A novel Pemphigus vulgaris patient-derived antibody with sequence homology to antibodies directed against desmosomal and non-desmosomal targets

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Pemphigus vulgaris (PV) is an autoimmune blistering skin disease Pemphigus (PV) are the keratinocyte associated desmosomal proteins desmoglein (Dg) 1 and 3. However, the presence of autoantibodies to these targets does not fully explain disease activity and phenotype. We and others have described the presence of numerous non-Dg PV autoantibodies. In this study, we characterized the scope, specificity, and particularly functional non-Dg autoantibodies has not been fully defined. Our group has previously reported the discovery of a patient derived antibody (AS13) that binds 74% heavy-chain homology to anti-thyroid peroxidase (TPO) antibody and 86% light-chain homology to an anti-desmosome antibody as per BLAST alignment. While this antibody did not bind to Dg3, -1 or TPO by ELISA and Western Blot and did not stain intercellular regions on monkey esophagus by IIF we did observe binding to a 55-60kDa protein in HaCaT keratinocyte lysates. Additionally, immunofluorescence defined a cytoplasmic target to HaCaT keratinocytes but no co-localization with the cell membrane or any component thereof, including Dg3. In order to investigate the functional role of this novel antibody and its potential to induce keratinocyte dissociation, HaCaT keratinocytes were grown to confluence and subjected to treatment with increasing concentrations of AS13, an established mouse anti-human Dg1 antibody, and/or AS15. We show that while AS15 induces a strong dose-dependent dissociation of keratinocytes in vitro, the rate of fragment formation at high concentrations of AS13 alone. AS15 in combination with AS13, however, induces an approximately 3-fold higher fragmentation rate than AS13 alone, indicating a synergistic effect of these autoAbs in vivo. While the exact epitope target of the AS13 antibody is yet to be defined, our data suggests a functional role of this novel patient-derived antibody in the skin with potential disease relevance.

Role of hippo signaling in apoptosis of lupus keratinocytes

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Skin inflammation and photosensitivity are common manifestations of systemic lupus erythematosus (SLE), yet mechanisms underlying heightedened cell death and epidermal inflammation following UV light remain unclear. We performed genome-wide DNA methylation analysis on keratinocyte (KCN) DNA from non-lesional, non-sun exposed skin of SLE patients and healthy controls and identified Hippo signaling as the top canonical pathway. Hippo mutations increase cell proliferation in oncogenesis models, including in UV-induced neoplasms. However, this pathway has not been studied in inflammatory skin disease. YAP is a critical component in the regulation of the Hippo pathway. Through a kinase cascade that includes LATS1/2, TAZ and WW1C, the Hippo pathway targets YAP for phosphorylation, preventing nuclear translocation and transcriptional activity of YAP. We found that patients with SLE had an increased YAP expression and an altered YAP expression pattern. We performed a YAP promoter reporter assay in HaCaT cells and showed that YAP expression did not affect IL-5 and IL-17 levels whereas a IL-4Ra treatments affected IL-5 levels only. IgE monitoring showed that both forms of IL-4Ra treatments reduced YAP expression in in vitro cultures. We found that the activity of IL-4Ra treatments in reducing YAP expression in vitro was dependent on the benefit of additional IL-4 inhibition in Th2 directed treatments of AD.

Six North American cases of cutaneous Pemphigus vulgaris with no history of mucosal disease

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Pemphigus is a group of autoimmune blistering diseases including Pemphigus (PV) and Pemphigus foliaceus (PF). Examination of lesion morphology has been elegantly proffered by the Desmoglein Compensation Hypothesis (DCH) based on the epidermal distribution of desmoglein proteins and autoantibody profiles. In this theory, PV is characterized by subcorneal lesions in the presence of only anti-Dg1 antibodies, while PV lesions are suprabasilar and associated with anti-Dg1 and Dg3 in mucocutaneous PV. However, logistical inconsistencies in the DHC have emerged and exceptions have been published in multiple small-scale studies. One of these inconsistencies described by our group and others is that some PV patients present with solely cutaneous disease (cPV). Our group recently reported the discovery of patients with cutaneous Pemphigus vulgaris, a novel PV phenotype characterized by involvement of cutaneous disease without a history of mucosal lesions, further challenging the fidelity of the DHC. To verify the first documented individual cases of PV patients that develop cutaneous disease without a history of mucosal lesions, further challenging the fidelity of the DHC. Two of the 3 patients reported did not fit the common PV-associated HLA genes or display anti-Dg antibodies while in active disease, suggesting some PV patients may display autoAbs via genetic and immune mechanisms that differ from typical mucosal or cutaneous PV.