskins. This inhibition was dose-dependent and was reversed by the addition of timolol, a beta blocker (FDA approved, and currently in use primarily as an anti-glaucoma drug). The results were confirmed by the "scratch wound assay", where a plate of confluent keratinocytes are scratched to make a wound, and the migration of cells are monitored by videomicroscopy and imaging. Similarly, addition of isoproterenol inhibited healing of human skin cultured *ex-vivo*, and healing is improved by adding the beta blocker. Finally, we have confirmed this in two *in vivo* animal studies- in an acute excisional wound model and a more chronic full thickness burn wound model. In both *in vivo* models, the treatment of the wound, with either topical or systemic beta blockers, improves healing, and results in more rapid wound epithelialization.

Additional pre-clinical tests could be done using a chronic wound model which is currently being implemented in collaboration with Dr. Kevin Grayson, DVM, PhD, Director, Clinical Investigation Facility, David Grant Medical Center, Travis Air Force Base, . The proposed model would more closely resemble the non healing wounds of venous stasis. An excision, made on the back of the animal's ear down to cartilage, is then further impaired by tying its feeder arteries. This chronic wound model is used widely in the preclinical testing of wound healing therapies.

Proposed Research Collaboration

We propose a research collaboration to identify novel b2AR antagonists utilizing our HTS assays and animal models. We envision several options:

1. De novo screening using our own or partner's compound libraries
2. Repositioning of one of the existing beta blockers, such as Timolol®
3. Generation of a new composition of matter based on chemical modifications of the existing b2AR antagonists

Proof of concept clinical study

Clinical validation of the efficacy of beta 2 adrenergic receptor antagonists in wound healing could be obtained from a clinical study with an existing beta blocker, such as Timolol®. The clinical study on patients with non-healing venous ulcers has already been approved by our local VA Northern California Health Systems IRB. Patients would be recruited from the VA Wound Healing Clinic, of which Dr. Isseroff is the director.

Clinical trial experience

Dr. Isseroff has been the site Principal Investigator of over 10 sponsored clinical trials. Most recently she has enrolled over 25 patients in a one-year period in a venous ulcer, industry sponsored trial. Dr. Isseroff is a full time faculty at UCD department of Dermatology, and in addition holds a 50% appointment at the VA, where she is a staff physician, Director of the Wound Clinic, and a VA Merit Award funded basic researcher in wound healing.

Patient Profile

The VA multispecialty Wound Clinic (housed within the Dermatology Section) sees most of the venous, diabetic and pressure ulcer patients enrolled in the VA Medical Center Northern California Health System. That translates to more than 50 patients with the diagnosis of venous ulcers seen in the clinic each year.

Intellectual Property

"Beta adrenergic Receptor Agonists and antagonists and modulation of wound healing", [WO 06/108176](#) published Dec 14, 2006

"In vitro wound healing assay and device "WO 2006/010161", filed July 2004

Selected Publications

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