Old Herborn University Seminar Monograph 28:
The epidermis of man: co-existing with commensals.
Editors: Peter J. Heidt, Pearay L. Ogra, Michael Zasloff, and Volker Rusch.
Old Herborn University Foundation, Herborn, Germany: 1-17 (2015).

EPIDERMIS AS A DYNAMIC INTERFACE

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INTRODUCTION

Too long viewed as a mere battle-ground for the immune system, the epidermis is asserting its rightful place at the centre of cutaneous biology and pathophysiology. While immunologists seek ever finer distinctions between T cell subsets in inflammatory lesions, it is now increasingly clear that the protective requirements of the skin dictate virtually every metabolic process (including adaptive immune responses) in its underlying layers. True, there are 'outside-to-inside-back-to-outside' vicious cycles, whereby immune responses further compromise epidermal function, and there are also examples of primary immune disorders, such as autoimmune and bullous diseases, HIV infections, and superantigen-initiated flares of erythrodermic psoriasis, where a primary inflammatory infiltrate can produce downstream abnormalities in epidermal function (e.g., for HIV, see *Gunathilake*, 2010). But as the example of filaggrin-deficient atopic dermatitis eloquently demonstrates, most cutaneous immune phenomena occur downstream of primary epidermal insults, whether inherited or acquired, and these responses are recruited only when epidermal homeostatic responses fail to promptly re-establish normal cutaneous function. In this brief review, we will consider:

i) a new 'holistic' view of epidermal defence;
ii) a concise review of the structural basis for the barrier with an update on tight junctions and the corneocyte lipid envelope;
iii) intra-epidermal metabolic processes that are regulated by barrier requirements; and
iv) certain homeostatic signalling mechanisms that regulate these metabolic responses.

BRIEF REVIEW OF BARRIER STRUCTURE AND FUNCTION

The two-compartment model

The protective functions of the skin, including the permeability barrier, largely localize to the outer epidermis and stratum corneum (SC) (Table 1) (Figure 1). The SC is an anucleate structure, arranged in a 'brick and mortar' mosaic of flattened corneocytes ('bricks'), embedded in lipid-enriched extracellular matrix ('mortar') that is organized into parallel stacks of lamellar bilayers, enriched in ceramides, cholesterol, and free fatty acids (FFA) (*Elias* and *Menon*, 1991). These water repellent lipids restrict the outward flow of water, while also impeding the inward absorption of toxins, allergens, and microbial pathogens (*Prausnitz* et al., 2012). It is the secretion of the contents of multiple, small ovoid lamellar

Table 1: Defensive gradients in the outer epidermis

Functions:	Outer surface (sebaceous glands)	Stratum corneum	Stratum granulosum
Antimicrobial:	AMP, FFA (↓pH)	FFA (↓pH), AMP, SPI	AMP, TLR
Permeability barrier:	—	Cholesterol, Cer, FFA in lamellar bilayers	Tight junction (larger xenobiotes)
Antioxidant:	Vit. E	Vit. E, Sprr2d, Sprr2h, Slpi	SOD, CoQ, catalase, GluTR
UV-B:	—	t-UCA (melanin)	Melanin
Mechanical:	—	Cornified envelopes	—
Cohesion:	—	Lipids, Corneodesmosomes	Desmosomes, Adherens junctions
Cytokine activation:	—	IL-1α/β release	TNFα, IL-1α/β, GMCSF, IL-6, NGF, AR, VEGF
Neurosensory:	—	—	TRPVs, TRPM8
Hydration:	Glycerol	FLG → NMF; glycerol, urea	AQP channels, Urea transporters

Abbreviations: AMP, antimicrobial peptide; Cer, ceramide; CoQ, co-enzyme Q; FFA, free fatty acid; GluTR, glutamyl tRNA reductase; GMCSF, granulocyte-macrophage colony-stimulating factor; IL, interleukin; NGF, nerve growth factor; NMF, natural moisturizing factor; Slpi, serine leukocyte protease inhibitor; SOD, superoxide dismutase; SPI, serine protease inhibitor; TLR, toll-like receptor; TNF, tumor necrosis factor; TRPM8, transient receptor potential melastatin-8; TRPV, transient receptor potential vanilloid; t-UCA, trans-urocanic acid; VEGF, vascular endothelial growth factor.

bodies (LB) (*Elias* and *Menon*, 1991) that delivers both lipid precursors and hydrolytic 'processing' enzymes that generate the hydrophobic species, ceramides (Cer), free fatty acids (FFA), and cholesterol, that mediate the permeability barrier (Figure 2). These three lipids, along with as-yet unidentified amphiphilic molecules, are required for the organization of the secreted lipids into mature lamellar bilayers (*Elias* and *Menon*, 1991).

The corneocyte-bound lipid envelope (CLE)

The external surface of the cornified envelope (CE) is coated with a monolayer of ω-hydroxyceramides (ω-OH-Cer) that is covalently bound to peptides (1° involucrin) within the CE (*Zheng* et al., 2011; *Breiden* and *Sandhoff*, 2014; *Rabionet* et al., 2014) (Figure 3). Both the origin and the function of this structure are still uncertain. While most workers believe that it is formed from a pool of secreted acylCer, the CLE also could derive from the insertion of a myriad of lamellar body limiting membranes during the exocytosis of these organelles (*Elias* et al., 2014). We noted that the CLE fails to form in several inherited and acquired disorders that compromise steps that either generate acylCer, or oxidize the ω-OH-linoleate moiety of acylCer (Figure 2). Since all of these disorders are characterized by a faulty permeability barrier, poor SC hydration, and impaired desquamation, it is tempting (but still not certain) that the CLE is linked to one or more of these functions (*Elias* et al., 2014).

The tight junction (TJ) controversy

How should we interpret an ever-expanding literature that proclaims a

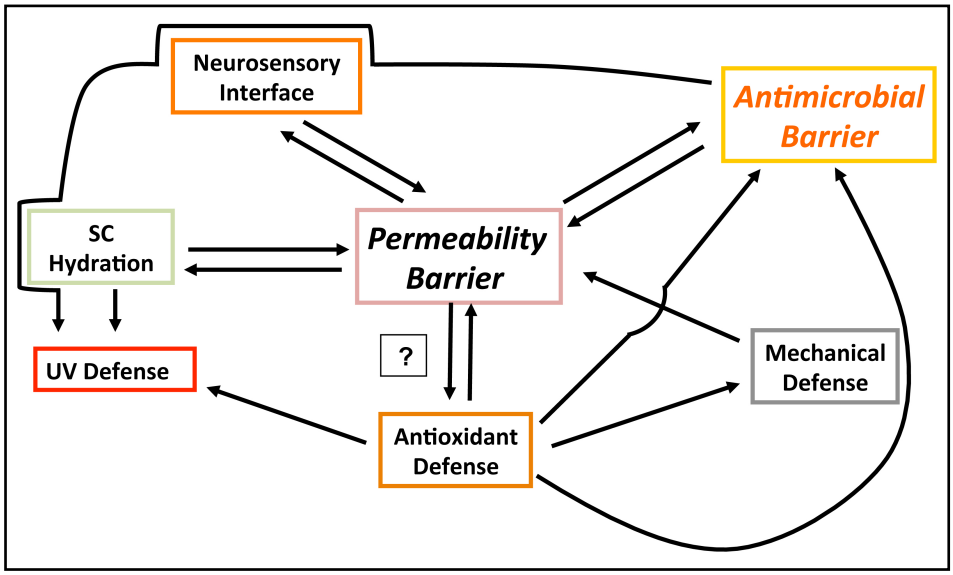

Figure 1: Protective (defensive) functions are related, co-regulated and interdependent.

potential role for TJ in normal permeability barrier function [e.g., (*Brandner* et al., 2002; *Kubo* et al., 2012)], as well as a potential role for abnormal TJ function in AD (De *Benedetto* et al., 2011)? We will attempt to navigate this heavily-invested subject as follows: First, complex TJ structures, such as those found in the kidney and gastrointestinal (GI) tract, do not occur in adult keratinizing epithelia (*Elias* et al., 1977). Second, with the exception of highly complex TJ in renal collecting tubules, where they comprise multitiered, overlapping sites of membrane fusion ('zonulae occludentes'), in other tubular epithelia, such as the trachea and GI tract, these junctions provide a relatively poor barrier against paracellular water movement (*Marchiando* et al., 2010; *Suzuki*, 2013). Much of the confusion in the skin-related literature has occurred because 'TJ *proteins*' are widely equated with 'TJ' (*Brandner* et al., 2002; *Furuse* et al., 2002; *Kubo* et al., 2012). Certainly, multiple TJ proteins heavily decorate the apical-lateral plasma membranes of cells in the outer stratum granulosum of normal adult epidermis, forming 'kissing points'. However, these focal attachments; i.e., 'maculae occludentes' (*Elias* et al., 1977), do not comprise true zonulae occludentes (=TJ), as occur in tubular epithelia. The most compelling evidence that these putative TJ play no direct role in the paracellular water barrier comes from solvent extraction studies, where removal of SC lipids by repeated, gentle, lipid solvent swabbing *completely abrogates* the permeability barrier (*Grubauer* et al., 1989). It should be noted that this observation also excludes a possible 'back-up' role for TJ-like structures in the *water* barrier, although it remains possible that true TJ eventually could begin to form in response to such repeated solvent wipes. Moreover, these incomplete structures could suffice to interdict the paracellular passage of larger xenobiotes, particularly when the overlying lipid-based barrier becomes defective, as occurs in atopic dermatosis (*De Benedetto* et al., 2011).

Yet, these structures, though insufficient to contribute directly to the nor-

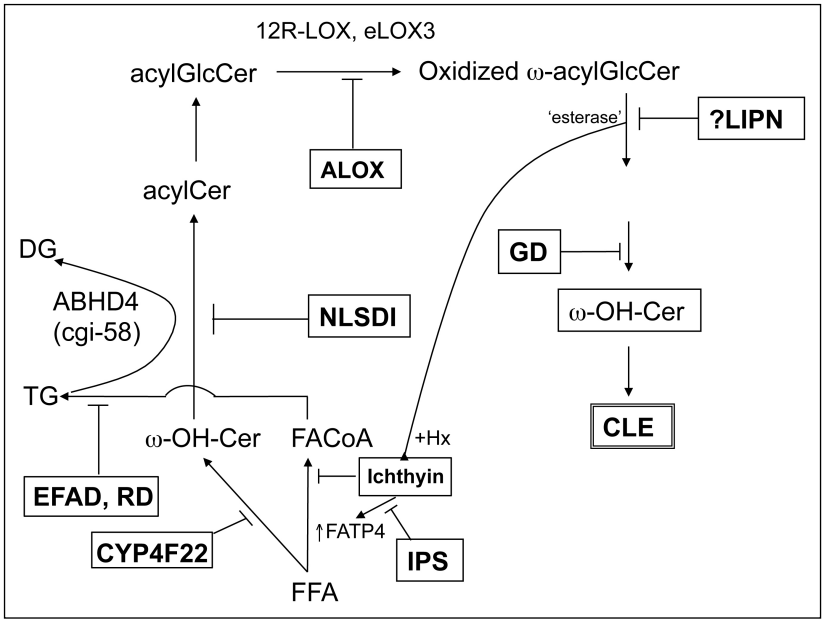

Figure 2: Pathways leading to CLE formation: insights from inherited and acquired lipid metabolic disorders. (Modified from: Elias et al., 2014).

mal water barrier, are nonetheless critical for the development of permeability barrier competence. Transgenic knockout of the key TJ protein, claudin 1, results in a fatal, post-natal permeability barrier abnormality (*Furuse* et al., 2002). Indeed, our recent studies show that replete TJ are present early in epidermal development, but they become functionally incompetent later in foetal life in parallel with establishment of the lipid-based barrier (*Celli* et al., 2012). Although an acquired reduction in the expression of the TJ protein, claudin 1, has been reported in atopic dermatitis (*De Benedetto* et al., 2011), treatment of cultured human keratinocytes with the Th2 cytokine, IL-4, instead *upregulates* claudin 1 expression, while simultaneously *downregulating* another TJ protein, occludin (Y. Hatano, personal communication). Moreover, occludin (but not claudin) protein levels decline in filaggrin-deficient human epidermis (*Gruber* et al., 2011). Hence, it is likely that abnormalities in TJ proteins in atopic dermatitis, should they occur, likely result from the Th2-dominant milieu, which is known to downregulate many other epidermal differentiation-linked proteins [e.g., (*Howell* et al., 2008)].

Since adult epidermis does not generate the types of complex zonulae occludentes necessary to impede paracellular water movement, attention should be focused instead on the possible functions of these incomplete junctions (maculae occludentes) in normal epidermis; and how acquired defects in such focal connections could contribute to disease pathogenesis. We believe that these structures perform important 'fence functions' in adult epidermis, including polarizing the direction of lamellar body secretion towards the apex of the outermost granular layer (*Elias* et al., 1998), while also restricting selected membrane transporters, such as the sodium-hydrogen antiporter 1 (NHE1), to the apical plasma membrane of these cells.

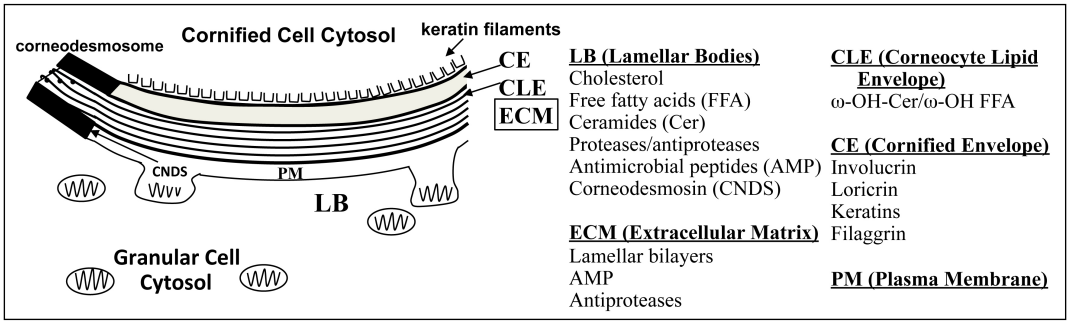

Figure 3: Diagram of stratum corneum membrane domains. (Modified from: Schmuth et al., 2008).

INTERDEPENDENCE OF, AND INTERRELATIONSHIP BETWEEN EPIDERMAL DEFENSIVE FUNCTIONS

While it is common practice to list the various defensive functions of the skin as discrete processes (Table 1), in most cases, these functions are interrelated, co-regulated, and interdependent. As is evident from Figure 1, more and more connections are emerging between these defensive functions, of which we will highlight only a few for consideration here. Best appreciated are the connections between the permeability barrier and antimicrobial defence. Shared structural and biochemical processes (*Elias*, 2007), as well as common metabolic processes, unite these two functions (Table 2).

Moreover, epidermal lamellar bodies provide a common delivery mechanism for components with overlapping functions, such as free fatty acids and antimicrobial peptides (AMP) (Table 2), of which at least one, the cathelicidin carboxyterminal peptide, LL-37, is required not only to restrict pathogen invasion, but also as an apparent structural component of lamellar bilayers (*Aberg* et al., 2008). In multiple clinical situations; in experimental perturbations; and after applications of therapeutic ingredients that either compromise or improve permeability barrier homeostasis, corresponding alterations occur in LL-37, and to a lesser extent, in hBD2 expression (*Aberg* et al., 2008; *Rodriguez-Martin* et al., 2011) (Figure 4).

Table 2: How permeability and antimicrobial barriers are linked

1. Co-localization of both functions to extracellular ('mortar') domains
2. Pathogens attempt to invade through SC extracellular domains
3. Some permeability barrier lipids (e.g., free fatty acids and sphingosine) exhibit potent antimicrobial activity
4. Certain antimicrobial peptides (AMP) localize to lamellar bodies (along with lipids), and are co-delivered to SC extracellular domains
5. Both AMP expression and secretion accelerate after permeability barrier disruption, paralleling up-regulation of lipid synthesis
6. At least one AMP (LL-37) is required for permeability barrier homeostasis
7. Certain serine proteases (e.g., secretory leukocyte protease inhibitor, SLPI) that regulate SC cohesion also exhibit potent antimicrobial activity.

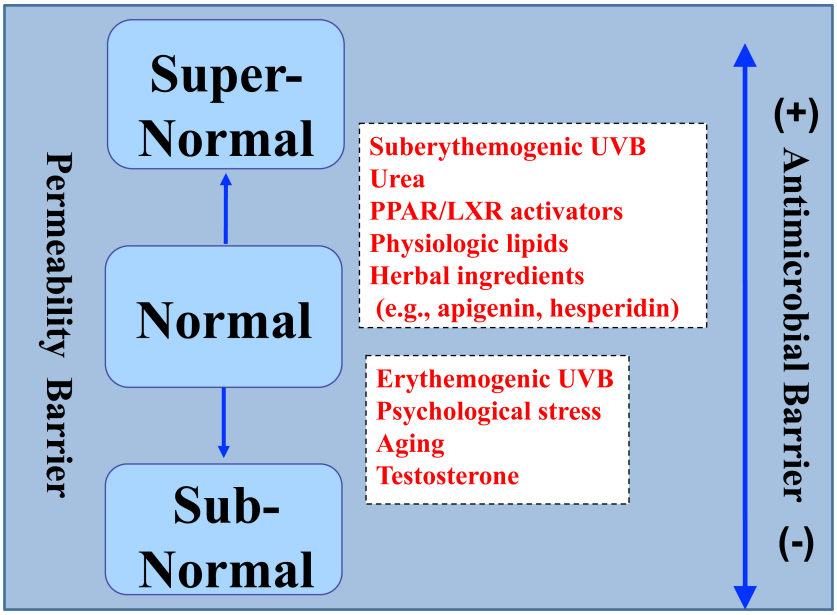

Figure 4: Parallel changes in barrier function and cathelicidin expression. (Modified from: Rodriguez-Martin et al., 2011).

Lamellar bodies also deliver proteases and anti-proteases that initially regulate SC cohesion, and then orchestrate the digestion of corneodesmosomes (*Caubet* et al., 2004; *Brattsand* et al., 2005) (Figure 3). But corneodesmosome degradation is only the first in a series of subsequent cellular events that leads to the eventual shedding of corneocytes from the skin surface (*Lin* et al., 2012) (Figure 5). Finally, as noted above, lamellar bodies also secrete at least two antimicrobial peptides, human beta-defensin2 (hBD2) and LL-37, into the SC extracellular domains (*Oren* et al., 2003; *Braff* et al., 2005; *Aberg* et al., 2007). Because they appear to be so intertwined, it becomes a matter of semantics as to whether not only these two, but also whether several other functions should be considered as discrete or interrelated processes (Figure 1).

The multiple functions that are impacted by the epidermal structural protein, filaggrin, serve as another illustrative example of the link between multiple defence functions. First, the full-length protein becomes a component of the corneocyte envelope (CE) (*Eckert* et al., 2004; *Presland*, 2009), contributing to epidermal mechanical defence (*Gruber* et al., 2011). We have shown that an intact CE is required for the supramolecular organization of secreted lipids into lamellar bilayers, as eloquently demonstrated in two disorders of cornification, transglutaminase 1-deficient lamellar ichthyosis (*Elias* et al., 2002), and loricrin keratoderma (*Schmuth* et al., 2004). But it is the subsequent, humidity-dependent proteolysis of FLG above the mid-SC (*Scott* and *Harding*, 1986), that impacts an even broader suite of functions (Figure 6). Following FLG hydrolysis, its constituent amino acids are further deiminated, both enzymatically and non-enzymatically, into a suite of polycarboxylic acids (= 'natural moisturizing factor') that not only account for much of SC hydration, but also contribute to defence against UV-B and to the acidification of the SC

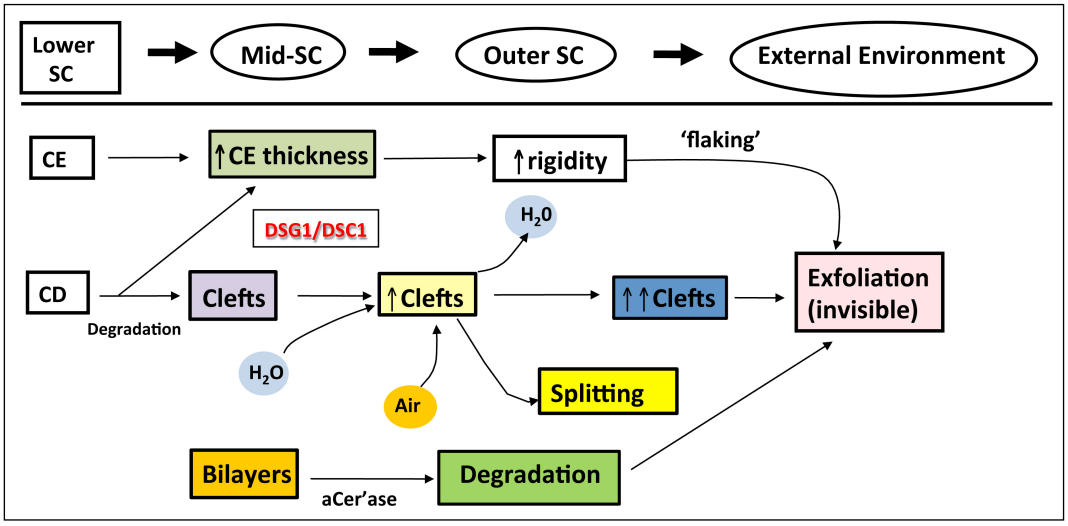

Figure 5: Basis for normal exfoliation: summary of observations. (Modified from Lin et al., 2012). Abbreviations: aCer'ase, acidic ceramidase; CD, corneodesmosome; CE, cornified envelope; DSC1, desmocollin 1; DSG1, desmoglein 1; SC, stratum corneum.

(Figure 6). The reduced pH of the SC in turn is critical for multiple functions, including not only antimicrobial defence, but also permeability barrier homeostasis (*Mauro* et al., 1998), SC cohesion, and pro-inflammatory cytokine activation.

We next highlight another recent example of linked functions that recently emerged from the laboratory of Sabine Werner (Inst. of Cell Biology, Zurich), who showed that a key transcription factor, nuclear factor erythroid 2-related factor 2 (Nrf2), regulates the expression of two cornified envelope precursors, small proline-rich proteins (Sprr2b and Sprr2h). This transcription factor also regulates expression of a potent antimicrobial protein, secretory leukocyte protease inhibitor (Slpi), which is also an inhibitor of serine proteases (kallikreins) that regulate SC cohesion (Figure 7). The cohesiveness of the SC in turn is critical for both permeability barrier function and antimicrobial defence. Together, these examples of functional links illuminate how discrete epidermal protective functions should instead be considered as components of a broader, protective 'superfunction' of the skin.

METABOLIC MECHANISMS THAT MAINTAIN EPIDERMAL HOMEOSTASIS

Life in a terrestrial environment requires constant vigilance, accompanied by responses, either draconian or subtle, to external perturbations that potentially threaten the organism with desiccation, microbial invasion, oxidant damage, UV-B-induced apoptosis, and/or impaired mechanical defence. Consider the most dramatic example, i.e., an external thermal burn, with its potentially devastating consequences. The foremost threat to such patients, of

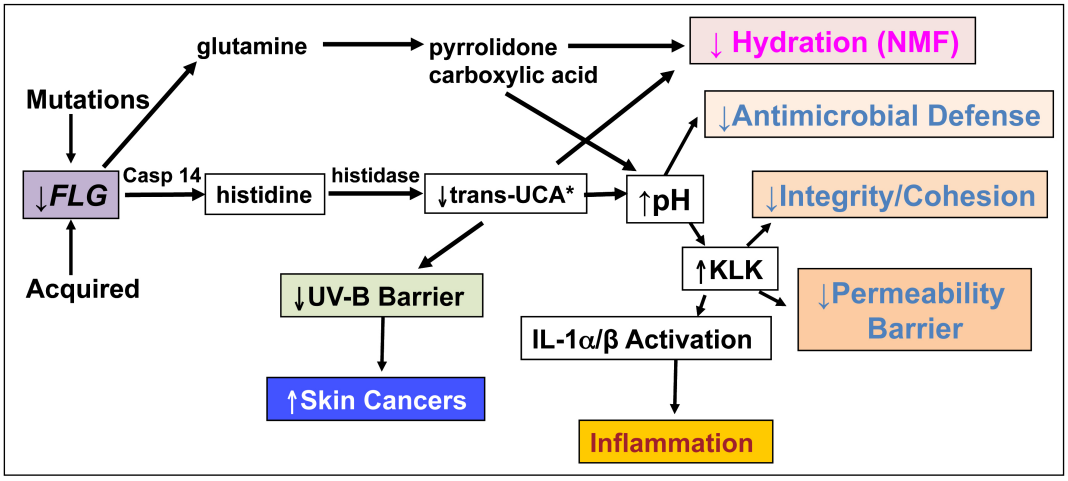

Figure 6: Multiple downstream consequences of filaggrin deficiency in atopic dermatitis: *Trans-urocanic acid (t-UCA) is the most potent endogenous UV-B filter in lightly-pigmented skin. Loss of t-UCA could account for the higher incidence of non-melanoma skin cancers in AD. (Modified from: Thyssen et al., 2013).
Abbreviations: Casp 14, Caspase 14; KLK, kallikrien, NMF, natural moisturizing factor

course, is rapid desiccation due to an unrestricted loss of internal fluids and electrolytes, as well as an increased susceptibility to pathogen invasion. Yet, even following such potentially catastrophic injuries, the skin attempts to repair itself. What is the driving force behind the repair of such wounds? Entire generations of surgeons and skin biologists have focused once again upon 'inside-to-outside' phenomena, related either to the initial inflammatory responses, platelet-derived growth factors, granulation tissue, collagen remodelling, and/or wound contracture as key 'drivers' of wound healing. Re-epithelialization often is noted only in passing as the inevitable downstream consequence of these earlier events. Few, if any of these investigators have considered the possibility that it could be the *imperative to re-establish permeability barrier homeostasis* that likely 'drives' much

Table 3: Chronology of Metabolic Response to Acute Barrier Disruption

Chronology:	0→20 min	20 min→2 hrs	30 min→6 hrs	6 hrs→12 hrs	16+ hrs
Event:	Secretion of preformed pool of lamellar bodies	Terminal differentiation (physiologic apoptosis)	↑Lipid synthesis + secretion	↑Lipid processing	↑DNA synthesis
Known signals:	$\downarrow Ca^{2+}$	KLK → PAR2	SREBPs; ↑IL-1α; $\downarrow Ca^{2+}$	?	AR, NGF, IL-1α
Effects of occlusion:	Blocks	Blocks	Blocks both lipid synthesis and transport	Blocks	Blocks DNA synthesis, AR, NGF and VEGF (but not cytokine) production

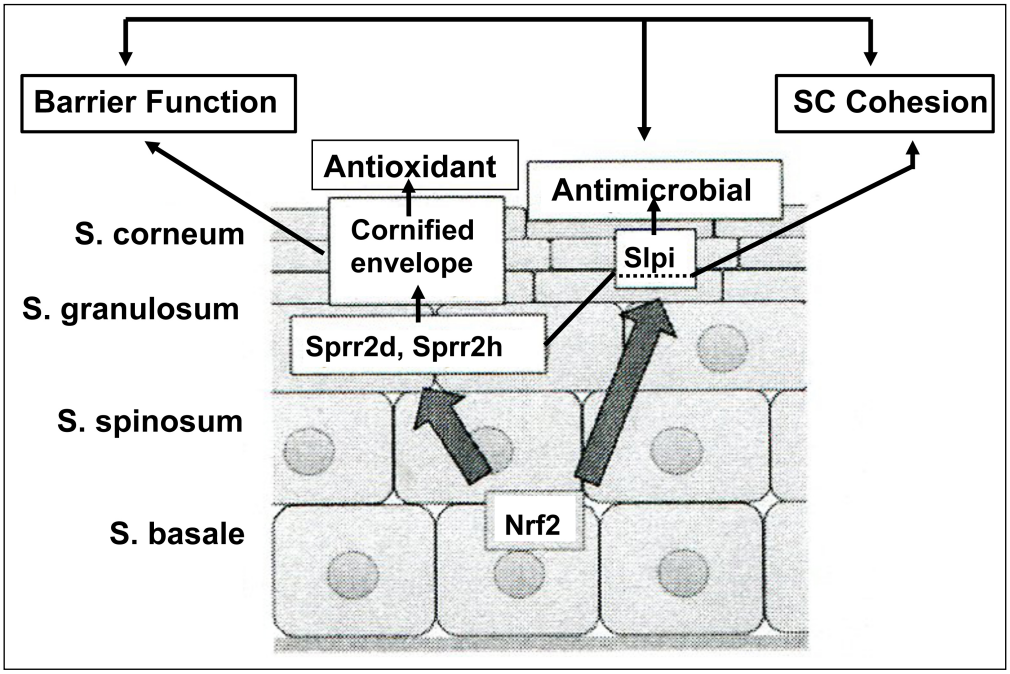

Figure 7: Nrf2 regulates not only antioxidant defense, but also barrier function and antimicrobial defense through increased cornified envelopes and secretory leukocyte protease inhibitor [Slpi] expression. (Modified from: Schäfer et al., 2012).
Abbreviations: Nrf2, nuclear factor erythroid 2-related factor 2; Slpi, secretory leukocyte protease inhibitor; Sprr, small proline-rich protein.

of the wound healing sequence, which includes re-epithelialization followed by stratification of epidermis into a functional stratum corneum. They need only observe that occlusion with vapour-permeable wraps delays wound healing, while applications of vapour-permeable wraps stimulate all of the processes described above, including re-epithelialization.

We view one of our standard laboratory models; i.e., sequential tape stripping, as a type of superficial wound. Tape stripping (no different than either detergent or solvent wipes) produces a defect in the permeability barrier, and all three of these unrelated, acute perturbations stimulate an identical series of metabolic responses in the underlying epidermis that rapidly re-establishes permeability barrier homeostasis in a predictable sequence, and with characteristic kinetics (Table 3). This approach (which we term the cutaneous stress test or 'treadmill of the skin') can be deployed to identify specific metabolic responses that bring about re-establishment of barrier homeostasis. The earliest response to acute barrier perturbations is the immediate secretion (within 15-20 minutes) of much of the pre-formed pool of lamellar bodies from cells of the outer stratum granulosum (SG) (*Elias* et al., 1998). After exteriorizing their cargo of lamellar body contents, these outermost SG cornify; i.e., they undergo physiologic apoptosis (*Demerjian* et al., 2008), followed immediately by the apical migration of subjacent SG cells (*Elias* et al., 1998) (Table 3).

Yet, barrier perturbations also stimulate injury responses that may be unrelated to the restoration of barrier

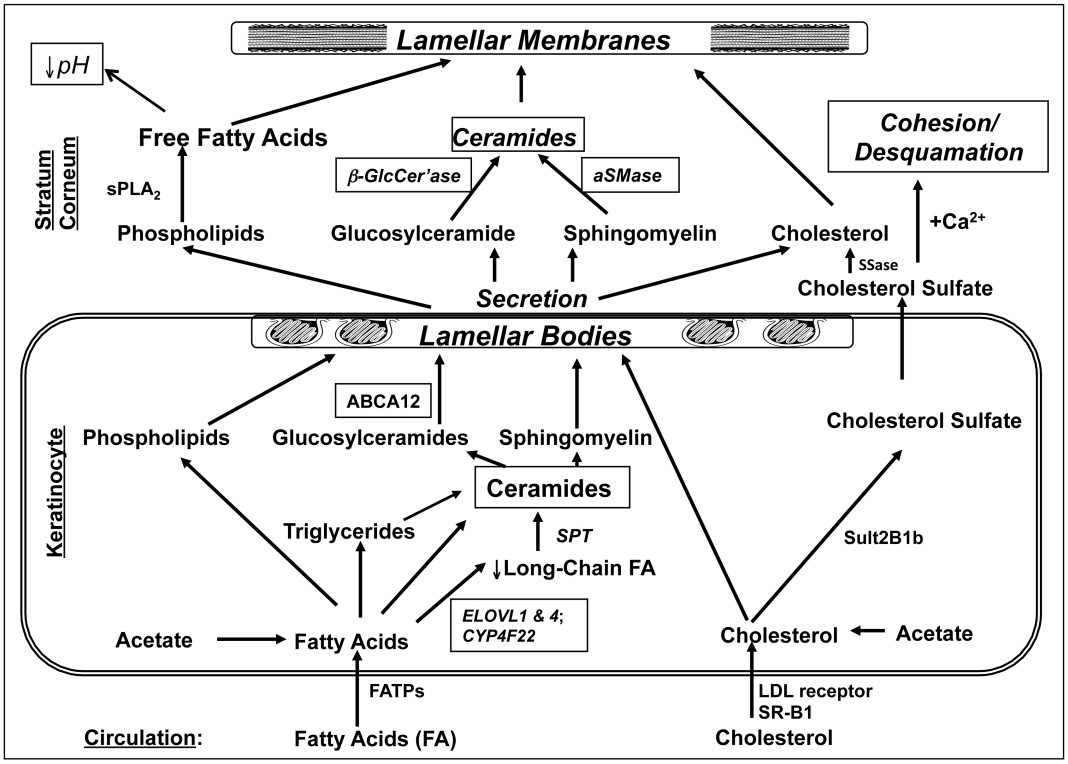

Figure 8: Lipid metabolic events leading to normal barrier formation. (Modified from: Feingold and Elias, 2014).
Abbreviations: β-GlcCer'ase, β-glucocerebrosidase; aSMase, acidic sphingomyelinase; FATPs, fatty acid transport proteins; sPLA2, secretory phospholipase A2; SSase, steriod sulfatase.

function. To distinguish between these two events, one can artificially restore barrier function with a vapour-impermeable wrap, such as a Latex® glove or a sheet of Saran® wrap. By sending a 'message' that the barrier function is now normal, these forms of occlusion shut down metabolic events that are solely directed at restoring barrier function, including virtually all of the changes shown in Figure 8 and Table 3 (*Feingold*, 2009). Yet, some responses, such as increased cytokine production (see below), are not blocked by occlusion. These could be dual-purpose; i.e., signals of both barrier homeostasis and an injury response. Finally, it should be noted that the same 'stress test' approach has allowed us to identify abnormalities in barrier function in:

i) developmental (neonatal and aged skin) settings (*Ghadially* et al., 1995; *Choi* et al., 2007);
ii) human populations, subjected to psychological stress (*Garg* et al., 2001) or endowed with different pigment types (Reed et al., 1995; Gunathilake et al., 2009); and
iii) disease settings (*Schmuth* et al., 2007; *Elias* et al., 2008).

Finally, the stress test led to the development of new generations of 'barrier repair' therapeutics (*Man* et al., 1996), as well as novel metabolically-based, drug delivery technologies (*Menon* and *Elias*, 2000).

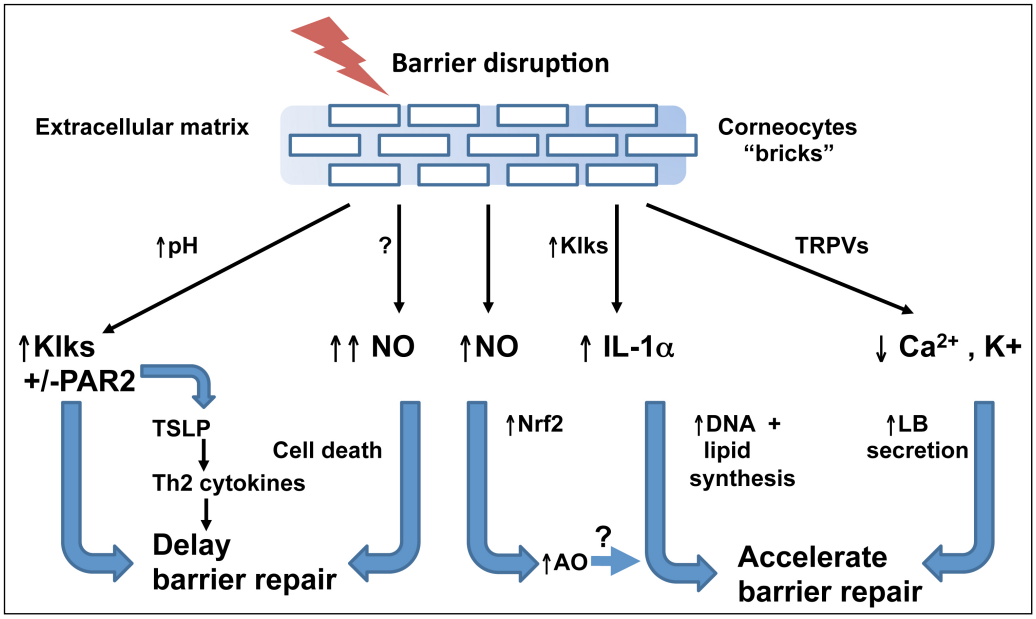

Figure 9: Regulation of permeability barrier repair. Examples of how disruption of the permeability barrier results in signals that can either accelerate or delay barrier repair. (Modified from: Feingold et al., 2007).
Abbreviations: AO, antioxidants; Ca, calcium;l IL-1α, interleukin-1alpha; K, potassium; Klks, kallikreins; LB, lamellar bodies; NO, nitric oxide; Nrf2; nuclear factor erythroid 2-related factor 2; PAR2, proteinase-activated receptor; TSLP, thymic stromal lymphopoietin.

SIGNALS OF BARRIER HOMEOSTASIS

It still is only partially understood how perturbations of the outer skin surface signal the underlying nucleated layers to initiate the metabolic responses that restore permeability barrier homeostasis. To date, several extracellular signalling mechanisms have been identified that are known to stimulate a broad array of metabolic responses in the underlying epidermis (Figure 9, Table 4). But it also should be noted that external perturbations 'turn on' intracellular signalling mechanisms (second messengers) that also regulate these metabolic responses (Table 5).

These include the 'liposensor' subclass of nuclear hormone receptors, PPARα, PPARβ/δ, PPARγ and LXR, which regulate the transcription of several genes that are critical for epidermal differentiation and lipid production (*Schmuth* et al., 2008) (Figure 10). Also carefully studied are sterol regulatory element binding proteins (SREBPs) that modulate epidermal sterol and triacylglyceride synthesis (*Harris* et al., 1998). Then, barrier disruption stimulates hyaluronic acid production which, depending upon fragment size, regulates epidermal proliferation, differentiation and cholesterol synthesis (*Bourguignon* et al., 2006), nitric oxide (NO) production, and endoplasmic reticulum (ER) stress responses. It should be noted, however, that egregious external insults result in cell death or apoptosis (Figure 9), by one or more of these mechanisms (*Park*et al., 2011). In addition, the sulphated sterol, cholesterol sulphate,

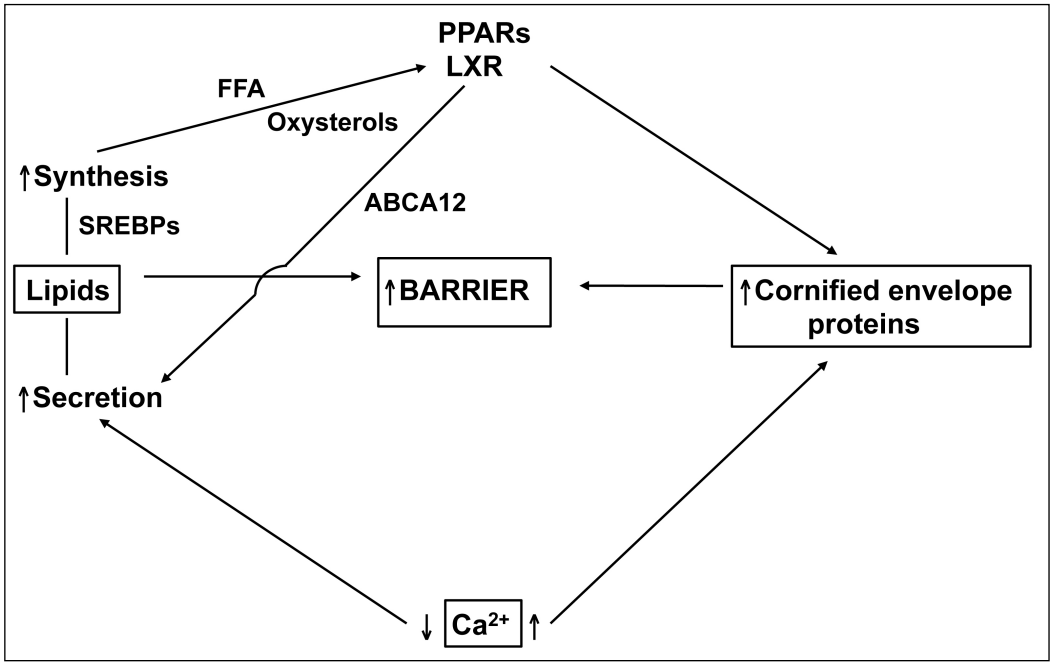

Figure 10: Speculative diagram that illustrates coordinate regulation of epidermal barrier homeostasis by changes in calcium, and activation of the liposensor sub-class of class-II nuclear hormone receptors (modified from: Elias and Feingold, 2001)
Abbreviations: ABCA12, ATP-binding cassette transporter A12; FFA, free fatty acid; LXR, liver-x receptor; PPAR, peroxisome proliferator-activated receptor; SREBP, sterol response element binding transcription factor.

which is generated late in epidermal differentiation, is a potent transcriptional regulator of epidermal differentiation (*Hanley* et al., 2001). Yet, new signalling networks, both extra and intracellular, that link external perturbations to metabolic response in the underlying epidermis continue to be discovered (Tables 4 and 5). While several of these signals broadly regulate epidermal differentiation and/or lipid production, perhaps in a redundant or overlapping fashion, others instead modulate more discrete metabolic pathways within the epidermis.

It is important to distinguish whether these signalling mechanisms represent purely homeostatic, or in part, injury-mediated responses. The 'gold standard' applies again - do these signals deploy following acute external perturbations, even when the barrier is immediately restored by occlusion (see also above)? Several cytokines, but not the growth factors NGF, AR and VEGF, continue to up-regulate, even in the face of barrier restoration by occlusion, indicating that they could represent, at least in part responses to injury, rather than purely homeostatic mechanisms alone (Tables 4 and 5).

Table 4: Signals that regulate permeability barrier homeostasis

Extracellular modulations:	Sensor	Signal	Homeostatic responses	Potential pathogenic signal*
External humidity:	TRPV4, TRPM8	Ca^{2+}	↑Lamellar body secretion; ↓-↑Epidermal differentiation	No
Osmolar stress (cell volume):	AQPs, UTs, TauT, TonEBP, Na-dependent myo-inosital transporter	Ca^{2+} BGT1 PSLC5A3	↑Epidermal differentiation; ↑Lipid synthesis; ↑AMP production, anti-apoptotic (↑HSPs)	No
Acidity:	TRPV1	Ca^{2+}	↑NHE1 + ? others	Yes (via SP→PAR2)
Barrier disruption:	ΔpH	Cytokines; Klk→PAR2	↑Epidermal proliferation; ↑lipid synthesis/secretion (IL-1α); terminal differentiation	Yes (inflammation, pruritis)
Heat:	TRPV3, Ca^{2+}	?Ca^{2+}	?	No
Injury (wounding):	TLR3	ncRNA	↑Lipid synthesis + secretion; ↑innate immunity	Yes (inflammation)

*Fail to downregulate with artificial barrier restoration following acute perturbation.
Abbreviations: AQP, aquaporin; BGT1, betaine/gamma-amino-n-butyric acid transporter 1; Chol, cholesterol; HSP, heat shock protein; Klk, kallikrein; ncRNA, non-coding RNA; NHE1, sodium-hydrogen antiporter 1; PAR2, protease-activator receptor 2; TauT, taurine transporter; TLR3, toll-like receptor 3; TonEBP, tonicity enhancer binding protein; TRPM8, transient receptor potential melastatin-8 ; TRPV, transient receptor potential vanilloid.

ACKNOWLEDGMENTS

Research reported in this publication was supported by the National Institute of Arthritis and Musculoskeletal and Skin Diseases of the National Institutes of Health under Award Number AR061106 and AR019098, administered by the Northern California Institute for Research and Education, with additional resources provided by the Veterans Affairs Medical Center, Merit Review, San Francisco, CA.
This content is solely the responsibility of the authors and does not necessarily represent the official views of either the National Institutes of Health or the Department of Veterans Affairs.
Ms. Joan Wakefield provided superb editorial assistance.

Table 5: Second messengers of permeability barrier homeostasis

External perturbations	Signal	Homeostatic response	Potential pathologic signal
Barrier disruption, UV-B, oxidative stress	ER stress→↑Cer→↑S1P	↑epidermal CAMP (LL-37) production	Cell death, apoptosis, if excessive
—	↑Cholesterol sulfate→ PKGη, AP1 elements	↑epidermal differentiation	Abnormal SC cohesion & barrier function
Barrier disruption	SREBPs	↑sterol, triacylglycerol synthesis	No
Barrier disruption	PPARs, LXR	↑epidermal differentiation, lipid synthesis	No
Barrier disruption	HA→CD44 receptor	↑epidermal proliferation, differentiation, sterol synthesis	No
Barrier disruption→ oxidative stress	nitric oxide→↑cGMP, ↑Ca^{2+}	↓Barrier repair	Inflammation; Apoptosis

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