Multiomic Tools to Characterize the Mechanism of Action (MOA) of Novel Therapies for Healing Chronic Wounds

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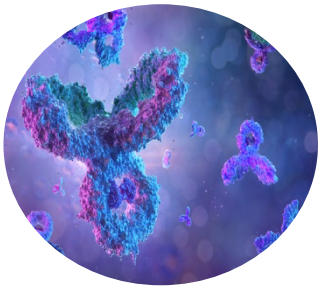
In the path to defining the mechanism of action (MOA) of advanced wound care products, it is beneficial to use the best model system and a variety of complementary multiomic approaches (proteomics, genomics, and lipidomics) to better understand the gene expression, proteins, and lipid composition that define the parameters for wound healing. Our model system of choice is the diabetic (DB) mouse from Jackson Laboratories (db/db BKS.Cg-Dock7m +/+ Lepr db/J). The DB mouse is a delayed healing model which tightly mimics the comorbidities of patients with diabetes. Porcine models have their purpose for biocompatibility studies and complex tissue injuries such as burns, but they are not as robust for MOA studies. With the advent of more sensitive technologies, we are now capable of supplying these services for clinical trials with CAMPs (cellular, acellular, matrix-like products), biologics, devices, and novel therapeutics.

In terms of tools of our trade, we use a battery of approaches to understand the dynamic interplay of molecular signals that stimulate the stages of wound healing. For mRNA **gene expression** studies our method of choice is to use NanoString with a custom code set that covers all the stages of wound healing, macrophage plasticity, fibrosis, and scarring. More recently we spiked our custom code set with additional target probes to include nitric oxide and oxygen responsive genes. Due to the reproducibility and robustness of NanoString, we have been able generate publication quality data with profound statistical significance.



For **proteomics**, we have refined methods to examine the protein expression level of over two thousand proteins from healed wound tissue. We utilize Jump SAS and multivariate analysis to define how a novel therapy compares to the standard of care (SOC) and untreated negative controls. From these studies we have been able to ascertain which biomarkers are critical to healing chronic wounds. In addition, we can identify differences in competitor products (1,2).

To better understand how to prevent wound recurrence, we use two complementary approaches to study the tissue integrity of healed wounds and skin barrier function. After the wounds have healed in the animal models, we take the healed tissue for **biomechanical testing** (ADMET, eXpert 7600) using validated ASTM and ISO methods to study parameters such as elasticity and tensile strength.

lipotype
Lipidomics for a better life

To have a more profound understanding of skin barrier function we have recently started to delve into **lipidomics** (3,4). We are collaborating with Lipotype to better understand how the lipid composition of a wound and surrounding skin can predict wound healing, and potentially prevent wound recurrence. With over a

decade of expertise in mass spectrometry-based lipidomics, Lipotype empowers researchers to identify and quantify lipids in diverse biological samples including skin, plasma and tissues. From skin barrier-specific ceramides to pro- and anti-inflammatory oxylipins, their cutting-edge technology delivers deep insights for biomarker discovery, drug development, and a comprehensive understanding of lipid metabolism in health and disease.

References

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[3] Sadowski, T et al. (2017). Large-scale human skin lipidomics by quantitative, high-throughput shotgun mass spectrometry. Scientific Reports, 7. https://doi.org/10.1038/srep43761

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