052 Targeting keratinocytes to potentiate non-viral DNA skin immunization

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Psoriasis is a complex, chronic inflammatory skin disease characterized by inflammation and hyperproliferation of the epidermis. In recent years, the epigenetic mechanism has been proposed to play vital roles in the pathogenesis of psoriasis. N6-methyladenosine (m6A) is the most prevalent internal modification of messenger RNA (mRNA) in eukaryotes, and it is involved in gene expression regulation and various biological processes. To reveal the relationship between m6A methylation and psoriasis, we analyzed the data from the GEO database and found that the expressions of m6A regulatory enzymes have changed in psoriatic lesions compared to the healthy controls, which was also confirmed by performing real-time PCR. The m6A methylation levels are significantly reduced in psoriatic lesions compared with the skin of healthy controls. The results of immunostaining showed that the expression of METTL3, which plays a central role in catalysis of m6A, showed a declining tendency in psoriatic lesions, while the expression of ALKBH5, a main demethylase of m6A, was upregulated in psoriatic lesions compared to the healthy controls. In the back skin of immunodeficient mice, the expression of METTL14 and WTAP these two writers as well as both erasers, FTO and ALKBH5, were all upregulated with the increase of modeling days, and the results of immunofluorescence showed the same tendency. We also separated the epidermis and dermis from the back skin of imiquimod-induced psoriatic mice as well as normal mice, and we found that both of the reduced expression of METTL1 and increased expression of ALKBH5 mainly exits in the epidermis instead of the dermis. This study demonstrates the link between m6A and the regulatory enzymes with psoriasis for the first time, and it provides a solid foundation for further research on the effect of m6A modification in the pathogenesis of psoriasis.

054 Highly Multiplexed Immunophenotyping of Dermatomyositis Skin Lesions

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Dermatomyositis (DM) is an autoimmune systemic disease that most often affects skin and muscle. The pathogenesis of cellular skin inflammation has yet to be investigated. Previous work revealed a type 1 interferon signature characterized predominantly by interferon-beta (IFN-β). To investigate the type 1 interferon signature, we identified pathways and cellular phenotypes in a subset of DM patients. 5 Healthy controls (HC) and 3 DM formalin-fixed, paraffin-embedded (FFPE) samples obtained from trunk, arm, or leg were stained with a panel of 35 metal conjugated antibodies. Regions of interest were imaged at a frequency of 200Hz on the Hyperion Imaging System (Fluidigm). The resulting files were used for unsupervised clustering of cell populations after thresholding each channel. Statistical analysis between groups was performed using the Mann-Whitney test all values reported as mean ± SDA. Skin lesions of DM patients contain an increased number of CD163+ cells compared to normal skin from HC patients (14.4±4.7 vs 2.1±1 cells/ROI; p<0.05). CD163+ cells had increased MPI of key inflammatory pathways: pSTING (3.4±4.9 vs 3.9±1.9), IFNβ (8.2±3.0 vs 4.4±2.7), and BCL2 (8.17±6.5±1.0 vs 1.3±1.0); all p<0.05. A population of CD4 cells was identified that produced higher IFNγ MPI compared to HC CD4 cells (16.4±4.5 vs 8.0±6.0; p<0.01). Lesional DM skin also contained more FOXP3+ CD4 cells when compared to HC (62.3±20.3 vs 6.1±1.3 cells/ROI; p<0.05). The function of these cells is unclear. Compared to HC CD163+ cells in DM appear to be an important source of IFNγ via activation of the STING pathway. IFNγ is produced by both CD163+ and a subset of CD4 cells.

055 Assessment of fidelity to the desmoglein compensation hypothesis in a large pemphigus cohort

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Pemphigus vulgaris (PV) and pemphigus foliaceous (PF) are autoimmune blistering diseases characterized by oral or mucosal lesions in the presence of autoantibodies (autoAb) targeting the cell-adhesion proteins desmoglein (Dsg)1 and Dsg3. Lesion location has been elegantly explained by the Desmoglein Compensation Hypothesis (DCH), which utilizes the epitope distribution of Dsg subtypes as well as autoAb profiles. According to this theory, PF presents with subcorneal lesions in the presence of anti-Dsg1 Abs only, while lesions in PV are suprabasilar and accompanied by anti-Dsg1 only in mucosal PV, or anti-Dsg1 and -Dsg3 in mucocutaneous PV. While the validity of this hypothesis has been supported in the literature, logical inconsistencies have been noted and exceptions have been published in several small-scale studies. Here we compared genome-wide maps of chromatin accessibility of CD8+ T cell subsets from PV patients with active disease at time of visit present with a combination of lesion morphology and anti-Dsg levels that contradict the DCH. The most common contradiction is cutaneous only PV at time of enrolment (n=14), including 7 patients who report no mucosal lesions at any time in their history. Other categories in which lesion morphology does not align with the predicted autoAb status include mucocutaneous disease in the absence of either Dsg1, Dsg3, or both (n=23). We find stark differences based on ethnicity, with the highest proportion of patients that follow the DCH among the Ashkenazi Jewish population (63.5%) and the lowest for African Americans (23%). These findings demonstrate that we need to expand our understanding of pemphigus morphology beyond the DCH, in particular for populations that have not been the focus of previous studies.