initiated by the valuable work of Speeckaert et al. on vitiligo.

CONFLICT OF INTEREST

The author states no conflict of interest.

REFERENCES

Cheong KA, Noh M, Kim CH, Lee AY. S100B as a potential biomarker for the detection of cytotoxicity of melanocytes. Exp Dermatol 2014;23:165–71.

Donato R. S100: a multigenic family of calcium-modulated proteins of the EF-hand type with intracellular and extracellular functional roles. Int J Biochem Cell Biol 2001;33:637–68.

Fujiya A, Nagasaki H, Seino Y, Okawa T, Kato J, Fukami A, et al. The role of S100B in the interaction between adipocytes and macrophages. Obesity 2014;22:371–9.

Ghanem G, Loir B, Morandini R, Sales F, Lienard D, Eggermont A, et al. On the release and half-life of S100B protein in the peripheral blood of melanoma patients. Int J Cancer 2001;94:586–90.

Hajduková L, Sobek O, Prchalová D, Bílková Z, Koudelková M, Lukášková J, Matuchová I. Biomarkers of brain damage: S100B and NSE concentrations in cerebrospinal fluid- a normative study. Biomed Res Int 2015;2015:379071.

Hu J, Ferreira A, Van Eldik LJ. S100beta induces neuronal cell death through nitric oxide release from astrocytes. J Neurochem 1997;69:2294–301.

Li S, Zhu G, Yang Y, Guo S, Dai W, Wang G, et al. Oxidative stress induced chemokine production mediates CD8(+) T cell skin trafficking in vitiligo. J Investig Dermatol Symp Proc 2015;17(1):32–3.

Lin J, Yang Q, Wilder PT, Carrier F, Weber DJ. The calcium-binding protein S100B down- regulates p53 and apoptosis in malignant melanoma. J Biol Chem 2010;285:27487–98.

Niven J, Hoare J, McGowan D, Devarajan G, Itohara S, Gannagé M, et al. S100B up- regulates macrophage production of IL1β and CCL22 and influences severity of retinal inflammation. PLoS One 2015;10:e0132688.

Paradisi A, Guidi B, Diociaiuti A, Forni F, Scribano D, Sisto T, et al. Increased S100B protein serum levels in psoriasis. J Dermatol Sci 2007;48:148–50.

Portela LVC, Brenol JCT, Walz R, Bianchin M, Tort ABL, Canabarro UP, et al. Serum S100B levels in patients with lupus erythematosus: preliminary observation. Clin Diagn Lab Immunol 2002;9:164–6.

Rothermundt M, Peters M, Prehn JH, Arolt V. S100B in brain damage and neurodegeneration. Microsc Res Tech 2003;60:614–32.

Speeckaert R, Voet S, Hoste E, van Geel N. S100B is a potential disease activity marker in non-segmental vitiligo. J Invest Dermatol 2017;137:1445–53.

Thelin EP, Nelson DW, Bellander BM. A review of the clinical utility of serum S100B protein levels in the assessment of traumatic brain injury. Acta Neurochir 2017;159:209–25.

Wilhelm KR, Yanamandra K, Gruden MA, Zamotin V, Malisauskas M, Casaite V, et al. Immune reactivity towards insulin, its amyloid and protein S100B in blood sera of Parkinson's disease patients. Eur J Neurol 2007;14:327–34.

Zimmer DB, Chessher J, Wilson GL, Zimmer WE. S100A1 and S100B expression and target proteins in type I diabetes. Endocrinology 1997;138:5176–83.

See related article on pg 1501

Act1: A Psoriasis Susceptibility Gene Playing its Part in Keratinocytes

Ryan P. Hobbs[1], Susan H. Smith[2] and Spiro Getsios[2]

Unchecked inflammation, impaired keratinocyte differentiation, and heightened host defense responses typify psoriasis. Lambert et al. make clever use of psoriasis patient genetics and whole transcriptome RNA-Seq analysis to implicate Act1 in these seemingly variegated processes by keeping IL-17 receptor signaling in check while supporting differentiation and limiting innate immune responses in human keratinocytes.

Journal of Investigative Dermatology (2017) **137,** 1410–1412. doi:10.1016/j.jid.2017.01.023

[1]*Department of Biochemistry and Molecular Biology, Johns Hopkins University Bloomberg School of Public Health, Baltimore, Maryland, USA; and* [2]*Discovery and Preclinical Development, Dermatology Therapy Area Unit, GlaxoSmithKline, Collegeville, Pennsylvania, USA*

Correspondence: Spiro Getsios, Discovery and Preclinical Development, Dermatology Therapeutic Area Unit, GlaxoSmithKline UP1410, 1250 S. Collegeville Road, Collegeville, Pennsylvania 19426, USA. E-mail: spiro.x.getsios@gsk.com

Psoriasis vulgaris is a chronic autoimmune inflammatory skin disorder characterized by erythematous, scaly plaques that tend to form at discrete body sites (Greb et al., 2016). Abnormal keratinocyte proliferation, differentiation, and host defense mechanisms are evident within psoriatic plaques in association with enhanced vascularization and pronounced leukocyte recruitment into the underlying dermis. For some time, keratinocytes were thought to be the main cellular culprits in the pathogenesis of psoriasis because epidermal morphology (i.e., hyperplasia, acanthosis, hypogranulosis, hyperkeratosis, parakeratosis) was changed so profoundly within plaques. The advent of successful therapies that target inflammation has since swayed the pathogenic pendulum for psoriasis in the direction of the immune system. In particular, existing and emerging biologic therapies that target receptors for proinflammatory cytokines, such as tumor necrosis factor-α (TNFα), IL-17 family members, and IL-23, have emphasized the central role that the immune system plays in the disease. More than likely, an intricate dialogue between keratinocytes and immune cells, in addition to vasculature, nerves, dermal fibroblasts, and possibly even microbes, contributes to the exacerbation of inflammatory cycles under susceptible metabolic and environmental conditions that drive disease progression in cutaneous lesions.

> "An intriguing role for Act1 in keratinocyte differentiation and host defense response emerged upon unbiased, whole transcriptomic evaluation of Act1-deficient keratinocytes that may not have surfaced by more directed analysis of its impact on IL-17 receptor signaling."

Although the causes of psoriasis are multifactorial, this disease clearly has a

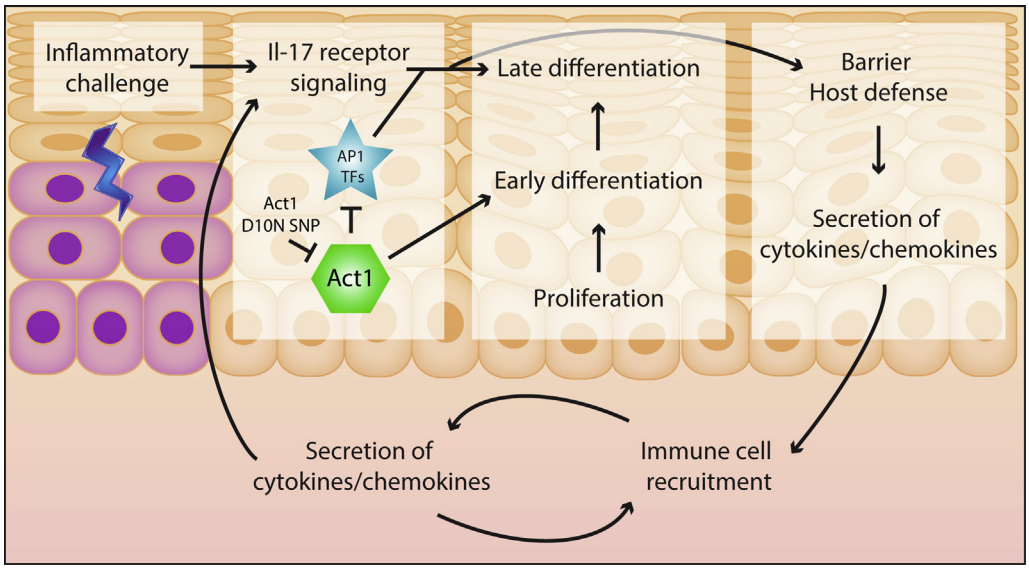

Figure 1. Model depicting the roles of Act1 in regulating epidermal homeostasis. After an inflammatory challenge (e.g., pathogen, allergen), IL-17R signaling becomes activated in a manner requiring Act1 (green hexagon) to suppress the expression of AP1 transcription factors (AP1 TFs, blue star). IL-17R signaling then promotes the expression of genes associated with both late differentiation and epidermal barrier/host defense mechanisms. Independent of IL-17 receptor signaling, Act1 also promotes the expression of early differentiation genes. Persistent inflammation can lead to sustained expression of inflammatory cytokines and chemokines and recruitment of immune cell effectors, which likely participate in a feed-forward inflammatory loop chronically interfering with epidermal differentiation and host defense. SNP, small nucleotide polymorphism; TFs, transcription factors.

strong genetic component (Greb et al., 2016; Harden et al., 2015). A wealth of information from genome-wide association studies and whole exome DNA sequencing has fortified the link between immune system dysregulation and psoriasis, as exemplified by the reasonably well-studied psoriasis susceptibility locus, *HLA-C* (formerly known as *PSORS1*), harboring human leukocyte antigen genes within the major histocompatibility complex. A number of additional psoriasis susceptibility loci as well as small nucleotide polymorphisms have been identified that further associate cytokine signaling pathways with the epidermal thickening and inflammation that is associated with psoriasis pathophysiology. Included among these candidate susceptibility genes for psoriasis is TNF receptor associated factor 3 interacting protein 2 (*TRAF3IP2*), which encodes for Act1 and also serves as a potential risk factor for psoriatic arthritis. The case for a *TRAF3IP2* small nucleotide polymorphism, resulting in an Act1 Asp10Asn variant, impacting immune surveillance and inflammation would seem relatively straightforward, given the known role of Act1 in dampening IL-17 receptor (IL-17R) signaling in mice (Chang et al., 2006; Qian et al., 2007; Wang et al., 2013). However, these compelling associations between Act1 and IL17 pathway regulation remain to be formally tested in human cells before drawing clear mechanistic links from patient genetics to disease phenotype.

The study by Lambert et al. (2017) rises to this challenge by taking advantage of an inducible gene silencing system for Act1 in a relatively normal human keratinocyte cell line, namely N/TERT keratinocytes. Because IL-17R is expressed in keratinocytes and is known to respond to IL-17A and related family members produced by CD4+ T cells, in a differentiation-dependent manner, the authors logically predicted that Act1 loss of function would impact IL-17R signaling in epidermal keratinocytes. This possibility was neatly substantiated on Act1 silencing, with amplified IL-17 responses evident in keratinocytes, particularly after an inflammatory challenge invoked by the addition of serum to these monolayer cultures. An intriguing role for Act1 in keratinocyte differentiation and host defense responses emerged from this study upon unbiased, whole transcriptomic evaluation of Act1-deficient keratinocytes, a role that may not have surfaced using more directed analysis of its impact on IL-17R signaling (Figure 1). In particular, Act1 loss interfered with normal execution of early stages of keratinocyte differentiation and led to upregulation of late differentiation products, similar to what is found in psoriatic epidermis. Moreover, epidermal host defense mechanisms that are alerted to microbial challenges via innate immune responses that feed into the IL-17 signaling pathways also seem to depend on intact Act1 function in keratinocytes. Interestingly, Act1 loss did not increase keratinocyte proliferation, which is a major feature of psoriatic epidermis, perhaps reflecting a later role for the signaling regulator in commitment to terminal differentiation by epidermal progenitor cells. Now that these RNA-Seq datasets are deposited in the public domain (GEO; GSE86451), it will be intriguing to determine how closely these Act1-deficient keratinocyte monolayer cultures reflect the psoriatic transcriptome, because multiple cutaneous cell types, including immune cells present in cutaneous lesions, do express Act1 (Wang et al., 2013).

The mechanisms by which Act1 regulates keratinocyte differentiation, epidermal host defense, and IL-17R signaling responses remain somewhat unclear from the Lambert et al. study. In silico analysis of DNA binding consensus sites, complemented with expression profiling, suggests that AP1 family transcription factors (e.g., FosB and Fra1) may be involved in Act1

pathway regulation, a finding that is supported by their known roles in bridging keratinocyte differentiation and cytokine signaling. The relative importance of AP1 family members in Act1-mediated IL-17R signaling and keratinocyte differentiation will need to be tested formally, as will the relative importance of Act1 in epidermal homeostasis in vivo or possibly by using more complex in vitro human skin culture systems where host defense responses are identified with greater certainty. The authors acknowledge these potential limitations of their study, as well as the fundamental differences between gene silencing and a *TRAF3IP2* point mutation that is expected to disrupt Act1 function. As gene editing approaches become adopted more routinely, it will become possible to assess the consequences of these specific *TRAF3IP2* genetic variants for keratinocyte differentiation and IL-17 signaling in a manner that more closely reflects patient circumstances. For this purpose, it may also be useful to derive somatic skin cells or iPS-derived keratinocytes from donors that carry these mutations for studying relevant signaling pathways, modeling disease states, and testing novel precision medicine-based therapeutic applications for psoriasis.

Conceptually, the Lambert et al. study illustrates nicely the use of genetics as a guide for interrogating the role of disease susceptibility variants in epidermal homeostasis and innate immunity in a patient-specific manner. Understanding how these discrete genetic changes manifest into integrated cellular and tissue-level responses that drive inflammatory disease phenotypes will allow for a more complete picture of psoriasis disease pathology, which remains a bit of a black box in spite of all the recent advances in treating this disease. Similar approaches can be applied readily to other chronic inflammatory dermatoses about which much less is known. For instance, atopic dermatitis is an intensely pruritic, chronic, relapsing, inflammatory skin disease that is associated with defective epidermal differentiation, skin barrier function, and unrestrained immune responses that are in some ways related to, but more notably distinct from, psoriasis, including genetic risk factors (Bin and Leung, 2016; Guttman-Yassky et al., 2011). Candidate gene association studies indicate that filaggrin (*FLG*) null gene mutations are the most significant known risk factor for atopic dermatitis, followed by type 2 T helper lymphocyte signaling pathways. A number of studies have already examined the consequences of *FLG* deficiency using knockdown or patient gene variant approaches in human keratinocyte culture models, with somewhat incongruous outcomes with respect to epidermal differentiation and barrier function (Niehues et al., 2016 and citations therein). More recently, altered expression within the claudin (*CLDN*) gene family has been implicated in barrier dysfunction of patients with atopic dermatitis, and this is supported by evidence of *CLDN1* small nucleotide polymorphisms associated with atopic dermatitis in some North American populations (De Benedetto et al., 2011). Although the prevailing model for claudin-1 function would suggest a primary role in tight junction barrier function, following the experimental paradigm laid out by Lambert et al., further study may reveal unexpected roles for *CLDN1* variants in innate immune responses within the epidermis, which could act as a trigger for subsequent inflammation. Identifying mechanistic links between patient-specific genetic variants in skin disease and their roles in epidermal homeostasis, barrier function, and host defense may help to elucidate common pathways of keratinocyte-immune cell crosstalk, pathways that could fuel bouts of inflammation in patients and which may provide an area ripe for drug targeting in dermatology.

CONFLICT OF INTEREST

SHS and SG are employees and shareholders of GlaxoSmithKline, llc. (GSK). This commentary does not relate to work being done at GSK, nor does their status as employees and shareholders unduly influence its contents.

REFERENCES

Bin L, Leung DY. Genetic and epigenetic studies of atopic dermatitis. Allergy Asthma Clin Immunol 2016;12:52.

Chang SH, Park H, Dong C. Act1 adaptor protein is an immediate and essential signaling component of interleukin-17 receptor. J Biol Chem 2006;281:35603–7.

De Benedetto A, Rafaels NM, McGirt LY, Ivanov AI, Georas SN, Cheadle C, et al. Tight junction defects in patients with atopic dermatitis. J Allergy Clin Immunol 2011;127:773–86. e1-7.

Greb JE, Goldminz AM, Elder JT, Lebwohl MG, Gladman DD, Wu JJ, et al. Psoriasis. Nat Rev Dis Primers 2016;2:16082.

Guttman-Yassky E, Nograles KE, Krueger JG. Contrasting pathogenesis of atopic dermatitis and psoriasis—part I: clinical and pathologic concepts. J Allergy Clin Immunol 2011;127:1110–8.

Harden JL, Krueger JG, Bowcock AM. The immunogenetics of psoriasis: a comprehensive review. J Autoimmun 2015;64:66–73.

Lambert S, Swindell WR, Tsoi LC, Stoll SW, Elder JT. Dual role of Act1 in keratinocyte differentiation and host defense: *TRAF3IP2* silencing alters keratinocyte differentiation and inhibits IL-17 responses. J Invest Dermatol 2017;137:1501–11.

Niehues H, Schalkwijk J, van Vlijmen-Willems IM, Rodijk-Olthuis D, van Rossum MM, Wladykowski E, et al. Epidermal equivalents of filaggrin null keratinocytes do not show impaired skin barrier function [epub ahead of print]. J Allergy Clin Immunol 2016; http://dx.doi.org/10.1016/j.jaci.2016.09.016 (accessed 26 February 2017).

Qian Y, Liu C, Hartupee J, Altuntas CZ, Gulen MF, Jane-Wit D, et al. The adaptor Act1 is required for interleukin 17-dependent signaling associated with autoimmune and inflammatory disease. Nat Immunol 2007;8:247–56.

Wang C, Wu L, Bulek K, Martin BN, Zepp JA, Kang Z, et al. The psoriasis-associated D10N variant of the adaptor Act1 with impaired regulation by the molecular chaperone hsp90. Nat Immunol 2013;14:72–81.