

SKIN BIOLOGY & DISEASES

RESOURCE-BASED CENTER



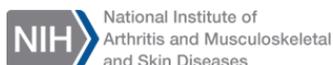
2021 Annual Symposium

Thursday, March 4, 2021

Michigan Medical, Ann Arbor, MI

Event Program

Our annual symposium is sponsored by:



UNIVERSITY OF MICHIGAN
DEPARTMENT OF DERMATOLOGY

2021 UM SKIN BIOLOGY & DISEASES RESOURCED-BASED CENTER ANNUAL SYMPOSIUM

March 4, 2021

The University of Michigan

Virtual on Zoom

8:00 am – 12 noon

8:00 - 8:20 am Welcome & UM-SBDRC Overview

Research Updates: UM-SBDRC PILOT AWARDS

8:20 – 8:30 am ***Single-Cell RNA-Seq analysis of the effect of NETs in TH17 polarization***
James T. Elder, M.D., Ph.D. (Principal Investigator)

8:30 – 8:40 am ***Dissecting the role of CD13/Aminopeptidase N in scleroderma pathogenesis***
Eliza Pei-Suen Tsou, Ph.D. (Principal Investigator); David A. Fox, M.D. (Co-Investigator); Dinesh Khanna, M.B.B.S. (Co-Investigator); M. Asif Amin, M.B.B.S. (Co-Investigator)

8:40 - 8:50 am ***Elucidating the cellular and molecular role of PIXT1 in skin inflammation and UVB-mediated keratinocyte apoptosis***
Michelle Kahlenberg, M.D., Ph.D. (Principal Investigator)

8:50 - 9:00 am ***Role of each member of the IL-36 cytokines in keratinocyte immune responses***
Mrinal K. Sarkar, Ph.D. (Principal Investigator)

9:00 - 9:10 am ***Live imaging of neutrophil infiltration in stressed skin in real time in vivo***
Carole Parent, Ph.D. (Principal Investigator); Pierre Coulombe, Ph.D. (Co-Investigator)

9:10 - 9:20 am ***Investigating the role of VGLL3 in antiviral immune responses***
Allison Billi, M.D., Ph.D. (Principal Investigator); Olesya Plazyo, Ph.D. (Post Doctorate Co-Investigator)

9:20 - 9:30 am ***The role of Yap/Taz in dermal extracellular matrix homeostasis and aging***
Gary Fisher, Ph.D. (Principal Investigator); Taihao Quan, M.D., Ph.D. (Co-Investigator)

Research Updates: UM-SBDRG INNOVATION AWARDS

9:30 - 9:35 am ***Transcriptomic profiling for nascent RNA to identify distinct mediators of early inflammatory response in keratinocytes***

Matthew T. Patrick, Ph.D. (Principal Investigator); Lam C. Tsoi, Ph.D. (Co-Investigator)

9:35 - 9:40 am ***Prediction of regulatory elements related to Th17 differentiation in skin-homing T Cells***

James T. Elder, M.D., Ph.D. (Principal Investigator); Zhaolin Zhang, Ph.D. (Post Doctorate Co-Investigator)

9:40 - 9:45 am ***Targeting CD200 in a Mouse Model of Basal Cell Carcinoma***

Sunny Wong, Ph.D. (Principal Investigator)

9:45 - 10:45 am Break & Poster Viewing/Nominations

10:45 - 11:45 am Keynote Address

**Richard Gallo, M.D., Ph.D., Professor & Chairman of the Dermatology Department
University of California San Diego School of Medicine**

"Lessons from the Skin Microbiome in Innate Immunity and Human Disease"

11:45 - 11:50 am Present & Future Collaborations

11:50 - 12 noon Closing Remarks & Poster Awards

ABSTRACTS FOR POSTER PRESENTATIONS

The gut microbe-derived metabolite trimethylamine N-oxide activates PERK to drive mesenchymal cell differentiation and fibrosis

Seok-Jo Kim¹, Qianqian Wan¹, Adam Gordon², Johann E. Gudjonsson³, Stanley L. Hazen^{4,5,6}, Karen J. Ho⁷, John Varga^{1*}

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Abstract

Systemic sclerosis (SSc) is characterized by synchronous vascular injury and fibrosis in multiple organs. While gut dysbiosis is prominent in patients with SSc, it remains unknown if and how it contributes to these pathogenic processes. Trimethylamine (TMA) generated from dietary choline by the gut microbiome is converted by Flavin-containing monooxygenase (FMO3) to trimethylamine N-oxide (TMAO). TMAO is increased in patients with chronic cardiovascular, neurodegenerative and renal disease, and contributes to pathogenesis. We now show that TMAO triggers myofibroblast reprogramming in skin fibroblasts and vascular endothelial cells. These cellular processes are mediated via the novel TMAO receptor activated protein kinase R-like endoplasmic reticulum kinase (PERK). Genetic variants of the highly pleomorphic FMO3 showed strong association with SSc. Remarkably, FMO3, which is principally expressed in the liver, was found to be present and inducible by TGF- β in both isolated skin fibroblasts and in skin explant (GSE109350), and its expression was elevated in skin biopsies from patients with SSc. Elevated FMO3 transcriptome datasets showed in SSc skin biopsies compared to healthy controls (1.69-fold: GSE95065, 1.78-fold: GSE130955, 2.71: GSE47162). Thus we uncover a novel mechanism linking TMAO generation by the gut microbe and vasculopathy and fibrosis in SSc involving stromal cell reprogramming. The TMAO meta-organismal pathway may therefore be targeted for SSc therapy.

Poster #2

A20 and its repressor DREAM expression govern susceptibility to fibrosis in systemic sclerosis (SSc)

Wenxia Wang¹, Swarna Bale^{1,2}, Swati Bhattacharyya^{1,2}, John Varga^{1,2},

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In addition to autoimmune and inflammatory diseases, variants of TNFAIP3 encoding the ubiquitin-editing enzyme A20 are also associated with fibrosis in systemic sclerosis (SSc). However, it remains unclear how genetic factors contribute to SSc pathogenesis, and which cell types drive disease due to SSc-specific genetic alterations. We therefore characterized the expression, function and role of A20, and its negative transcriptional regulator DREAM, in SSc and disease models. Using unbiased transcriptome analysis of skin and lung biopsies from SSc patients, we found significantly decreased A20 levels, and robust anti-correlation with fibrotic TGF- β signaling ($r=-0.84$, $p<0.005$). In contrast, the negative regulator of A20 DREAM was significantly elevated in SSc biopsies and anti-correlated with A20 ($r=-0.41$). In human skin and lung fibroblasts and adipose-derived stem cells (ADSC), and mouse preadipocytes, A20 potently inhibited both profibrotic gene expression and myofibroblast transition via blocking multiple SSc-relevant pathways including canonical and non-canonical TGF- β . Mice haploinsufficient for A20, or harboring fibroblasts-specific A20 deletion, recapitulated major pathological and genomic features of SSc, whereas DREAM-null mice with elevated A20 expression were protected. In DREAM-null fibroblasts TGF- β induced A20 expression, while in wildtype fibroblasts it had the opposite effect. Moreover, fibrotic TGF- β responses showed DREAM dependence. An anti-fibrotic small molecule targeting the adiponectin receptors stimulated A20 expression *in vitro* in wildtype but not A20-deficient fibroblasts, and in bleomycin-treated mice. Thus, A20 has a novel cell-intrinsic function in negative regulation of fibroblast activation, and together with DREAM, constitutes a critical regulatory network governing the fibrotic process in SSc. A20 and DREAM represent novel druggable targets for fibrosis therapy.

Poster #3

Name: Shannon Estadt

Title: Graduate Student

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Interferon alpha promotes extrinsic apoptosis of keratinocytes following exposure to ultraviolet B radiation.

Shannon N. Estadt, Mehrnaz Gharaee-Kermani, Bin Xu, Tyson M. Moore, J. Michelle Kahlenberg

Systemic lupus erythematosus (SLE) is a chronic autoimmune disease characterized by sensitivity to ultraviolet radiation (UVR). UVR triggers cutaneous and systemic disease flares, yet mechanisms driving this are not fully characterized. Type I interferons (IFNs) are highly expressed in non-lesional SLE skin and promote increased death of SLE keratinocytes (KCs) after UVR, but the manner in which they do so is currently unknown. This study explores activation and regulation of cell death pathways in KCs exposed to type I IFNs and UVR. We treated immortalized humans KCs (N/TERTs) overnight with IFN α prior to UVB exposure. Four hours post UVB, IFN α priming significantly increased the percentage of cleaved-caspase-3⁺ (CC3⁺) and Annexin V⁺propidium iodide⁻ (AV⁺PI⁻) cells compared to treatment with UVB alone. Further, IFN α promoted apoptosis at a lower dose of UVB. Inhibiting RIPK1, RIPK3, or caspase-1 had no effect on death suggesting this enhanced death does not involve necroptosis or pyroptosis. Use of a pan-caspase inhibitor or a caspase-8 inhibitor significantly reduced the percentage of AV⁺PI⁻ cells following IFN α and UVB treatment while caspase-9 inhibition did not, suggesting enhanced activation of the extrinsic apoptosis pathway. RNA-sequencing identified pro-apoptotic genes *XAF1* and *TNFSF10* as highly upregulated by IFN α treatment, and more so in SLE compared to healthy control KCs. Knockdown of XAF1 in N/TERTs did not abrogate the IFN α -induced increase in UVB-driven apoptosis, suggesting this occurs in an XAF1-independent manner. Future studies will determine if IFN-induced upregulation of TRAIL (encoded by *TNFSF10*), a known inducer of extrinsic apoptosis, is responsible for UVB-enhanced apoptosis in an autocrine fashion. Together, these data suggest that photosensitive responses exhibited by SLE patients may be due to type I IFN priming of KCs that sensitizes cells to undergo increased extrinsic apoptosis after minimal exposures to UVB. Continued investigation into mechanisms by which this occurs will provide important prophylactic options to prevent SLE flares.

Roles and regulation of nuclear keratins in cellular stress response and psoriasis

Erez Cohen and Pierre A. Coulombe

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The ability of cells to respond to a variety of stressors is required to maintain homeostasis and utilizes many regulatory pathways that are aberrant during chronic hyperproliferative diseases. Keratins have traditionally been studied as cytoplasmic structural components in epithelial cells, yet accumulating evidence suggests that keratins regulate several cellular signaling pathways and can translocate to the nucleus. Keratin 16 (K16) is prominently induced in cellular stress and is highly upregulated in psoriasis-activated keratinocytes. Importantly, nuclear K16 has recently been observed in tumor keratinocytes and following treatment with chemical irritants. Preliminary observations suggest that nuclear K16 is also present in psoriatic human tissue. While K16 is highly abundant in psoriasis and is a known regulator of differentiation and innate immune responses in keratinocytes, the cellular functions of K16 and interacting signaling pathways remain incompletely defined. Utilizing single-cell RNA-seq, proteomics, and available K16 transgenic mouse models, we have begun to define the K16 interactome and the role(s) of nuclear K16 during cellular stress response and in psoriasis. These efforts are poised to advance our understanding of how K16 and related stress protein impact the molecular pathways that underlie altered proliferation, differentiation and tissue homeostasis, and immune regulation, in psoriasis.

Epidemiology and Genomic Investigation of Relationship Between Inflammatory Skin Conditions and COVID-19

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Inflammatory skin conditions have previously been shown to be associated with pneumonia, pharyngitis and other respiratory tract infections. However, little is known about their relationship with Coronavirus disease 2019 (COVID-19). Preliminary reports from Italy, Turkey and the USA suggest psoriasis and lupus patients may be at greater risk of COVID-19, and when combined together with rheumatoid arthritis, a previous epidemiological study found these diseases were associated with elevated in-hospital death. To investigate whether patients with different skin conditions are more susceptible to COVID-19, we applied an epidemiological and multi-omics approach. Among 435,019 patients (1,115 with Covid-19) who had at least one encounter with the University of Michigan health system since 2019, having a skin condition was found to increase the risk of COVID-19 (OR=1.55, $p=1.4 \times 10^{-9}$), but decrease the risk of mechanical ventilation (OR=0.22, $p=8.5 \times 10^{-5}$), controlling for race, age, gender, obesity and socioeconomic disadvantage. We compared differential gene expression across nine different inflammatory skin conditions and SARS-CoV-2 infected bronchial epithelial cell lines, finding significant overlap of genes, including four S100 family members located in the epidermal differentiation complex (EDC). Furthermore, IL-17 signaling ($p=4.9 \times 10^{-77}$) was one of the most significantly enriched pathways. Finally, we performed a trans-disease genetic meta-analysis between psoriasis (11,024 cases, 16,336 controls) and Covid-19 (1,678 cases and 674,635 controls), revealing a locus in the EDC whose lead marker (rs12564811) is suggestive significant for psoriasis ($p=1.4 \times 10^{-5}$) and Covid-19 ($p=5.8 \times 10^{-5}$) but genome-wide significant ($p=2.7 \times 10^{-8}$) in the trans-disease meta-analysis. These findings could potentially be explained by disruption to oral/respiratory epithelium as a result of the impact of inflammatory skin conditions on uninvolved skin.

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Poster #6

Novel Targeting of Intracellular Fibro-inflammatory Kinases TAK1 and IRAK4 for the Treatment of Systemic sclerosis

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Abstract

Systemic sclerosis (SSc) is characterized by inflammation coupled with fibrosis in multiple organs. Currently, no approved SSc therapies exist. We identify the kinase TGF β -activated kinase 1 (TAK1) as a key signaling node in SSc-associated fibro-inflammatory signaling driven by both TGF- β and toll-like receptor (TLR) activation. We hypothesize that selective targeting of TAK1, alone or combined with interleukin kinase 4 (IRAK4) represents a novel approach for suppressing inflammatory and fibrotic signaling for the treatment of SSc. We employed a drug-like novel potent small molecule inhibitor of TAK1; HS-276 (orally bioavailable Takinib analogue) as well as a selective IRAK 4 inhibitor, HS-243 in cell cultures with normal and SSc fibroblasts, and in skin organoid systems. Our pre-clinical studies with primary human skin fibroblasts demonstrated that both TAK1 and IRAK4 inhibitors potently attenuated TGF- β 1-induced fibrotic responses, including stimulation of collagen, fibronectin and other profibrotic markers, and fibroblast-myofibroblasts differentiation. The anti-fibrotic effects were equally potent in both preventive and reversal approaches, supporting the role of TAK1 and IRAK4 in fibrosis. Importantly, the inhibitors abrogated collagen synthesis and myofibroblasts differentiation in explanted constitutively active SSc fibroblasts. These findings implicated TAK1 as a major target for therapy in SSc, and identify HS243 and HS276 as potential novel anti-fibrotic agents in the treatment of SSc and other fibrotic diseases.

A novel MX1 reporter line to identify the regulators of autocrine type I interferons in skin

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ABSTRACT

We have recently identified a critical role for Interferon-kappa (IFN κ), an epidermal type I IFN, in inflammatory skin diseases including autoimmune connective tissue diseases¹. Through an autocrine signaling that is unique to keratinocytes (KCs), IFN κ maintains basal IFN responses in KCs that are heightened in diseases like lupus, contact hypersensitivity and select cutaneous malignancies^{2,3,4}. This further leads to increased expression of IFN stimulated genes (ISGs) like *MX1*, *OASL*, and others by KCs. The evolutionary basis of IFN- κ expression may be explained by the role of KCs as a critical barrier against infections, particularly viral⁵. But the molecules and ISGs involved in regulating type I IFNs in skin remain unknown, identifying the regulators will be crucial to understand their biology and identify novel drug targets for autoimmune and inflammatory skin diseases. Hence, we designed a KC reporter line using *MX1* to dissect the role of autocrine IFN responses and identify key molecules/pathways involved in this process. MX1 reporter line was established by infecting KCs with a lentivirus encoding GFP or Luciferase gene under *MX1* promoter. KCs transduced with MX1 construct exhibited higher reporter expression after stimulating with 5ng/ml IFN- α for 24 hours (n=3, p<0.0001). This increase was blocked with the JAK1/JAK2 inhibitor, baricitinib (n=3, 10 μ M). Furthermore, baricitinib suppressed reporter gene expression below basal levels. Genome-wide CRISPR/Cas9 screens are on-going on GFP reporter line, for both coding genes, and lncRNAs, and small-molecular screens on luciferase reporter line. Identified targets from these screens will be functionally validated and characterized. Alongside a novel role for HERC6 (E3 Ubiquitin Ligase), an ISG which is strongly correlated with MX1 expression (r=0.93) in KCs and induced upon IFN- α stimulation (5-fold change) will be determined.

References:

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The deacylase SIRT5 supports melanoma viability by regulating chromatin dynamics

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#These authors contributed equally.

Abstract

Cutaneous melanoma remains the most lethal skin cancer, and ranks third among all malignancies in terms of years of life lost. Despite the advent of immune checkpoint and targeted therapies, only roughly half of patients with advanced melanoma achieves a durable remission. SIRT5 is a member of the sirtuin family of protein deacylases that regulate metabolism and other biological processes. Germline *Sirt5* deficiency is associated with mild phenotypes in mice. Here we show that SIRT5 is required for proliferation and survival across all cutaneous melanoma genotypes tested, as well as uveal melanoma, a genetically distinct melanoma subtype that arises in the eye and is incurable once metastatic. Likewise, SIRT5 is required for efficient tumor formation by melanoma xenografts and in an autochthonous mouse *Braf;Pten*-driven melanoma model. Via metabolite and transcriptomic analyses, we find that SIRT5 is required to maintain histone acetylation and methylation levels in melanoma cells, thereby promoting proper gene expression. SIRT5-dependent genes notably include *MITF*, a key lineage-specific survival oncogene in melanoma, and the *c-MYC* proto-oncogene. SIRT5 may represent a novel, druggable genotype-independent addiction in melanoma.

Poster #9

Interferon kappa Enhanced Psoriasis Disease Severity

Mehrnaz Gharaee-Kermani^{1,3}, Shannon N. Estadt^{1,2},^{1,2} Jianhua Liu¹, Xianying Xing³, Jonathon Theros¹, Tammi J. Reed¹, Dennis Gruszka⁴, Nicole L. Ward⁴, Johann E. Gudjonsson³, J. Michelle Kahlenberg^{1,3,5}

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Psoriasis is a common, chronic inflammatory autoimmune skin disease. Early detection of a type I interferon (IFN) signature occurs in many psoriasis lesions. Interferon kappa (IFN- κ) is an important source of type I IFN production in the epidermis, and psoriasis lesions express an elevated type I IFN signature. In our human data, we identified a correlation between IFN-regulated and psoriasis-associated genes in the skin. We thus wanted to explore the effects of IFN- κ a keratinocyte produced type I IFN, in psoriasis. We used the well-characterized imiquimod (IMQ) psoriasis model for these studies. Three mouse strains at 10-weeks of age were used: wild type (WT) C57Bl/6, C57Bl/6 that overexpress *Ifnk* in the epidermis (TG), and total body *Ifnk*^{-/-} (KO) were used. Psoriasis was induced by topical application of IMQ on both ears for 8 consecutive days. On day 8, the severity of skin lesions and inflammatory cell infiltration was significantly increased in TG>WT>KO mice. H&E staining revealed significantly increased inflammatory cell infiltrates in IMQ treated TG>WT>KO mice. Gene expression analysis via qPCR identified TG>WT>KO for expression of *Mxa*, *Il1b*, *Tnfa*, *Il6*, *Il12*, *Il23*, *Il17* and *Ifng* following IMQ treatment. Further, CD8⁺ and CD4⁺ T cells were increased in TG>WT>KO mice. In summary, we identified a role for IFN- κ as a rheostat for initiation of psoriasis. This suggests that targeting of type I IFNs early in disease may be an effective way of controlling psoriatic inflammation.

STING-IFN- κ -APOBEC3G pathway mediates resistance to CRISPR transfection in keratinocytes

Mrinal Sarkar, Department of Dermatology, University of Michigan, Ann Arbor, MI

Abstract

CRISPR-Cas9 has been proposed for treatment of genetically inherited disorders including of the skin. However, a limitation for widespread use of CRISPR for correction of inherited skin diseases is the poorly understood transfection resistance of keratinocytes (KCs). Here we report that CRISPR transfection activates STING dependent antiviral responses in KCs, resulting in heightened endogenous interferon (IFN) responses through induction of IFN κ , and decreased plasmid stability secondary to induction of the cytidine deaminase *APOBEC3G*. Notably, CRISPR generated KO KCs had permanent suppression of IFN κ and IFN stimulated genes (ISGs) expression, secondary to hypermethylation of the *IFN κ* promoter region by the DNA methyltransferase DNMT3B. Pre-treatment with the JAK1/JAK2 inhibitor, baricitinib prior to CRISPR transfection led to enhanced transfection efficiency, absence of *IFN κ* promoter hypermethylation, and normal IFN κ activity and ISG responses. These results provide insights into the transfection resistance of KCs and indicate that CRISPR mediated gene-correction can lead to permanent alteration of antiviral responses in skin, which can be prevented by JAK1/JAK2 inhibition. This work has major implications for future gene therapy of inherited skin diseases using CRISPR technology.

Poster #11

Genome-wide DNA methylation analysis in lupus keratinocytes identifies differential methylation of genes that regulate apoptosis

Grace A Hile, Patrick Coit, Shannon Estadt, Mitra Maz, Bin Xu, Chang Zeng, Rachael Wasikowski, Lam C. Tsoi, Allison C. Billi, Johann E. Gudjonsson, Amr H. Sawalha, J. Michelle Kahlenberg

Skin inflammation and photosensitivity are common manifestations of cutaneous (CLE) and systemic lupus erythematosus (SLE), yet the mechanisms underlying heightened cell death and epidermal inflammation following ultraviolet (UV) light remain unclear. We performed genome-wide DNA methylation analysis on cultured keratinocyte (KC) DNA from non-lesional, non-sun exposed skin biopsies of SLE patients and healthy controls and identified Hippo signaling as the top canonical pathway. Mutations in this pathway have been shown to increase cell proliferation in oncogenesis models, including in UV-induced neoplasms. However, this pathway has not been studied in inflammatory skin diseases, such as CLE. YAP is a critical component in the regulation of cell proliferation in the Hippo pathway. Through a kinase cascade that includes LATS1/2, TAZ, and WWC1, the Hippo pathway targets YAP for phosphorylation, preventing nuclear translocation and transcriptional activation of TEAD 1-4. YAP's transcriptional activation of TEAD is the principal mechanism for growth and tumor suppression by the Hippo pathway, and inhibition through phosphorylation of YAP may play a role in heightened epidermal cell death. We found significant hypomethylation of LATS1/2 and WWC1 in lupus KC compared to control ($\Delta\beta = -0.17$ and $\Delta\beta = -0.153$, respectively), both of which serve to phosphorylate YAP and thus cause cytosolic sequestration and inhibition of TEAD signaling. Lupus KCs also showed hypomethylation of TEAD1 ($\Delta\beta = -0.17$, $P = 4.36 \times 10^{-9}$) and hypermethylation of upstream co-activator TAZ ($\Delta\beta = 0.12$, $P = 2.20 \times 10^{-4}$). To determine functional relevance of our methylation data, we compared paired RNA-seq samples from the same SLE and control keratinocytes stimulated with IFN α and IFN γ . Indeed, we found a negative correlation between IFN induced genes and methylation signatures, suggesting that the methylation changes we identified result in functional expression differences *in vivo*. To further evaluate the activity of Hippo signaling *in situ*, we analyzed expression and localization of these proteins using immunofluorescent microscopy of frozen lesional biopsies and found a significant increase in cytoplasmic retention of phosphorylated YAP in lupus skin lesions compared to control using Mander's colocalization coefficient. The mean intensity of unphosphorylated YAP was not significantly different in SLE vs. control. Collectively, our work describes a novel mechanistic paradigm for how Hippo signaling through restriction of YAP transcriptional activity is a mechanism of dysregulated apoptosis and photosensitivity in lupus keratinocytes.

Poster #12

Differential gene expression and chromatin accessibility in skin-homing T-cells implicate Th17 differentiation

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To pinpoint genetic determinants of psoriasis, we generated 855 RNA-seq and 989 ATAC-seq libraries passing QC from blood-derived, flow-sorted CD4 and CD8 memory T cells either directly upon isolation or after 24h of CD3-CD28 stimulation (Dynabeads). The filtered and aligned RNA-seq or ATAC-seq reads (>24M per sample) were analyzed by PCA, revealing clear separations on the basis of activation, CD4/CD8, and skin homing (identified by CLA). We utilized DESeq2 to identify differentially expressed genes (DEGs) as function of skin homing, in the context of T-cell subset (CD4/CD8) and CD3/CD28 activation (FDR < 0.05, |log₂ FC| > 0.585). We identified 3,731 skin-homing related DEGs in resting CD4 cells (3,238 for CD8). With respect to activation, 11,273 and 10,010 DEGs were identified in CD4+CLA+ and CD8+CLA+ cells, respectively. For CLA- T-cells, the corresponding DEGs numbered 10,452 and 9,536. We intersected skin-homing and stimulation-related DEGs to identify dual DEGs (dDEGs) with both characteristics, yielding 3,335 dDEGs in CD4+CLA+ and 2972 in CD4+CLA-. Pathway enrichment studies found that dDEGs in both CD4+CLA+ and CD4+CLA- T-cells were enriched for the KEGG term "Th17 cell differentiation" with $p_{adj} = 0.003$ and 0.028 , respectively. The corresponding analysis of CD8 T-cells yielded 2,445 significant dDEGs in CD8+CLA+ ($p_{adj} = 0.001$) and 2,265 in CD8+CLA- ($p_{adj} = 0.014$). Using GREAT to associate differentially accessible regions (DARs) identified by ATAC-seq with nearby genes, a preliminary analysis of 40 subjects found significant enrichment for genes involved in "Th17 differentiation" in activated vs. resting skin-homing CD4 T-cells ($p_{adj} = 7 \times 10^{-6}$). These results identify skin-homing related, T-cell activation-enhanced DEGs in blood-derived immunocytes, highlighting the subtle but important systemic component of psoriatic immune-mediated inflammation.