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Inflammasome activation of IL-1 family mediators in response to cutaneous photodamage†

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Abstract

Although keratinocytes are relatively resistant to ultraviolet radiation (UVR) induced damage, repeated UVR exposure result in accumulated DNA mutations that can lead to epidermal malignancies. Keratinocytes play a central role in elaborating innate responses that lead to inflammation and influence the generation of adaptive immune responses in skin. Apart from the minor cellular constituents of the epidermis, specifically Langerhans cells and melanocytes, keratinocytes are the major source of cytokines. UVR exposure stimulates keratinocytes to secrete abundant pro-inflammatory IL-1-family proteins, IL-1 α , IL-1 β , IL-18 and IL-33. Normal skin contains only low levels of inactive precursor forms of IL-1 β and IL-18, which require caspase 1-mediated proteolysis for their maturation and secretion. However, caspase-1 activation is not constitutive, but depends on the UV-induced formation of an active inflammasome complex. IL-1 family cytokines can induce a secondary cascade of mediators and cytokines from keratinocytes and other cells resulting in wide range of innate processes including infiltration of inflammatory leukocytes, induction of immunosuppression, DNA repair or apoptosis. Thus, the ability of keratinocytes to produce a wide repertoire of proinflammatory cytokines can influence the immune response locally as well as systematically, and alter the host response to photodamaged cells.

We will highlight differential roles played by each IL-1 family molecule generated by UV-damaged keratinocytes, and reveal their complementary influences in modulating acute inflammatory and immunological events that follow cutaneous UV exposure.

INTRODUCTION

The epidermal barrier

Keratinocytes make up to 95% of the total cells in the epidermis, and as such, they function as the major protectors of the body by forming an impenetrable boundary against the entry of foreign and infectious agents through the skin. The stratified keratinocyte differentiation

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program changes as daughter cells move progressively from the germinative basal layer (*stratum basale*) towards the skin surface. They form well-defined layers of non-proliferative cells as they traverse the thick *stratum spinosum* layer to reach the outermost living cell layer of the *stratum granulosum*, where they form the epidermal tight junction barrier. These cells lose their nuclei and form the keratinized corneocytes of the *stratum corneum*, where they are finally shed. This differentiation program has evolved to maintain barrier integrity in the face of chronic exposure to the sun's toxic and mutagenic ultraviolet rays.

UVR and photodamage mechanisms

UVR is a potent complete carcinogen since it acts as both an initiator and promoter of cancer. Most (94–97%) of the UVR that reaches the earth is comprised of UVA (320–400 nm), while UVB (290–320 nm) makes up the remainder (3–6%). Negligible amounts of mutagenic UVC (200–290 nm) reach the earth's surface by virtue of absorption by the ozone layer in the stratosphere (1). Like the earth's ozone layer, the skin's *stratum corneum* can effectively absorb around 90% of UVC wavelengths, hence only minute amounts might reach the basal layer in human skin in areas where ozone is diminished. However, a significant proportion of the UVA and UVB spectrum can penetrate the epidermal layers and reach the dermis, thus effecting differentiated cells of *stratum spinosum* (or spinous layer), dividing cells in the *stratum basale* (or basal layer), as well as cellular constituents in the dermis (2). While UVB makes up a minor fraction (< 5%) of the sun's irradiation on earth, it is well characterized as the most potent mutagenic component; however, recent investigation on the role of UVA, which makes up the majority (95%) of the sun's radiation and used by commercial tanning salons, has raised awareness of its importance in developing melanoma and non-melanoma skin cancers (3–7). UVR photodamage occurs through direct and indirect mechanisms. Direct absorption of UV energy drives biochemical reactions that result in molecular changes and production of reactive oxygen species (ROS). UVA wavelengths, which penetrate deeply into skin, induce formation of ROS, which act as mediators of indirect photodamage resulting in oxidative stress and adduct formation of biomolecules. DNA has absorption maxima in UV wavelength range and is therefore a direct target of UV damage, forming cyclobutane pyrimidine dimers (CPDs) and 6-4 photoproducts (6-4PPs) by UVB and UVA, respectively. These photoproducts interfere with biological processes and induce signals that inform cellular decisions for entry into one of two main pathways: cell survival through initiation of cellular repair processes or cell death by apoptosis (8). At low UV doses, homeostatic apoptosis occurs in the absence of concomitant production of pro-inflammatory signals, and can facilitate immune tolerance to immunogenic UV “altered-self” components (9). With increasing UV dose, damaged cells will produce danger signals mediated by stress related host “alarmins” and pro-inflammatory cytokines that activate neighboring healthy cells to secrete antimicrobial peptides and support wound healing processes to protect epidermal barrier integrity. Thus, keratinocytes undergo an immunogenic inflammatory type of cell death that has mainly been described for macrophages and antigen presenting cells and is termed pyroptosis to distinguish it from non-inflammatory apoptosis.

Photo damage-induced inflammasome activation

One of the earliest responses made by viable keratinocytes in response to UV damage is the formation of the inflammasome within the cytoplasm. This complex assembly of proteins is made up, in part, by a highly conserved family of intracellular receptors termed nucleotide binding and oligomerization domain (NOD)-like receptors (NLRs). The type of inflammasome that is assembled is specific for a set of cell damage associated molecular patterns (DAMPs) as well as pathogen associated microbial patterns (PAMP), which are recognized by different NLR subtypes. Inactive procaspase-1 molecules are recruited to the inflammasome complex where inter-molecular processing events required for caspase-1

activation occurs. Caspase-1-mediated cleavage is the limiting step for processing IL-1 related molecules IL-1 β and IL-18, into their secreted active forms. These secreted cytokines induce autocrine and juxtocrine signals of the skin damage status throughout the adjacent epidermal and dermal layers. Similar to IL-1 α , the newest member of the IL-1 family, IL-33, does not require direct caspase-1 processing for its activation, but depends on inflammasome-associated unconventional mechanisms for its secretion (10). Interestingly, IL-33 is, in fact, susceptible to inactivation by direct caspase-1 cleavage (11). Thus, the mechanisms that regulate inflammasome assembly and subsequent activation of these IL-1 family molecules play an important role in influencing keratinocyte fate with regard to initiating cell survival or cell death pathways. Importantly, IL-1 family members have been shown to promote alternate pathways for T cell development; therefore, understanding how photodamage induces inflammasome activation will also provide insight into mechanisms that contribute to induction of tolerance, promotion of chronic inflammation, and ultimately progression of neoplasias.

In this review, we will focus on what is known about mechanisms that govern the expression and activation of these IL-1 family keratinocyte-derived cytokines, with respect to photodamage responses and their targeted activities favoring survival or death. Further, we will examine their differential roles in modulating the cutaneous adaptive immune response with respect to influencing the induction and function of cutaneous effector or regulatory T cell subsets.

INDUCTIVE SIGNALS FOR PRO-INFLAMMATORY MEDIATORS: CROSS-TALK BETWEEN NLR AND TLR SIGNALING

Source of IL-1 β differs in mouse and human epidermis

Following exposure to UVR, keratinocytes secrete increased levels of activated IL-1 family proteins, IL-1 β , IL-18 as well as IL-33 and IL-1 α , which depend on inflammasome activation (12, 13). In human skin, keratinocytes constitutively produce low levels of inactive precursors pro-IL-1 β and pro-IL-18, which accumulate in the cell cytoplasm without active secretion. Like macrophages and dendritic cells, keratinocytes require stimulation of both TLR and NLR receptors for secretion of mature IL-1 β and IL-18 (14). This supports the notion that keratinocytes are immunologically active components of the skin-associated immune system and the cytokines they produce in response to UV damage may counter or promote UV-induced immune suppression (15). However, in murine skin, IL-1 β production cannot be detected in keratinocytes following UVR or other inflammatory stimuli *in vivo*. Instead, infiltrating bone marrow-derived cells and to a lesser extent resident Langerhans cells (LCs) are responsible for generating IL-1 β . This has been demonstrated in studies using radiation bone-marrow chimeras of wild type and *IL-1 β ^{-/-}* mice (16), and more recently verified in elegant imaging studies using IL-1 β promoter-driven DsRED fluorescent protein reporter mice. In both cases, investigators used hapten stimulation, so the precise nature of IL-1 β secreting cells in mouse skin following UVR has yet to be defined. (17). LCs are key players in early UV-induced photodamage responses which have been reviewed by us and others previously (18, 19). LCs are found suprabasally in the epidermis and can sense UV damage directly through ROS formation and DNA damage, or indirectly through recognition of apoptotic bodies and soluble mediators derived from damaged keratinocytes (18). In addition, IL-1 mediators affect resident dermal leukocytes, including dermal dendritic cells (dDC), mast cells, macrophages and endothelial cells, impacting both local and systemic immune processes. The differences between human and murine keratinocyte-mediated signal amplification of photodamage responses is not, yet, fully characterized. Therefore, as indicated above, the source of cutaneous IL-1 β production in

response to UV must be considered when comparing human and murine photodamage response pathways.

Sterile inflammation is induced by alarmins

UV-induced damage causes physical and metabolic disruption of epidermal cells, resulting in the accumulation and release of highly conserved endogenous cellular constituents termed alarmins. Alarmins are extracellular DAMPS that induce “sterile” inflammation, as these endogenous molecules are perceived by damaged and undamaged cells as potent “danger” signals that indicate the potential loss of epidermal barrier integrity. IL-1 α and IL-33 are two such alarmins. Both are normally sequestered in the nucleus, where they are thought to modulate transcriptional activity (20, 21). They are secreted or passively released from cells under conditions that induce inflammasome-dependent unconventional secretion (since both are leaderless and neither contains caspase-1 processing sites) or necrosis, respectively. Other alarmins that mediated photodamage responses are highlighted in the following sections.

Toll-like receptors

Because of the varying range of cross-talk between toll-like receptors (TLRs) and NLRs with respect to inflammasome complex formation (22), we will highlight what is known about the role of TLRs in inflammasome activation generally, and specifically in skin photodamage responses. TLRs are transmembrane receptors that detect external danger signals chiefly by recognition of microbial products. They comprise a subset of pattern-recognition receptors (PRRs), which recognize certain invariant microbial motifs called pathogen-associated molecular patterns (PAMPs). TLRs are expressed by cells of the innate immune system, such as macrophages, monocytes, dendritic cells, neutrophils, and epithelial cells, as well as cells of the adaptive immune system (23) and have been reviewed in detail (24, 25). Up to 13 TLRs have been identified in mice and humans, most of which are well characterized (26). Human keratinocytes express TLRs 1–10 except 7 and 8 (27). Their activation in keratinocytes induces production of inflammatory cytokines and chemokines (27–29). How UVR activates TLRs is not fully understood, but many reports indicate that they play a role in UV-induced inflammatory processes in mice and humans. It is likely that in addition to recognition of microbial PAMPs, TLRs also bind endogenous cellular constituents released upon UV damage, which act as alarmins (defined in the section above) and recognized as DAMPs. For example HSP60, HSP70, gp96 (a HSP90 family protein), fibrinogen, surfactant protein-A, extra domain A of fibronectin, heparan sulfate, oligosaccharide of hyaluronan (soluble hyaluronan), β -defensins, high-mobility group box 1 (HMGB1) protein, and mRNA have been shown to ligate TLRs (30). All these endogenous ligands have been found to stimulate the release of cytokines IL-1 β , TNF α , IL-12 and IL-6. In TLR 2, 4 or 9 deficient mice cytokine secretion is severely limited, supporting the requirement of a two-step mechanism for IL-1 β maturation (30, 31). The adapter molecule MyD88 recruited by all TLRs, except TLR3, activates NF- κ B, and has been shown to be upregulated after UV stimulation. In MyD88 knockout mice or MyD88 null human keratinocytes the UVB stimulation of these cytokines is severely impaired as compared to wild-type counterparts (32, 33). TLR-mediated induction of IL-1 β and IL-18 transcription is required to accumulate sufficient levels of precursor proteins in the cytoplasm, while NLR activated inflammasome assembly is required for caspase-1 activation and subsequent caspase-1-dependent maturation of those cytoplasmic precursors. What is important here is that upon UV stimulation these endogenous alarmins (or DAMPs) are induced in keratinocytes and other cell types of the epidermis like LCs and melanocytes (34–39). Upon UVB exposure TLRs are activated which stimulate pro-IL-1 β synthesis that is further processed by the inflammasome machinery to generate active IL-1 β cytokine, one of the most important pro-inflammatory cytokines. Like macrophages, keratinocytes also require

inflammasomes for active IL-1 secretion (40, 41); however the amount of active IL-1 family cytokines seems to be dependent on the activation of TLRs. The extent to which UVR stimulates different types of TLRs has been shown to influence gene activation programs. For example TLR3 or TLR7 activation in conjunction with UVR produces IL-12, but TLR2 or TLR4 activation in conjunction with UVR induces IL-10, (42). IL-10 inhibits IL-1 β production (43) and inhibits IL-18 function by inhibiting the IL-18 induced cytokines like IFN- γ (44). In contrast, IL-12 activates both IL-1 β and IL-18 (discussed in detail below). Thus, differential TLR activation during UVR can lead to opposing cytokine profiles with respect to promoting or inhibiting immune responses.

NOD-like receptors

The NOD-like receptors have been reviewed in great detail by many investigators (23, 45, 46). Various autoinflammatory disorders have let to discover the role of NLRs and describing the mechanism and regulation of IL-1 β (46). NLR family is composed of 22 human genes and in mice there are additional genes making the total to 34 (47). The NLR families are defined by their characteristic domain structures in addition to the proteins they bind to form an active complex. A subset of these are expressed by keratinocytes and myeloid cells, and are diagramed in Figure 1A {and described in detail elsewhere (23)}. The NLR family has a characteristic central nucleotide-binding and oligomerization (NACHT) domain, with flanking leucine-rich repeats (LRR) on C-terminus that can sense ligands and caspase-binding domain (CARD) on the N-terminus that is important for downstream signaling. All the NLR members have the common NACHT domain, which utilize ATP for activation of signaling complex. The three NLR subfamilies have been characterized on the basis of phylogenetic analysis of their NACHT domain. They are the NOD subfamily comprising of (NOD1-2, NOD3/NLRC3, NOD4/NLRC5, NOD5/NLRX1 and CIITA), the NLRP or NALP subfamily comprising of (NLRP1-14) and the IPAF subfamily, comprising of IPAF (NLRC4) and NAIP. Here we are going to describe the role of two important members of the first subfamily (NODs 1 and 2) and then NLRP1 and NLRP3 receptors. The latter two form the important inflammasome complexes in UV-induced inflammatory processes of keratinocytes and are the means of activating proinflammatory cytokines IL-1, IL-18 and possibly IL-33 (Figure 1B). Although little is known about the role of NOD1 and NOD2 in keratinocyte photodamage responses, it is known that both are expressed by the keratinocytes and, like TLRs, they not only serve as sentinels against microbes but also regulate pro-inflammatory pathways (48) independent of TLRs. NOD1 recognizes a component found in many Gram-negative and various Gram-positive bacteria termed meso-diaminopimelic acid (iE-DAP) while NOD2 recognizes muramyl dipeptide (MDP), a component of peptidoglycan mainly found in Gram-negative bacteria (49, 50). Further NLRP1 and NLRP3 are also activated by various bacterial and fungal components (23). Most of the DAMPs that activate TLRs can also activate NLRs like NLRP3 (51). Further, reactive oxygen species (ROS), hyaluronan (HA), ATP and uric acid can also activate NLRs (52–54). Activated by cellular stress and injury they induce secretion of proinflammatory cytokines including IL-12, a very important cytokine in DNA repair mechanisms (55). The role of NOD1 and NOD2 in UV-induced processes is suggested by studies done on human corneal epithelial cells (HCE-T), where increased mRNA levels of NOD1 and NOD2 was detected following UVB treatment, in addition to increased expression of IL-12, IL-6, IL-1 β and various other proinflammatory cytokines (56). While human corneal epithelial cells differ from skin keratinocytes in keratin-specific expression, expressing K3/K12 in place of K5/K14 or K10 for epidermal keratinocytes (57) they may behave similarly to cutaneous keratinocytes with respect to photodamage response mechanisms.

INFLAMMASOMES

NLRP1

The NLRP1 (NACHT, LRR and PYD domains-containing protein 1) inflammasome is the first discovered inflammasome complex that not only activates caspase 1 but also caspase 5. The NLRP1 structure has been described in detail elsewhere (45). NLRP1 inflammasome complex consisting of caspase 1, caspase 5, ASC (apoptosis-associated speck-like protein) and NLRP1 (58). The mechanism of NLRP1 activation is still not well understood, however all the crucial NALP1 inflammasome components are expressed by primary human keratinocytes (41, 59). NLRP1 is expressed constitutively in keratinocytes, and therefore is likely to be the first NLR to respond to alarmins induced by UV-damage. NLRP1 activates both caspase 1 and caspase 5 – both of which are able to activate IL-1 β in human cells (58), however knockdown of caspase 5 in keratinocytes did not affect the secretion of IL-1 β indicating that caspase 5 may not be required for UV-induced inflammatory processes in keratinocytes. Thus, different NLR components can be activated by different stimuli and/or in different cell types (13). Investigation of NLRP1 activation *in vitro* indicates that it forms self-oligomers and binds with procaspase 1, NTP (ribonucleotide- triphosphate) as well as ASC (apoptosis-associated speck-like protein) (13). However in case of mouse NLRP1, ASC is unable to bind, owing to the deficiency in PYD (pyrin domain) that is present in the human homologue. As a result ASC binding can be bypassed for caspase activation (60). In resting cells anti-apoptotic proteins Bcl-2 and Bcl-XL are bound to NLRP1, thus suppressing caspase 1 activation and IL-1 β secretion. In macrophages it was shown that muramyl dipeptide (MDP) releases the anti-apoptotic proteins from NLRP1 and thus activates the inflammasome assembly (61). UVB irradiated keratinocytes show decreased levels of these anti-apoptotic proteins (Bcl-2 and Bcl-XL) and suggests a possible mechanism by which UV-induced NLRP1 activation may occur (62). In cell free extracts, it was shown that NLRP1 alone was able to form the inflammasome complex and activate caspase 1. However, what happens *in vivo* is not well understood. It has been shown that NOD2 interacts with NLRP1 *in vitro* and activates NF- κ B thus providing a superior signal for IL-1 β production (45).

NLRP3

The NLRP3 inflammasome is well characterized among the inflammasome complexes. It consists of the NLRP3, the ASC adaptor, and caspase-1. NLRP3 is activated by a wide range of pathogens, their components, toxins and many chemical irritants (63–66). NLRP3 has been shown to require ATP as its NBD domain possesses ATPase activity (67). Interestingly many toxins that stimulate NLRP3 inflammasome happen to induce pore formation leading to potassium efflux from the cells which is ATP driven, an important condition for NLRP3 activation (45). ATP is an important stress signal and was the first to be described (68). Under normal conditions the extracellular levels of ATP are low. Upon injury, dead cells release large quantities of ATP in the extracellular space. ATP has a high intracellular concentration and is kept low in the extracellular space by the activity of ATPases. ATP is released from dying cells and hence can be a fitting “danger signal” or “alarmin”. Indeed stimulated T-cells and monocytes release ATP and IL-1 β through activation of NLRP3 (69, 70). ATP release into the cytoplasm might be one of the mechanisms by which UV-irradiated keratinocytes activate formation of the NLRP3 inflammasome complex. This ATP release might be a mechanism to influence other cells to activate their inflammasome machinery as well. It is understood that UVB radiation induces rapid extracellular ATP release from HaCaT keratinocytes (71). In contact in the hypersensitivity model it has been shown that ATP release from keratinocytes also stimulates Langerhans cells (72) to produce inflammatory cytokines. Mizumoto *et al* did not report the release of IL-1 β in response to the ATP treatment, various others have shown that

LPS can stimulate macrophages which is dependent on ATP-mediated K^+ efflux from cells triggered by the P2X₇ receptor (73). Further, when the macrophages that lacked P2X₇ were stimulated with LPS, they failed to secrete mature IL-1 β . It is interesting that the keratinocytes also express many P2X receptors and in presence of ATP or UVB- irradiation there was increased expression of purinergic P2X₁, P2X₂, P2X₃, and P2X₇ receptors with the inward flux of calcium (74). Mice that are deficient in P2X₇ receptors are unable to mount an IL-1 β NLRP3 response again emphasizing the role ATP plays in NLRP3 activation (75). Though the efflux of K^+ seems essential for the activation of NLRP3 in macrophages (63, 76), in keratinocytes influx of Ca^{2+} seems to do the job. No other ionophore was able to activate NLRP3 except Ca^{2+} and UV-induced IL-1 β secretion was suppressed upon Ca^{2+} chelation (12). This might be true as in keratinocytes Ca^{2+} regulates growth and differentiation. The authors found that K^+ efflux did not activate NLRP3 in this system. Interestingly the cytoplasmic level of K^+ should be lower than half the concentration of normal levels to activate inflammasomes. The efflux of K^+ might not have been large enough as required for inflammasome activation. However, the influx of Ca^{2+} and efflux of K^+ at the same time might also be necessary for the activation of inflammasome in keratinocytes. It has been shown that ATP-induced changes mediated via the activation of purinergic P2X₇ nucleotide receptor facilitates the rapid influx of extracellular Na^+ and Ca^{2+} and efflux of intracellular K^+ (77). Moreover, studies in various other models of HEK-293, THP-1 and Bac1.2 macrophages point towards the importance of Ca^{2+} in activation of caspase 1 and IL-1 β secretion. Further with the efflux of intracellular K^+ there is increased acceleration in procaspase-1 activation and thus enhanced IL-1 β production (78, 79).

Some reports also suggest that HA a structural component of the extracellular matrix, thought to be another danger signal of the cell, activates the NLRP3 inflammasome (53). Indeed it has been shown that UVB radiation caused significant increases of HA in the basal layer of the epidermis(80). And in neonatal foreskin keratinocyte cultures, increased expression of hyaluronan synthase, in addition to IL-1 β secretion, was induced by UVB irradiation (81).

NLRP3 activation is also induced by the production of ROS (82). It is well appreciated that UV-induced ROS production in keratinocytes leads to DNA damage and ultimately skin cancer (83). The production of ROS in skin can take place either directly through photochemical interactions between UV and chromophores or through the activation of the epidermal growth factor receptor (EGF-R), which leads to NADP oxidase activation and delayed ROS production. Suppression of the NADP subunit p22 inhibits inflammasome activation (82). ROS can also be generated by mitochondria. Alum or nigericin treatment in macrophages leads to the production of mitochondrial ROS. It has been shown that the induction of mitochondrial ROS induces the recruitment of NLRP3 to mitochondrial-associated ER membranes. NLRP3 then recruits ASC and finally pro-caspase-1. This results in the activation of caspase-1 and the production of IL-1 β (84, 85). Jurg Tschopp's laboratory recently identified a relevant ligand that is induced by ROS, which activates the NLRP3 inflammasome. They observed that in resting cells the anti-oxidant protein thioredoxin (TXN) is normally associated with its binding partner, the thioredoxin interacting protein (TXNIP). In the presence of oxidative stress, ROS molecules bind TXN, releasing TXNIP, which then can directly bind the LRR domain of NLRP3 and induce inflammasome assembly (86) (Figure 2). Regardless of the role that ROS has been shown to play in NLRP3 activation, still more evidence is needed to clarify why ROS activates NLRP3 in some cases and inhibits it in others and why many other ROS promoting agents do not employ the NLRP3 inflammasome (23). Further, UV-induced immunosuppression seems to depend on ROS production, since none was generated in mice treated with ROS inhibitors (87). These observations suggest that UVR induced ROS might promote

production of inflammatory mediators that favor production of immunosuppressive cytokines rather than immune enhancing ones (Figure 3).

AIM2

Another inflammasome has recently been identified that can sense double-stranded DNA (dsDNA) and is a non-NLR member that activates IL-1 β maturation in a caspase-1 dependent manner (88, 89). Unlike the NLR-family of inflammasomes, the AIM2 (Absent In Melanoma 2) inflammasome oligomerizes by crowding on numerous binding sites on the ligand to which AIM2 binds, through its C-terminal HIN domain (23, 88). (Figure 1A) The final complex assembly is comprised of AIM2, ASC, and caspase-1. AIM2 has demonstrated direct binding of a cytoplasmic DAMP – dsDNA. AIM2 knockdown in human and mouse macrophages severely attenuated inflammasome activation by cytoplasmic DNA, while AIM2 transfection of a cytoplasmic DNA non-responsive cell line conferred responsiveness (88). The AIM2 inflammasome has been found to be present in keratinocytes of psoriatic skin but not in healthy skin; however, normal keratinocytes were shown to release IL-1 β when cultured in the presence of cytosolic DNA (90). It is tempting to speculate that AIM2 may also be activated in UV-damaged cells, since UV irradiation induces DNA fragmentation that is associated with caspase activation and IL-1 β release (12, 91).

INFLAMMASOME-INDUCED CYTOKINES AND THEIR IMPACT ON PHOTODAMAGE RESPONSES

Caspases belong to the family of enzymes known as cysteine proteases, most of which play a central role as mediators of apoptosis. Caspase activation, therefore, is regulated at multiple levels. They are present in the cells as inactive precursors or zymogens. Different caspase families fall into functional categories involved with either regulating apoptosis, by inhibiting or inducing apoptosis - or activating inflammatory pathways, by virtue of their controlled proteolytic cleavage (23). In both human and mice, caspase-1 is categorized as the prototype inflammatory caspase (92). Its activity is tightly regulated by activation of inflammasome complex. Caspase-1 is also expressed in its precursor form, as procaspase-1, and once the inflammasome complex assembled through ASC oligomerization, it is then activated by ASC. As discussed above, and diagrammed in Figure 2, oligomerization is dependent on depletion of intracellular K⁺ through activation of the purinergic receptor P2X₇-induced potassium channel (93). ASC oligomerizes the components of each inflammasome subtype to form a structurally unique molecular platform for processing pro-IL-1 β and pro-IL-18 into their secreted, active forms. Caspase 1 has greater specificity for pro-IL-1 β than for pro-IL-18, and thus favors IL-1 β production in the presence of equivalent precursor stoichiometry (94). Caspase-1 knockout mice showed decreased production of both IL-1 β and IL-18 and reduced inflammation, clearly demonstrating caspase-1 dependent cytokine activation (95). However, caspase-1 is not required for IL-33, as it is constitutively expressed in the nucleus (96), but it is upregulated by UVR and can be passively released into the extracellular space by necrotic cells. The role these cytokines play in UVB induced inflammation and immunosuppression is the subject of ongoing investigation, but it is becoming clear that they have distinct, often non-overlapping activities that critically impact how innate and adaptive responses are elaborated. These simplified relationships are diagrammed in Figure 3.

IL-1 α

While IL-1 α was the first pro-inflammatory cytokine described, its function(s) and its need for caspase-1 processing is still poorly defined. The precursor form is constitutively expressed and retained intracellularly in human and murine keratinocytes as well as in

myeloid cells. It is detected in multiple subcellular locations, depending on the cell type and status. In monocytes, it translocates to the nucleus following LPS stimulation, where it is proposed to promote transcription (20). In resting human monocytes, pro-IL-1 α is endogenously expressed as a myristoylated protein that is anchored to the plasma membrane surface through interaction with a mannose-like receptor, where it retains stimulatory activity. This membrane-associated form has not been reported for keratinocytes. Thus, IL-1 α performs multiple functions: (1) as an intracellular modulator of transcription, (2) as a local immune adjuvant in its membrane-associated form on myeloid cells, and (3) as a secreted, extracellular alarmin produced by epidermal keratinocytes that can act in a long-range fashion to impact innate and adaptive immune responses. While pro-IL-1 α is fully active, its activity can be greatly enhanced following proteolysis by a variety of immune-relevant proteases, such as granzyme B, calpain, elastase and mast cell chymase (97). This model suggests that the secreted pro-IL-1 α protein travels distally in a relatively benign state until it encounters an area of tissue damage where it can be locally targeted for proteolysis and activated as a potent initiator of sterile inflammation. The IL-1 receptor is a heterodimer composed of the IL-1RI binding chain and the shared common chain, IL-1 receptor accessory protein (IL-1RAcP). Both IL-1 α and IL-1 β bind to this receptor, which is ubiquitously expressed on all cells except platelets (98); While expression patterns differ remarkably for IL-1 α and IL-1 β , it seems the extracellular biological activities of these two pro-inflammatory cytokines have not been discernible until recently. Studies have ascribed different biological functions based on their physiologic differences in sub-cellular compartmentalization and expression levels (99). Studies using experimental models of tumor evasion compared the growth of tumor lines transfected with different cytokine isoforms and observed that, under conditions that mimic expression patterns during acute inflammation, high levels of secreted IL-1 β promoted tumor growth while IL-1 α , in its membrane-associated form, promoted tumor regression (100). In keratinocytes, intracellular IL-1 α is normally chromatin bound, but its pro-inflammatory function depends on its extracellular availability. Following UVR of epidermal human and murine keratinocytes, IL-1 α gene expression and protein secretion is upregulated to induce sterile inflammation. Non-inflammatory cell death occurs through apoptosis where nuclear alarmins, such as IL-1 α , remain sequestered within membrane bound apoptotic vesicles. In contrast, necrotic cell death leads to passive release of “secreted” IL-1 α , in its active form (21). While, pro-IL-1 α does not possess a caspase-1 cleavage site, it has been shown to bind caspase-1 and is dependent on caspase-1 for its secretion by “unconventional” mechanisms, which are not well defined. In studies using human monocytic cells, it was shown that secretion, (but not surface membrane translocation), of IL-1 α depended on its binding to IL-1 β during caspase-1 processing and secretion (101). However, it is not clear how IL-1 α is secreted from murine keratinocytes, since endogenous IL-1 β is not expressed in those cells. Comparative analyses are still needed to elucidate differences in expression patterns and functions for IL-1 α and IL-1 β during cutaneous photodamage responses in humans versus mice.

IL-1 β

In general, IL-1 β cannot be detected, or is expressed in its pro-form at very low levels, in resting tissues and cells. The secretion of its active form can be induced in a wide variety of cells including human (but not mouse) keratinocytes and myeloid cell subsets (macrophages, monocytes and dendritic cells) in response to infection, physical injury or photodamage. IL-1 β also plays a role in determining cell fate with regard to modulating pathways of apoptosis, cell proliferation and differentiation (102). It influences nearly all cell types and often works in concert with other cytokines. IL-1 α production in resting and stimulated cultures of human keratinocytes is high compared to IL-1 β levels. Because IL-1 β expression is undetectable in normal human skin, keratinocytes must be “primed” by TLR signals.

Priming is accomplished by TLR signaling through DAMP-mediated stimulation of NF κ B-dependent transcription of pro-cytokine and inflammasome component genes. Only low levels of IL-1 β precursor proteins need to accumulate, in contrast to IL-1 α , as IL-1 β potency is much greater at physiologically low concentrations in vivo. Therefore, the parameters that influence IL-1 β toxicity versus benefits are narrow (102, 103). This might be the reason why the immune system has evolved various mechanisms to strictly regulate its production (46). Elevated levels of IL-1 β has been reported in the sera of people who have been irradiated with UV, bear tumors, or suffer from inflammatory diseases (103–105). At the inflammasome complex, IL-1 β production can be down-regulated by several regulatory proteins, including CARD-only protein (COP), Iceberg, the caspase 1 inhibitor proteinase inhibitor 9 (PI-9), and pyrin, which interfere with caspase recruitment or inhibit caspase-1 activity (46, 106, 107).

Following UVR exposure, human keratinocytes are induced to express and secrete IL-1 β in an inflammasome dependent manner (10). When keratinocytes sustain unrecoverable photodamage, they will undergo apoptosis to eliminate damaged and/or mutated cells to maintain barrier integrity. UVR-induced apoptosis may be triggered by ROS generation or the activation of CD95 receptor (108). However, up regulation of CD95 alone does not account for UV-induced apoptosis, indicating other factors are involved (109). In vitro studies on cultured keratinocytes demonstrate that IL-1 β can protect against TNF- α and CD95 mediated apoptosis, but not UV-induced cell death (109). It is well known that TNF- α is up regulated in keratinocytes after UVB irradiation (110) and IL-1 β can counter that effect. Therefore, IL-1 β might play a prominent role in promoting the survival of mutant clones and hence tumors. Several lines of evidence support this notion. In humans, IL-1 β levels are higher in melanoma patients than in healthy individuals (111). Similarly, in mice bearing UVB induced tumors, increased levels of IL-1 β can be detected in the skin as compared to levels detected in control mice (112). Further studies done in IL-1 β knockout and transgenic mouse models demonstrate a strong correlation of IL-1 β expression with tumor development and progression (113). It is proposed that UVR-induced IL-1 β expression can set up a chronic inflammation microenvironment that promotes tumor progression. However, IL-1 β can also synergize with IL-2 to induce tumor-specific CD8+ cytotoxic T lymphocytes (CTLs) that generate increased IFN- γ and display potent lytic activity (114). The outcome of T-cell activation is dependent on the quantity of IL-1 β secreted; lower quantities promote naïve and memory T-cell activation, while prolonged secretion results in inflammation and tissue damage (115). We have shown that CD8+ T-cells are important in overcoming UVB-immunosuppression and tumor development (116). The regulatory T-cells play an important role in inhibiting the effector T-cell responses and play a pivotal role in UV-induced immune-suppression (116–118). However, IL-1 β has the capacity to inhibit regulatory T-cells (119). Watanabe *et al.* have provided evidence in this regard. They have shown that inflammasome activation results in the secretion of cytokines which favor Th1 and Th17 responses and inhibition of Treg activity and tolerance (120). They have also shown that IL-1 β results in the activation of IL-12 and IL-6 which polarize naïve T-cells to Th1 and Th17 respectively and hence suppress Treg development. Further, IL-17 and IFN- γ decrease the activity of Treg cells (120). Their results indicate that IL-1 β breaks tolerance by favoring the expansion of CD25 negative effector cells and, in contrast to our current understanding, predicts that UV-induced production of IL-1 β should attenuate immunosuppression mechanisms. A wide variety of skin-derived biochemical mediators, cytokines, and other cellular constituents contribute to the mechanism of UV-induced immunosuppression (18). Therefore, cellular responses to IL-1 β signaling will be influenced by the presence of particular UV-induced mediators that inform the direction of immune activation. For example, the profile of IL-12 induced gene activation differs in response to UVR (121). IL-1 β treatment induces many repair genes in keratinocytes in a time dependent manner indicating it may be promoting DNA repair enzyme activity after UVR irradiation

(122). Further, as discussed above, UVR irradiation results in the expression of heat-shock proteins that activate TLRs to secrete cytokines important in DNA repair and apoptosis (122).

IL-1 impact on UV-induced tumor development

The initial secretion of IL-1 β seems to be beneficial for keratinocytes after UV-exposure, however prolonged IL-1 β secretion results in tissue damage that can lead to enhanced inflammation and promote tumor development. IL-33, which is normally localized in the nucleus, will, under conditions of stress, be secreted as a cell associated “alarmin” and likely plays a prominent role in UV-induced inflammatory cell death, a process termed pyroptosis (123). Pyroptosis was originally described in macrophages infected with intracellular microbes as a specialized inflammasome-dependent pathway of programmed cell death that, unlike apoptosis, activates the secretion of alarmins or DAMPs to potentiate proinflammatory signals (124). Interestingly, Feldmeyer *et al* have shown that UV-irradiated keratinocytes mimic macrophages in having the ability to secrete the inflammasome components outside the cell membrane, without inducing cell death or apoptosis (12, 13, 125). While some tumors produce IL-1 β , likely due to their dependence on chronic inflammation for growth (115), others have been shown to expel inflammasome components, thereby reducing IL-1 secretion. Hence, Faustin and Reed argue tumor expulsion of inflammasome components is an immune-evasion mechanism that silences the innate “Incoming Danger” alarm, without killing themselves. The mechanism by which the components are released is still not understood, however it seems likely that mutant keratinocytes after UV exposure might expel inflammasome components to avoid initial inflammasome activation of IL-1 β and subsequent immune surveillance. Further studies need to be done in order to establish how inflammasome activation and tumor-mediated expulsion impacts tumor progression and elicit an immunosuppressive microenvironment.

IL-18: As a DNA repair cytokine

Like IL-1 β , IL-18 is also produced in a pro-form, which is 24kDa and is processed by inflammasomes to its secreted 18 kDa active form. However, unlike IL-1 β , its proform is constitutively synthesized and hence does not need priming (45). IL-18 is expressed by macrophages and in epidermal keratinocytes that make up all layers of the epidermis, except *stratum corneum*. It signals through an 80 kDa IL-18 receptor which is also present in keratinocytes supporting an autocrine activation loop (126). Upon UVR, keratinocytes secrete higher levels of mature IL-18 and it is thought that enhanced IL-18 secretion is dependent on ROS inhibition as ROS decreased IL-18 production (127), indicating that NLRP3 inflammasome might be involved in IL-18 production in keratinocytes. The role of IL-18 in cancer development is controversial. High levels of IL-18 in cancer patients is associated with poor survival, perhaps due to its role in promoting angiogenesis and metastasis (128). On the other hand, IL-18 has the capacity to activate both Th1 and Th2 cell types. It also has antitumor effects that are owed to its activation of NK cells, inhibition of angiogenesis and activation of apoptosis (128–130). It has been shown to inhibit prostate cancer and is also a potent adjuvant in dendritic cell based vaccines exhibiting enhanced antitumor potential irrespective of the route of injection (131). With IL-12, it synergistically enhances IFN- γ production by T-cells and increases cytotoxic activity of NK cells (131).

It has been shown that IL-12 treatment reduces UV-induced immunosuppression upon DNA damage (18, 132). The effects of IL-18 on immunosuppression mimic IL-12, although the two cytokines are structurally different. The DNA repair mechanism of IL-18 is also thought to be similar to IL-12. It is believed to activate nucleotide excision repair enzymes (NER) as NER deficient mice were unable to benefit from the DNA repair effects of IL-18 (55, 132). However, unlike IL-12, IL-18 was unable to inhibit regulatory T-cell development, which is

central in immunosuppressive effects of UV (55). Consistent with its immunogenic properties, keratinocytes cultured in the presence of IL-18 demonstrated enhanced expression of MHC class II and also increased the expression of IFN- γ in T-cells co-cultured with those keratinocytes (133). Further, IL-18 inhibited UV-induced apoptosis in keratinocytes supporting a protective role for this cytokine in promoting keratinocyte recovery(132).

IL-33: An activator of Th2 cells

The newest member of IL-1 family of cytokines, IL-33 exerts its biological function through binding its receptor, composed of the ST2 binding chain, and common IL-1RA beta chain, present on many cells types (134). In epidermis, keratinocytes are reported to constitutively produce IL-33, where its function is thought to be closely related to the alarmin, high mobility group protein-1 (HMGB1) (135), and IL-1 α (11, 135). IL-33 is further up regulated in keratinocytes after UV-irradiation and induces the secretion of cytokines like IL-10, IL-5 and IL-13 and many chemokines that are important in UV-mediated immunosuppressive functions (134, 136, 137). Very high levels of IL-33 have been found in both murine and human epidermis and dermis following UVB exposure, but not upon UVA exposure (134). In vitro, human and mouse keratinocytes and fibroblasts also secrete IL-33 upon UVB exposure (134). IL-33 promotes UV-induced immune suppression, and may be an important regulator of IL-1 β -mediated inflammation during UV photodamage responses. Evidence in support of this hypothesis comes in part from studies demonstrating that while bone marrow-derived DCs were shown to up-regulate production of all inflammasome-dependent cytokine secretion, the presence of prostoglandin E2 (PGE2) induced selective secretion of IL-33 without concomitant increases in IL-1 β and IL-18 (138). PGE2 is synthesized by the enzyme cyclooxygenase-2 (Cox-2), and is induced by the immunosuppressive chemical compound cis-urocanic acid (UCA), both of which are, respectively, induced or synthesized by UVR (139). PGE2 is a key mediator of UV-induced immunosuppression, where it signals through its receptors EP1 and EP4 present on many cell types including dendritic cells (140). Mice pretreated with an antagonist drug specific for EP4 had a diminished capacity to develop UV-induced immune suppress. Development of immunosuppression, concomitant with increased levels of Foxp3+ regulatory T cells (Treg cells) was rescued when indomethecin treated (Cox-2 inhibited) mice were given an EP4 agonist drug (141). Further, IL-33 is a potent stimulator of PGE2 secretion from immune cells (142). Mice injected with IL-33 developed poor contact hypersensitivity Th1 responses. Further, UV-induced squamous cell carcinomas express significantly higher levels of IL-33 demonstrating a novel tumor immune evasion mechanism (134). Experiments looking at the impact of IL-33 on tumor promotion of γ -irradiated skin fibroblasts demonstrated that blocking IL-33 resulted in apoptosis and IL-33 expression rescued cell survival (143). Furthermore, IL-33 expression is associated with recruitment of increased myeloid derived suppressor cells (MDSCs) and regulatory T-cells in a heart allograft model(144). IL-33 has been shown to activate TLR-4 induced inflammatory cytokines (145). These cytokines are immunosuppressive in UV-induced processes and loss of TLR4 inhibits UV-induced immunosuppression (146), further emphasizing that IL-33 acts in opposition to IL-1 β and IL-18 function. The alarmin activity is atypical in that it seems to induce immunosuppressive inflammation. IL-33 is self-regulated to some extent, since its activity can be abrogated by caspase-1 cleavage. (11). Thus IL-33 activities are in opposition to IL-1 β functions.

Downstream cytokines of IL-1 β ; Role of IL-12/IL-23 in UVB

Keratinocytes constitutively secrete low levels of both IL-23 and IL-12 (147), but upon UV-induced skin damage the expression is enhanced (148, 149). Whether IL-1 β secretion has an effect on the production of IL-12 and IL-23 in keratinocytes and whether the production of

these cytokines upon UV stimulation is IL-1 β dependent has yet to be investigated. However, in murine dendritic cells, IL-1 β induces expression of both subunits for IL-12 (p35 and p40). This was blocked by anti-IL-1 β antibody (150). The same may be true for human keratinocytes. Further, in fibroblast-like synoviocytes, IL-1 β induces the expression of both IL-12 and IL-23, demonstrating that IL-1 β can activate both of these cytokines (151). The role of IL-12 protection against UV-induced immunosuppression and tumorigenesis has been described in detail, especially with respect to its enhancement of DNA repair mechanisms (55, 152, 153). IL-12 reduces IL-10 production and enhances the activity of NER enzymes in the skin. IL-12 must be present in skin prior to UV exposure to obtain protective benefits. IL-12 stimulates fibronectin and various other HSPs that can act ligands for TLRs, necessary for priming NF κ B activation of IL-1 β , and IL-18 gene transcription(121). This can be regarded as another cross talk mechanism between NLRs and TLRs. Recently, IL-23 has been shown to play a similar role to IL-12 in DNA repair and immunosuppression against UV (154). However, its role in tumorigenesis is controversial (155).

The inhibition of UV-induced immunosuppression is also attributed to synergistic signals generated by the presence of both IL-12 and IL-18 or IL-1 β , which act in concert to polarize development of T cells into interferon gamma producing Th1 cells (156). Similarly, IL-1 β acts in concert with IL-23 to induce and expand Th17 cells, which are also able to block T-reg-mediated UV-tolerance induction (157, 158).

CONCLUSION

The activation of inflammasomes is an early photodamage response which engages an ancient protective mechanism that responds to the release of highly conserved DAMPS, to initiate “sterile” inflammatory responses required for cutaneous wound healing responses and for maintaining epidermal barrier integrity. Proinflammatory cytokines IL-1 β , IL-18 as well as IL-12 and IL-23 act directly and indirectly on keratinocytes to activate potent pro-survival mechanisms, including apoptosis inhibition and enhanced activation of DNA repair mechanisms. The production of IL-18, IL-1 β and IL-33 act locally and systemically to selectively expand particular arms of the cellular immune system; Th1, Th17, Th2 and T reg development, respectively. Complex cross regulation is evident among these mediators as demonstrated by UV-induced increases in IL-1 β , which can act to promote or inhibit Th1 responses, depending, in part on IL-1 β concentrations. UV-exposure increases production of IL-33, which in turn promotes development of Th2 and Treg cells – the critical effector cells that mediate UV-induced immunosuppression. While additional processes are involved in UV-induced immunosuppression, the context in which keratinocytes are induced to activate the inflammasome during the photodamage response is key in determining the fate of cellular repair and in setting up cutaneous inflammatory or adaptive immune responses that are ultimately responsible for maintaining immune surveillance against cancer.

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REFERENCES

1. Diffey BL. Sources and measurement of ultraviolet radiation. *Methods*. 2002; 28:4–13. [PubMed: 12231182]
2. Anderson RR, Parrish JA. The optics of human skin. *The Journal of investigative dermatology*. 1981; 77:13–19. [PubMed: 7252245]
3. Agar NS, Halliday GM, Barnetson RS, Ananthaswamy HN, Wheeler M, Jones AM. The basal layer in human squamous tumors harbors more UVA than UVB fingerprint mutations: a role for UVA in human skin carcinogenesis. *Proceedings of the National Academy of Sciences of the United States of America*. 2004; 101:4954–4959. [PubMed: 15041750]
4. Godar DE, Landry RJ, Lucas AD. Increased UVA exposures and decreased cutaneous Vitamin D(3) levels may be responsible for the increasing incidence of melanoma. *Med Hypotheses*. 2009; 72:434–443. [PubMed: 19155143]
5. Miller SA, Hamilton SL, Wester UG, Cyr WH. An analysis of UVA emissions from sunlamps and the potential importance for melanoma. *Photochemistry and photobiology*. 1998; 68:63–70. [PubMed: 9679452]
6. Hildesheim J, Awwad RT, Fornace AJ Jr. p38 Mitogen-activated protein kinase inhibitor protects the epidermis against the acute damaging effects of ultraviolet irradiation by blocking apoptosis and inflammatory responses. *The Journal of investigative dermatology*. 2004; 122:497–502. [PubMed: 15009736]
7. Runger TM. Role of UVA in the pathogenesis of melanoma and non-melanoma skin cancer. A short review. *Photodermatol Photoimmunol Photomed*. 1999; 15:212–216. [PubMed: 10599968]
8. Jans J, Garinis GA, Schul W, van Oudenaren A, Moorhouse M, Smid M, Sert YG, van der Velde A, Rijksen Y, de Gruijl FR, van der Spek PJ, Yasui A, Hoeijmakers JH, Leenen PJ, van der Horst GT. Differential role of basal keratinocytes in UV-induced immunosuppression and skin cancer. *Molecular and cellular biology*. 2006; 26:8515–8526. [PubMed: 16966369]
9. Fisher MS, Kripke ML. Further studies on the tumor-specific suppressor cells induced by ultraviolet radiation. *Journal of immunology*. 1978; 121:1139–1144.
10. Keller M, Ruegg A, Werner S, Beer HD. Active caspase-1 is a regulator of unconventional protein secretion. *Cell*. 2008; 132:818–831. [PubMed: 18329368]
11. Cayrol C, Girard JP. The IL-1-like cytokine IL-33 is inactivated after maturation by caspase-1. *Proceedings of the National Academy of Sciences of the United States of America*. 2009; 106:9021–9026. [PubMed: 19439663]

12. Feldmeyer L, Keller M, Niklaus G, Hohl D, Werner S, Beer HD. The inflammasome mediates UVB-induced activation and secretion of interleukin-1beta by keratinocytes. *Current biology : CB*. 2007; 17:1140–1145. [PubMed: 17600714]
13. Faustin B, Reed JC. Sunburned skin activates inflammasomes. *Trends in cell biology*. 2008; 18:4–8. [PubMed: 18083030]
14. Chen G, Pedra JH. The Inflammasome in Host Defense. *Sensors*. 2010; 10:97–111. [PubMed: 22315529]
15. Norval M. The mechanisms and consequences of ultraviolet-induced immunosuppression. *Prog Biophys Mol Biol*. 2006; 92:108–118. [PubMed: 16564073]
16. Shornick LP, Bisarya AK, Chaplin DD. IL-1beta is essential for langerhans cell activation and antigen delivery to the lymph nodes during contact sensitization: evidence for a dermal source of IL-1beta. *Cellular immunology*. 2001; 211:105–112. [PubMed: 11591114]
17. Matsushima H, Ogawa Y, Miyazaki T, Tanaka H, Nishibu A, Takashima A. Intravital imaging of IL-1beta production in skin. *The Journal of investigative dermatology*. 2010; 130:1571–1580. [PubMed: 20147964]
18. Timares L, Katiyar SK, Elmetts CA. DNA damage, apoptosis and langerhans cells--Activators of UV-induced immune tolerance. *Photochemistry and photobiology*. 2008; 84:422–436. [PubMed: 18248501]
19. Moodycliffe AM, Kimber I, Norval M. The effect of ultraviolet B irradiation and urocanic acid isomers on dendritic cell migration. *Immunology*. 1992; 77:394–399. [PubMed: 1478685]
20. Werman A, Werman-Venkert R, White R, Lee JK, Werman B, Krelin Y, Voronov E, Dinarello CA, Apte RN. The precursor form of IL-1alpha is an intracrine proinflammatory activator of transcription. *Proceedings of the National Academy of Sciences of the United States of America*. 2004; 101:2434–2439. [PubMed: 14983027]
21. Cohen I, Rider P, Carmi Y, Braiman A, Dotan S, White MR, Voronov E, Martin MU, Dinarello CA, Apte RN. Differential release of chromatin-bound IL-1alpha discriminates between necrotic and apoptotic cell death by the ability to induce sterile inflammation. *Proceedings of the National Academy of Sciences of the United States of America*. 2010; 107:2574–2579. [PubMed: 20133797]
22. Fritz JH, Girardin SE. How Toll-like receptors and Nod-like receptors contribute to innate immunity in mammals. *J Endotoxin Res*. 2005; 11:390–394. [PubMed: 16303096]
23. Schroder K, Tschopp J. The inflammasomes. *Cell*. 2010; 140:821–832. [PubMed: 20303873]
24. Akira S, Takeda K. Toll-like receptor signalling. *Nat Rev Immunol*. 2004; 4:499–511. [PubMed: 15229469]
25. Takeda K, Akira S. Toll-like receptors in innate immunity. *International immunology*. 2005; 17:1–14. [PubMed: 15585605]
26. Mishra BB, Gundra UM, Teale JM. Expression and distribution of Toll-like receptors 11–13 in the brain during murine neurocysticercosis. *J Neuroinflammation*. 2008; 5:53. [PubMed: 19077284]
27. Lebre MC, van der Aar AM, van Baarsen L, van Capel TM, Schuitemaker JH, Kapsenberg ML, de Jong EC. Human keratinocytes express functional Toll-like receptor 3, 4, 5, and 9. *The Journal of investigative dermatology*. 2007; 127:331–341. [PubMed: 17068485]
28. Miller LS, Modlin RL. Human keratinocyte Toll-like receptors promote distinct immune responses. *The Journal of investigative dermatology*. 2007; 127:262–263. [PubMed: 17228303]
29. Pivarsci A, Bodai L, Rethi B, Kenderessy-Szabo A, Koreck A, Szell M, Beer Z, Bata-Csorgoo Z, Magocsi M, Rajnavolgyi E, Dobozy A, Kemeny L. Expression and function of Toll-like receptors 2 and 4 in human keratinocytes. *International immunology*. 2003; 15:721–730. [PubMed: 12750356]
30. Tsan MF, Gao B. Endogenous ligands of Toll-like receptors. *Journal of leukocyte biology*. 2004; 76:514–519. [PubMed: 15178705]
31. Ivanov S, Dragoi AM, Wang X, Dallacosta C, Louten J, Musco G, Sitia G, Yap GS, Wan Y, Biron CA, Bianchi ME, Wang H, Chu WM. A novel role for HMGB1 in TLR9-mediated inflammatory responses to CpG-DNA. *Blood*. 2007; 110:1970–1981. [PubMed: 17548579]
32. Lee Y, Kim H, Kim S, Shin MH, Kim YK, Kim KH, Chung JH. Myeloid differentiation factor 88 regulates basal and UV-induced expressions of IL-6 and MMP-1 in human epidermal

- keratinocytes. *The Journal of investigative dermatology*. 2009; 129:460–467. [PubMed: 18719610]
33. Lee, Y.; Kong, K.; Lee, SR.; Yoon, HS.; Kim, KH.; Chung, JH. The 20th Academic Meeting of Korean Society for Investigative Dermatology. Korea: 2010. MyD88 mediates UV-induced inflammatory responses and cell apoptosis in mouse skin in vivo.
 34. Matsuda M, Hoshino T, Yamashita Y, Tanaka K, Maji D, Sato K, Adachi H, Sobue G, Ihn H, Funasaka Y, Mizushima T. Prevention of UVB radiation-induced epidermal damage by expression of heat shock protein 70. *The Journal of biological chemistry*. 2010; 285:5848–5858. [PubMed: 20018843]
 35. Perluigi M, Di Domenico F, Blarzino C, Foppoli C, Cini C, Giorgi A, Grillo C, De Marco F, Butterfield DA, Schinina ME, Coccia R. Effects of UVB-induced oxidative stress on protein expression and specific protein oxidation in normal human epithelial keratinocytes: a proteomic approach. *Proteome Sci*. 2010; 8:13. [PubMed: 20298559]
 36. Glaser R, Navid F, Schuller W, Jantschitsch C, Harder J, Schroder JM, Schwarz A, Schwarz T. UV-B radiation induces the expression of antimicrobial peptides in human keratinocytes in vitro and in vivo. *The Journal of allergy and clinical immunology*. 2009; 123:1117–1123. [PubMed: 19342087]
 37. Abeyama K, Stern DM, Ito Y, Kawahara K, Yoshimoto Y, Tanaka M, Uchimura T, Ida N, Yamazaki Y, Yamada S, Yamamoto Y, Yamamoto H, Iino S, Taniguchi N, Maruyama I. The N-terminal domain of thrombomodulin sequesters high-mobility group-B1 protein, a novel antiinflammatory mechanism. *The Journal of clinical investigation*. 2005; 115:1267–1274. [PubMed: 15841214]
 38. Shi B, Grahn JC, Reilly DA, Dizon TC, Isseroff RR. Responses of the 27-kDa heat shock protein to UVB irradiation in human epidermal melanocytes. *Exp Dermatol*. 2008; 17:108–114. [PubMed: 18031542]
 39. Bivik C, Rosdahl I, Ollinger K. Hsp70 protects against UVB induced apoptosis by preventing release of cathepsins and cytochrome c in human melanocytes. *Carcinogenesis*. 2007; 28:537–544. [PubMed: 16950797]
 40. Netea MG, van de Veerdonk FL, Kullberg BJ, Van der Meer JW, Joosten LA. The role of NLRs and TLRs in the activation of the inflammasome. *Expert Opin Biol Ther*. 2008; 8:1867–1872. [PubMed: 18990074]
 41. Watanabe H, Gaide O, Petrilli V, Martinon F, Contassot E, Roques S, Kummer JA, Tschopp J, French LE. Activation of the IL-1 β -processing inflammasome is involved in contact hypersensitivity. *The Journal of investigative dermatology*. 2007; 127:1956–1963. [PubMed: 17429439]
 42. Gaspari, AA.; Tying, SK. SpringerLink (Online service). *Clinical and Basic Immunodermatology*. London: Springer London; 2008.
 43. Wang P, Wu P, Anthes JC, Siegel MI, Egan RW, Billah MM. Interleukin-10 inhibits interleukin-8 production in human neutrophils. *Blood*. 1994; 83:2678–2683. [PubMed: 8167346]
 44. Schroder M, Meisel C, Buhl K, Profanter N, Sievert N, Volk HD, Grutz G. Different modes of IL-10 and TGF- β to inhibit cytokine-dependent IFN- γ production: consequences for reversal of lipopolysaccharide desensitization. *Journal of immunology*. 2003; 170:5260–5267.
 45. Stutz A, Golenbock DT, Latz E. Inflammasomes: too big to miss. *The Journal of clinical investigation*. 2009; 119:3502–3511. [PubMed: 19955661]
 46. Church LD, Cook GP, McDermott MF. Primer: inflammasomes and interleukin 1 β in inflammatory disorders. *Nat Clin Pract Rheumatol*. 2008; 4:34–42. [PubMed: 18172447]
 47. Ting JP, Lovering RC, Alnemri ES, Bertin J, Boss JM, Davis BK, Flavell RA, Girardin SE, Godzik A, Harton JA, Hoffman HM, Hugot JP, Inohara N, Mackenzie A, Maltais LJ, Nunez G, Ogura Y, Otten LA, Philpott D, Reed JC, Reith W, Schreiber S, Steimle V, Ward PA. The NLR gene family: a standard nomenclature. *Immunity*. 2008; 28:285–287. [PubMed: 18341998]
 48. Harder J, Nunez G. Functional expression of the intracellular pattern recognition receptor NOD1 in human keratinocytes. *The Journal of investigative dermatology*. 2009; 129:1299–1302. [PubMed: 19122645]

49. Inohara N, Ogura Y, Fontalba A, Gutierrez O, Pons F, Crespo J, Fukase K, Inamura S, Kusumoto S, Hashimoto M, Foster SJ, Moran AP, Fernandez-Luna JL, Nunez G. Host recognition of bacterial muramyl dipeptide mediated through NOD2. Implications for Crohn's disease. *The Journal of biological chemistry*. 2003; 278:5509–5512. [PubMed: 12514169]
50. Chamaillard M, Hashimoto M, Horie Y, Masumoto J, Qiu S, Saab L, Ogura Y, Kawasaki A, Fukase K, Kusumoto S, Valvano MA, Foster SJ, Mak TW, Nunez G, Inohara N. An essential role for NOD1 in host recognition of bacterial peptidoglycan containing diaminopimelic acid. *Nature immunology*. 2003; 4:702–707. [PubMed: 12796777]
51. Pais TF, Figueiredo C, Peixoto R, Braz MH, Chatterjee S. Necrotic neurons enhance microglial neurotoxicity through induction of glutaminase by a MyD88-dependent pathway. *J Neuroinflammation*. 2008; 5:43. [PubMed: 18844999]
52. Dostert C, Meylan E, Tschopp J. Intracellular pattern-recognition receptors. *Adv Drug Deliv Rev*. 2008; 60:830–840. [PubMed: 18280002]
53. Yamasaki K, Muto J, Taylor KR, Cogen AL, Audish D, Bertin J, Grant EP, Coyle AJ, Misaghi A, Hoffman HM, Gallo RL. NLRP3/cryopyrin is necessary for interleukin-1beta (IL-1beta) release in response to hyaluronan, an endogenous trigger of inflammation in response to injury. *The Journal of biological chemistry*. 2009; 284:12762–12771. [PubMed: 19258328]
54. Williams, A. *Immunology: Mucosal and Body Surface Defences*. Wiley-Blackwell; 2012.
55. Schwarz T, Schwarz A. DNA repair and cytokine responses. *J Invest Dermatol Symp Proc*. 2009; 14:63–66.
56. Benko S, Tozser J, Miklossy G, Varga A, Kadas J, Csutak A, Berta A, Rajnavolgyi E. Constitutive and UV-B modulated transcription of Nod-like receptors and their functional partners in human corneal epithelial cells. *Molecular vision*. 2008; 14:1575–1583. [PubMed: 18769647]
57. Pearton DJ, Ferraris C, Dhouailly D. Transdifferentiation of corneal epithelium: evidence for a linkage between the segregation of epidermal stem cells and the induction of hair follicles during embryogenesis. *Int J Dev Biol*. 2004; 48:197–201. [PubMed: 15272385]
58. Martinon F, Burns K, Tschopp J. The inflammasome: a molecular platform triggering activation of inflammatory caspases and processing of proIL-beta. *Molecular cell*. 2002; 10:417–426. [PubMed: 12191486]
59. Kummer JA, Broekhuizen R, Everett H, Agostini L, Kuijk L, Martinon F, van Bruggen R, Tschopp J. Inflammasome components NALP 1 and 3 show distinct but separate expression profiles in human tissues suggesting a site-specific role in the inflammatory response. *J Histochem Cytochem*. 2007; 55:443–452. [PubMed: 17164409]
60. Hsu LC, Ali SR, McGillivray S, Tseng PH, Mariathasan S, Humke EW, Eckmann L, Powell JJ, Nizet V, Dixit VM, Karin M. A NOD2-NALP1 complex mediates caspase-1-dependent IL-1beta secretion in response to *Bacillus anthracis* infection and muramyl dipeptide. *Proc Natl Acad Sci U S A*. 2008; 105:7803–7808. [PubMed: 18511561]
61. Faustin B, Chen Y, Zhai D, Le Negrat G, Lartigue L, Satterthwait A, Reed JC. Mechanism of Bcl-2 and Bcl-X(L) inhibition of NLRP1 inflammasome: loop domain-dependent suppression of ATP binding and oligomerization. *Proceedings of the National Academy of Sciences of the United States of America*. 2009; 106:3935–3940. [PubMed: 19223583]
62. Reagan-Shaw S, Breur J, Ahmad N. Enhancement of UVB radiation-mediated apoptosis by sanguinarine in HaCaT human immortalized keratinocytes. *Molecular cancer therapeutics*. 2006; 5:418–429. [PubMed: 16505117]
63. Gross O, Poeck H, Bscheider M, Dostert C, Hanneschlager N, Endres S, Hartmann G, Tardivel A, Schweighoffer E, Tybulewicz V, Mocsai A, Tschopp J, Ruland J. Syk kinase signalling couples to the Nlrp3 inflammasome for anti-fungal host defence. *Nature*. 2009; 459:433–436. [PubMed: 19339971]
64. Martinon F, Petrilli V, Mayor A, Tardivel A, Tschopp J. Gout-associated uric acid crystals activate the NALP3 inflammasome. *Nature*. 2006; 440:237–241. [PubMed: 16407889]
65. Kanneganti TD, Ozoren N, Body-Malapel M, Amer A, Park JH, Franchi L, Whitfield J, Barchet W, Colonna M, Vandenabeele P, Bertin J, Coyle A, Grant EP, Akira S, Nunez G. Bacterial RNA and small antiviral compounds activate caspase-1 through cryopyrin/Nalp3. *Nature*. 2006; 440:233–236. [PubMed: 16407888]

66. Muruve DA, Petrilli V, Zaiss AK, White LR, Clark SA, Ross PJ, Parks RJ, Tschopp J. The inflammasome recognizes cytosolic microbial and host DNA and triggers an innate immune response. *Nature*. 2008; 452:103–107. [PubMed: 18288107]
67. Duncan JA, Bergstralh DT, Wang Y, Willingham SB, Ye Z, Zimmermann AG, Ting JP. Cryopyrin/NALP3 binds ATP/dATP, is an ATPase, and requires ATP binding to mediate inflammatory signaling. *Proceedings of the National Academy of Sciences of the United States of America*. 2007; 104:8041–8046. [PubMed: 17483456]
68. Mariathasan S, Weiss DS, Newton K, McBride J, O'Rourke K, Roose-Girma M, Lee WP, Weinrauch Y, Monack DM, Dixit VM. Cryopyrin activates the inflammasome in response to toxins and ATP. *Nature*. 2006; 440:228–232. [PubMed: 16407890]
69. Filippini A, Taffs RE, Sitkovsky MV. Extracellular ATP in T-lymphocyte activation: possible role in effector functions. *Proceedings of the National Academy of Sciences of the United States of America*. 1990; 87:8267–8271. [PubMed: 2146684]
70. Netea MG, Nold-Petry CA, Nold MF, Joosten LA, Opitz B, van der Meer JH, van de Veerdonk FL, Ferwerda G, Heinhuis B, Devesa I, Funk CJ, Mason RJ, Kullberg BJ, Rubartelli A, van der Meer JW, Dinarello CA. Differential requirement for the activation of the inflammasome for processing and release of IL-1 β in monocytes and macrophages. *Blood*. 2009; 113:2324–2335. [PubMed: 19104081]
71. Takai E, Tsukimoto M, Harada H, Kojima S. Involvement of P2Y6 receptor in p38 MAPK-mediated COX-2 expression in response to UVB irradiation of human keratinocytes. *Radiation research*. 2011; 175:358–366. [PubMed: 21388279]
72. Mizumoto N, Kumamoto T, Robson SC, Seigny J, Matsue H, Enjoji K, Takashima A. CD39 is the dominant Langerhans cell-associated ecto-NTPDase: modulatory roles in inflammation and immune responsiveness. *Nature medicine*. 2002; 8:358–365.
73. Ferrari D, Wesselborg S, Bauer MK, Schulze-Osthoff K. Extracellular ATP activates transcription factor NF- κ B through the P2Z purinoreceptor by selectively targeting NF- κ B p65. *The Journal of cell biology*. 1997; 139:1635–1643. [PubMed: 9412459]
74. Inoue K, Denda M, Tozaki H, Fujishita K, Koizumi S. Characterization of multiple P2X receptors in cultured normal human epidermal keratinocytes. *The Journal of investigative dermatology*. 2005; 124:756–763. [PubMed: 15816834]
75. Weber FC, Esser PR, Muller T, Ganesan J, Pellegatti P, Simon MM, Zeiser R, Idzko M, Jakob T, Martin SF. Lack of the purinergic receptor P2X(7) results in resistance to contact hypersensitivity. *The Journal of experimental medicine*. 2010; 207:2609–2619. [PubMed: 21059855]
76. Shio MT, Eisenbarth SC, Savaria M, Vinet AF, Bellemare MJ, Harder KW, Sutterwala FS, Bohle DS, Descoteaux A, Flavell RA, Olivier M. Malarial hemozoin activates the NLRP3 inflammasome through Lyn and Syk kinases. *PLoS Pathog*. 2009; 5:e1000559. [PubMed: 19696895]
77. Surprenant A, Rassendren F, Kawashima E, North RA, Buell G. The cytolytic P2Z receptor for extracellular ATP identified as a P2X receptor (P2X7). *Science*. 1996; 272:735–738. [PubMed: 8614837]
78. Perregaux D, Gabel CA. Interleukin-1 β maturation and release in response to ATP and nigericin. Evidence that potassium depletion mediated by these agents is a necessary and common feature of their activity. *The Journal of biological chemistry*. 1994; 269:15195–15203. [PubMed: 8195155]
79. Gudipaty L, Munetz J, Verhoef PA, Dubyak GR. Essential role for Ca²⁺ in regulation of IL-1 β secretion by P2X7 nucleotide receptor in monocytes, macrophages, and HEK-293 cells. *American journal of physiology. Cell physiology*. 2003; 285:C286–C299. [PubMed: 12660148]
80. Werth BB, Bashir M, Chang L, Werth VP. Ultraviolet irradiation induces the accumulation of chondroitin sulfate, but not other glycosaminoglycans, in human skin. *PLoS One*. 2011; 6:e14830. [PubMed: 21829593]
81. Kakizaki I, Itano N, Kimata K, Hanada K, Kon A, Yamaguchi M, Takahashi T, Takagaki K. Up-regulation of hyaluronan synthase genes in cultured human epidermal keratinocytes by UVB irradiation. *Archives of biochemistry and biophysics*. 2008; 471:85–93. [PubMed: 18158910]

82. Dostert C, Petrilli V, Van Bruggen R, Steele C, Mossman BT, Tschopp J. Innate immune activation through Nalp3 inflammasome sensing of asbestos and silica. *Science*. 2008; 320:674–677. [PubMed: 18403674]
83. Heck DE, Vetrano AM, Mariano TM, Laskin JD. UVB light stimulates production of reactive oxygen species: unexpected role for catalase. *The Journal of biological chemistry*. 2003; 278:22432–22436. [PubMed: 12730222]
84. Sorbara MT, Girardin SE. Mitochondrial ROS fuel the inflammasome. *Cell research*. 2011; 21:558–560. [PubMed: 21283134]
85. Zhou R, Yazdi AS, Menu P, Tschopp J. A role for mitochondria in NLRP3 inflammasome activation. *Nature*. 2011; 469:221–225. [PubMed: 21124315]
86. Zhou R, Tardivel A, Thorens B, Choi I, Tschopp J. Thioredoxin-interacting protein links oxidative stress to inflammasome activation. *Nature immunology*. 2010; 11:136–140. [PubMed: 20023662]
87. Yao Y, Wolverson JE, Zhang Q, Marathe GK, Al-Hassani M, Konger RL, Travers JB. Ultraviolet B radiation generated platelet-activating factor receptor agonist formation involves EGF-R-mediated reactive oxygen species. *Journal of immunology*. 2009; 182:2842–2848.
88. Fernandes-Alnemri T, Yu JW, Datta P, Wu J, Alnemri ES. AIM2 activates the inflammasome and cell death in response to cytoplasmic DNA. *Nature*. 2009; 458:509–513. [PubMed: 19158676]
89. Roberts TL, Idris A, Dunn JA, Kelly GM, Burnton CM, Hodgson S, Hardy LL, Garceau V, Sweet MJ, Ross IL, Hume DA, Stacey KJ. HIN-200 proteins regulate caspase activation in response to foreign cytoplasmic DNA. *Science*. 2009; 323:1057–1060. [PubMed: 19131592]
90. Dombrowski YP, Koglin M, Kammerbauer S, Göb C, Anz C, Simanski D, Gläser M, Harder R, Hornung J, Gallo V, Ruzicka R, Besch R, R Schaubert J. Cytosolic DNA Triggers Inflammasome Activation in Keratinocytes in Psoriatic Lesions. *Science translational medicine*. 2011; 3
91. Higuchi Y, Mizukami Y, Yoshimoto T. Ultraviolet ray induces chromosomal giant DNA fragmentation followed by internucleosomal DNA fragmentation associated with apoptosis in rat glioma cells. *Annals of the New York Academy of Sciences*. 2003; 1010:326–330. [PubMed: 15033744]
92. Martinon F, Tschopp J. Inflammatory caspases and inflammasomes: master switches of inflammation. *Cell death and differentiation*. 2007; 14:10–22. [PubMed: 16977329]
93. Petrilli V, Papin S, Dostert C, Mayor A, Martinon F, Tschopp J. Activation of the NALP3 inflammasome is triggered by low intracellular potassium concentration. *Cell death and differentiation*. 2007; 14:1583–1589. [PubMed: 17599094]
94. Fiorucci S, Santucci L, Cirino G, Mencarelli A, Familiari L, Soldato PD, Morelli A. IL-1 beta converting enzyme is a target for nitric oxide-releasing aspirin: new insights in the antiinflammatory mechanism of nitric oxide-releasing nonsteroidal antiinflammatory drugs. *Journal of immunology*. 2000; 165:5245–5254.
95. Melnikov VY, Ecder T, Fantuzzi G, Siegmund B, Lucia MS, Dinarello CA, Schrier RW, Edelstein CL. Impaired IL-18 processing protects caspase-1-deficient mice from ischemic acute renal failure. *The Journal of clinical investigation*. 2001; 107:1145–1152. [PubMed: 11342578]
96. Ohno T, Oboki K, Kajiura N, Morii E, Aozasa K, Flavell RA, Okumura K, Saito H, Nakae S. Caspase-1, caspase-8, and calpain are dispensable for IL-33 release by macrophages. *Journal of immunology*. 2009; 183:7890–7897.
97. Afonina IS, Tynan GA, Logue SE, Cullen SP, Bots M, Luthi AU, Reeves EP, McElvaney NG, Medema JP, Lavelle EC, Martin SJ. Granzyme B-dependent proteolysis acts as a switch to enhance the proinflammatory activity of IL-1alpha. *Molecular cell*. 2011; 44:265–278. [PubMed: 22017873]
98. Jensen LE. Targeting the IL-1 family members in skin inflammation. *Curr Opin Investig Drugs*. 2010; 11:1211–1220.
99. Apte RN, Dotan S, Elkabets M, White MR, Reich E, Carmi Y, Song X, Dvozkin T, Krelin Y, Voronov E. The involvement of IL-1 in tumorigenesis, tumor invasiveness, metastasis and tumor-host interactions. *Cancer metastasis reviews*. 2006; 25:387–408. [PubMed: 17043764]
100. Song X, Voronov E, Dvorkin T, Fima E, Cagnano E, Benharroch D, Shendler Y, Bjorkdahl O, Segal S, Dinarello CA, Apte RN. Differential effects of IL-1 alpha and IL-1 beta on tumorigenicity patterns and invasiveness. *Journal of immunology*. 2003; 171:6448–6456.

101. Fettelschoss A, Kistowska M, LeibundGut-Landmann S, Beer HD, Johansen P, Senti G, Contassot E, Bachmann MF, French LE, Oxenius A, Kundig TM. Inflammasome activation and IL-1 β target IL-1 α for secretion as opposed to surface expression. *Proceedings of the National Academy of Sciences of the United States of America*. 2011; 108:18055–18060. [PubMed: 22006336]
102. Dinarello CA. Interleukin-1, interleukin-1 receptors and interleukin-1 receptor antagonist. *Int Rev Immunol*. 1998; 16:457–499. [PubMed: 9646173]
103. Li L, Fei Z, Ren J, Sun R, Liu Z, Sheng Z, Wang L, Sun X, Yu J, Wang Z, Fei J. Functional imaging of interleukin 1 β expression in inflammatory process using bioluminescence imaging in transgenic mice. *BMC Immunol*. 2008; 9:49. [PubMed: 18710581]
104. Shahbakhthi H, Watson RE, Azurdia RM, Ferreira CZ, Garmyn M, Rhodes LE. Influence of eicosapentaenoic acid, an omega-3 fatty acid, on ultraviolet-B generation of prostaglandin-E2 and proinflammatory cytokines interleukin-1 β , tumor necrosis factor- α , interleukin-6 and interleukin-8 in human skin in vivo. *Photochemistry and photobiology*. 2004; 80:231–235. [PubMed: 15362934]
105. Keystone EC, Jaglal S, Shore A. Interleukin 1 and interleukin 2 generation by peripheral blood cells from patients with ankylosing spondylitis. *J Rheumatol*. 1986; 13:944–947. [PubMed: 3493347]
106. Lee SH, Stehlik C, Reed JC. Cop, a caspase recruitment domain-containing protein and inhibitor of caspase-1 activation processing. *The Journal of biological chemistry*. 2001; 276:34495–34500. [PubMed: 11432859]
107. Humke EW, Shriver SK, Starovasnik MA, Fairbrother WJ, Dixit VM. ICEBERG: a novel inhibitor of interleukin-1 β generation. *Cell*. 2000; 103:99–111. [PubMed: 11051551]
108. Katiyar SK, Mantena SK, Meeran SM. Silymarin protects epidermal keratinocytes from ultraviolet radiation-induced apoptosis and DNA damage by nucleotide excision repair mechanism. *PLoS One*. 2011; 6:e21410. [PubMed: 21731736]
109. Kothny-Wilkes G, Kulms D, Luger TA, Kubin M, Schwarz T. Interleukin-1 protects transformed keratinocytes from tumor necrosis factor-related apoptosis-inducing ligand- and CD95-induced apoptosis but not from ultraviolet radiation-induced apoptosis. *J Biol Chem*. 1999; 274:28916–28921. [PubMed: 10506136]
110. Schwarz A, Bhardwaj R, Aragane Y, Mahnke K, Riemann H, Metze D, Luger TA, Schwarz T. Ultraviolet-B-induced apoptosis of keratinocytes: evidence for partial involvement of tumor necrosis factor- α in the formation of sunburn cells. *J Invest Dermatol*. 1995; 104:922–927. [PubMed: 7769259]
111. Yurkovetsky ZR, Kirkwood JM, Edington HD, Marrangoni AM, Velikokhatnaya L, Winans MT, Gorelik E, Lokshin AE. Multiplex analysis of serum cytokines in melanoma patients treated with interferon- α 2b. *Clin Cancer Res*. 2007; 13:2422–2428. [PubMed: 17438101]
112. Meeran SM, Punathil T, Katiyar SK. IL-12 deficiency exacerbates inflammatory responses in UV-irradiated skin and skin tumors. *J Invest Dermatol*. 2008; 128:2716–2727. [PubMed: 18509359]
113. Voronov E, Dayan M, Zinger H, Gayvoronsky L, Lin JP, Iwakura Y, Apte RN, Mozes E. IL-1 β -deficient mice are resistant to induction of experimental SLE. *Eur Cytokine Netw*. 2006; 17:109–116. [PubMed: 16840029]
114. Baxevasis CN, Dedoussis GV, Gritzapis AD, Stathopoulos GP, Papamichail M. Interleukin 1 β synergises with interleukin 2 in the outgrowth of autologous tumour-reactive CD8+ effectors. *Br J Cancer*. 1994; 70:625–630. [PubMed: 7917907]
115. Nazarenko I, Marhaba R, Reich E, Voronov E, Vitacolonna M, Hildebrand D, Elter E, Rajasagi M, Apte RN, Zoller M. Tumorigenicity of IL-1 α - and IL-1 β -deficient fibrosarcoma cells. *Neoplasia*. 2008; 10:549–562. [PubMed: 18516292]
116. Nasti TH, Iqbal O, Tamimi IA, Geise JT, Katiyar SK, Yusuf N. Differential roles of T-cell subsets in regulation of ultraviolet radiation induced cutaneous photocarcinogenesis. *Photochemistry and photobiology*. 2011; 87:387–398. [PubMed: 21143237]
117. Loser K, Apelt J, Voskort M, Mohaupt M, Balkow S, Schwarz T, Grabbe S, Beissert S. IL-10 controls ultraviolet-induced carcinogenesis in mice. *Journal of immunology*. 2007; 179:365–371.

118. Schwarz T. Regulatory T cells induced by ultraviolet radiation. *Int Arch Allergy Immunol.* 2005; 137:187–193. [PubMed: 15947474]
119. O'Sullivan BJ, Thomas HE, Pai S, Santamaria P, Iwakura Y, Steptoe RJ, Kay TW, Thomas R. IL-1 beta breaks tolerance through expansion of CD25+ effector T cells. *J Immunol.* 2006; 176:7278–7287. [PubMed: 16751371]
120. Watanabe H, Gehrke S, Contassot E, Roques S, Tschopp J, Friedmann PS, French LE, Gaide O. Danger signaling through the inflammasome acts as a master switch between tolerance and sensitization. *Journal of immunology.* 2008; 180:5826–5832.
121. Molenda M, Mukkamala L, Blumenberg M. Interleukin IL-12 blocks a specific subset of the transcriptional profile responsive to UVB in epidermal keratinocytes. *Molecular immunology.* 2006; 43:1933–1940. [PubMed: 16466796]
122. Steinberg T, Dannewitz B, Tomakidi P, Hoheisel JD, Mussig E, Kohl A, Nees M. Analysis of interleukin-1beta-modulated mRNA gene transcription in human gingival keratinocytes by epithelia-specific cDNA microarrays. *Journal of periodontal research.* 2006; 41:426–446. [PubMed: 16953820]
123. Bergsbaken T, Fink SL, Cookson BT. Pyroptosis: host cell death and inflammation. *Nat Rev Microbiol.* 2009; 7:99–109. [PubMed: 19148178]
124. Lamkanfi M, Walle LV, Kanneganti TD. Deregulated inflammasome signaling in disease. *Immunol Rev.* 2011; 243:163–173. [PubMed: 21884175]
125. Mariathasan S, Newton K, Monack DM, Vucic D, French DM, Lee WP, Roose-Girma M, Erickson S, Dixit VM. Differential activation of the inflammasome by caspase-1 adaptors ASC and Ipaf. *Nature.* 2004; 430:213–218. [PubMed: 15190255]
126. Koizumi H, Sato-Matsumura KC, Nakamura H, Shida K, Kikkawa S, Matsumoto M, Toyoshima K, Seya T. Distribution of IL-18 and IL-18 receptor in human skin: various forms of IL-18 are produced in keratinocytes. *Arch Dermatol Res.* 2001; 293:325–333. [PubMed: 11550805]
127. Cho D, Seung Kang J, Hoon Park J, Kim YI, Hahm E, Lee J, Yang Y, Jeon J, Song H, Park H, Kim T, Pang S, Kim CW, Il Hwang Y, Jae Lee W. The enhanced IL-18 production by UVB irradiation requires ROI and AP-1 signaling in human keratinocyte cell line (HaCaT). *Biochemical and biophysical research communications.* 2002; 298:289–295. [PubMed: 12387830]
128. Vidal-Vanaclocha F, Mendoza L, Telleria N, Salado C, Valcarcel M, Gallot N, Carrascal T, Egilegor E, Beaskoetxea J, Dinarello CA. Clinical and experimental approaches to the pathophysiology of interleukin-18 in cancer progression. *Cancer metastasis reviews.* 2006; 25:417–434. [PubMed: 17001512]
129. Coughlin CM, Salhany KE, Wysocka M, Aruga E, Kurzawa H, Chang AE, Hunter CA, Fox JC, Trinchieri G, Lee WM. Interleukin-12 and interleukin-18 synergistically induce murine tumor regression which involves inhibition of angiogenesis. *The Journal of clinical investigation.* 1998; 101:1441–1452. [PubMed: 9502787]
130. Thong-Ngam D, Tangkijvanich P, Lerknimitr R, Mahachai V, Theamboonlers A, Poovorawan Y. Diagnostic role of serum interleukin-18 in gastric cancer patients. *World journal of gastroenterology : WJG.* 2006; 12:4473–4477. [PubMed: 16874857]
131. Tse BW, Russell PJ, Lochner M, Forster I, Power CA. IL-18 Inhibits Growth of Murine Orthotopic Prostate Carcinomas via Both Adaptive and Innate Immune Mechanisms. *PLoS One.* 2011; 6:e24241. [PubMed: 21935389]
132. Schwarz A, Maeda A, Stander S, van Steeg H, Schwarz T. IL-18 reduces ultraviolet radiation-induced DNA damage and thereby affects photoimmunosuppression. *Journal of immunology.* 2006; 176:2896–2901.
133. Wittmann M, Purwar R, Hartmann C, Gutzmer R, Werfel T. Human keratinocytes respond to interleukin-18: implication for the course of chronic inflammatory skin diseases. *The Journal of investigative dermatology.* 2005; 124:1225–1233. [PubMed: 15955098]
134. Byrne SN, Beaugie C, O'Sullivan C, Leighton S, Halliday GM. The immune-modulating cytokine and endogenous Alarmin interleukin-33 is upregulated in skin exposed to inflammatory UVB radiation. *The American journal of pathology.* 2011; 179:211–222. [PubMed: 21703403]

135. Moussion C, Ortega N, Girard JP. The IL-1-like cytokine IL-33 is constitutively expressed in the nucleus of endothelial cells and epithelial cells in vivo: a novel 'alarmin'? PLoS One. 2008; 3:e3331. [PubMed: 18836528]
136. Schmitz J, Owyang A, Oldham E, Song Y, Murphy E, McClanahan TK, Zurawski G, Moshrefi M, Qin J, Li X, Gorman DM, Bazan JF, Kastelein RA. IL-33, an interleukin-1-like cytokine that signals via the IL-1 receptor-related protein ST2 and induces T helper type 2-associated cytokines. Immunity. 2005; 23:479–490. [PubMed: 16286016]
137. Dawes JM, Calvo M, Perkins JR, Paterson KJ, Kiesewetter H, Hobbs C, Kaan TK, Orengo C, Bennett DL, McMahon SB. CXCL5 mediates UVB irradiation-induced pain. Science translational medicine. 2011; 3:90ra60.
138. Yanagawa Y, Suzuki M, Matsumoto M, Togashi H. Prostaglandin E(2) enhances IL-33 production by dendritic cells. Immunology letters. 2011
139. Kaneko K, Walker SL, Lai-Cheong J, Matsui MS, Norval M, Young AR. cis-Urocanic acid enhances prostaglandin E2 release and apoptotic cell death via reactive oxygen species in human keratinocytes. The Journal of investigative dermatology. 2011; 131:1262–1271. [PubMed: 21412256]
140. Tsutsumi R, Xie C, Wei X, Zhang M, Zhang X, Flick LM, Schwarz EM, O'Keefe RJ. PGE2 signaling through the EP4 receptor on fibroblasts upregulates RANKL and stimulates osteolysis. Journal of bone and mineral research : the official journal of the American Society for Bone and Mineral Research. 2009; 24:1753–1762. [PubMed: 19419302]
141. Soontrapa K, Honda T, Sakata D, Yao C, Hirata T, Hori S, Matsuoka T, Kita Y, Shimizu T, Kabashima K, Narumiya S. Prostaglandin E2-prostaglandin E receptor subtype 4 (EP4) signaling mediates UV irradiation-induced systemic immunosuppression. Proceedings of the National Academy of Sciences of the United States of America. 2011; 108:6668–6673. [PubMed: 21460251]
142. Miller AM. Role of IL-33 in inflammation and disease. J Inflamm (Lond). 2011; 8:22. [PubMed: 21871091]
143. Ivanov VN, Zhou H, Ghandhi SA, Karasic TB, Yaghoubian B, Amundson SA, Hei TK. Radiation-induced bystander signaling pathways in human fibroblasts: a role for interleukin-33 in the signal transmission. Cellular signalling. 2010; 22:1076–1087. [PubMed: 20206688]
144. Brunner SM, Schiechl G, Falk W, Schlitt HJ, Geissler EK, Fichtner-Feigl S. Interleukin-33 prolongs allograft survival during chronic cardiac rejection. Transpl Int. 2011; 24:1027–1039. [PubMed: 21797940]
145. Espinassous Q, Garcia-de-Paco E, Garcia-Verdugo I, Synguelakis M, von Aulock S, Sallenave JM, McKenzie AN, Kanellopoulos J. IL-33 enhances lipopolysaccharide-induced inflammatory cytokine production from mouse macrophages by regulating lipopolysaccharide receptor complex. Journal of immunology. 2009; 183:1446–1455.
146. Lewis W, Simanyi E, Li H, Thompson CA, Nasti TH, Jaleel T, Xu H, Yusuf N. Regulation of ultraviolet radiation induced cutaneous photoimmunosuppression by toll-like receptor-4. Archives of biochemistry and biophysics. 2011; 508:171–177. [PubMed: 21236239]
147. Piskin G, Sylva-Steenland RM, Bos JD, Teunissen MB. In vitro and in situ expression of IL-23 by keratinocytes in healthy skin and psoriasis lesions: enhanced expression in psoriatic skin. Journal of immunology. 2006; 176:1908–1915.
148. Enk CD, Mahanty S, Blauvelt A, Katz SI. UVB induces IL-12 transcription in human keratinocytes in vivo and in vitro. Photochemistry and photobiology. 1996; 63:854–859. [PubMed: 8992506]
149. Kennedy-Crispin M, Billick E, Mitsui H, Gulati N, Fujita H, Gilleaudeau P, Sullivan-Whalen M, Johnson-Huang LM, Suarez-Farinas M, Krueger JG. Human Keratinocytes' Response to Injury Upregulates CCL20 and Other Genes Linking Innate and Adaptive Immunity. The Journal of investigative dermatology. 2011
150. Wesa AK, Galy A. IL-1 beta induces dendritic cells to produce IL-12. International immunology. 2001; 13:1053–1061. [PubMed: 11470775]
151. Liu FL, Chen CH, Chu SJ, Chen JH, Lai JH, Sytwu HK, Chang DM. Interleukin (IL)-23 p19 expression induced by IL-1beta in human fibroblast-like synoviocytes with rheumatoid arthritis

- via active nuclear factor-kappaB and AP-1 dependent pathway. *Rheumatology (Oxford)*. 2007; 46:1266–1273. [PubMed: 17569750]
152. Meeran SM, Mantena SK, Meleth S, Elmets CA, Katiyar SK. Interleukin-12-deficient mice are at greater risk of UV radiation-induced skin tumors and malignant transformation of papillomas to carcinomas. *Molecular cancer therapeutics*. 2006; 5:825–832. [PubMed: 16648552]
153. Emanuel P, Scheinfeld N. A review of DNA repair and possible DNA-repair adjuvants and selected natural anti-oxidants. *Dermatology online journal*. 2007; 13:10. [PubMed: 18328204]
154. Majewski S, Jantschitsch C, Maeda A, Schwarz T, Schwarz A. IL-23 antagonizes UVR-induced immunosuppression through two mechanisms: reduction of UVR-induced DNA damage and inhibition of UVR-induced regulatory T cells. *The Journal of investigative dermatology*. 2010; 130:554–562. [PubMed: 19727119]
155. Zou W, Restifo NP. T(H)17 cells in tumour immunity and immunotherapy. *Nat Rev Immunol*. 2010; 10:248–256. [PubMed: 20336152]
156. Tominaga K, Yoshimoto T, Torigoe K, Kurimoto M, Matsui K, Hada T, Okamura H, Nakanishi K. IL-12 synergizes with IL-18 or IL-1beta for IFN-gamma production from human T cells. *International immunology*. 2000; 12:151–160. [PubMed: 10653850]
157. Gasse P, Riteau N, Vacher R, Michel ML, Fautrel A, di Padova F, Fick L, Charron S, Lagente V, Eberl G, Le Bert M, Quesniaux VF, Huaux F, Leite-de-Moraes M, Ryffel B, Couillin I. IL-1 and IL-23 Mediate Early IL-17A Production in Pulmonary Inflammation Leading to Late Fibrosis. *PLoS One*. 2011; 6:e23185. [PubMed: 21858022]
158. Mus AM, Cornelissen F, Asmawidjaja PS, van Hamburg JP, Boon L, Hendriks RW, Lubberts E. Interleukin-23 promotes Th17 differentiation by inhibiting T-bet and FoxP3 and is required for elevation of interleukin-22, but not interleukin-21, in autoimmune experimental arthritis. *Arthritis and rheumatism*. 2010; 62:1043–1050. [PubMed: 20131264]

INFLAMMASOME SUBSETS

Figure 1.

A. The inflammasome. The ligand binding scaffold subtypes bind their cognate agonist (DAMPs or PAMPs) that promotes heptameric oligomerization and multimeric assembly. Subfamilies are defined by domain structure and binding partner proteins. AIM2 (Absent In Melanoma 2); ASC, apoptosis-associated speck-like protein containing a caspase recruitment domain; CARD, caspase activation and recruitment domain; Casp1, enzymatically active caspase-1; dsDNA, double stranded deoxyribonucleic acid; IL-18, interleukin-18; IL-1β, interleukin-1β; LMP, lysosomal membrane permeabilization; MDP, muramyl dipeptide; NLRC4, NLR family CARD domain-containing protein 4; NLRP1, NOD-like receptor family, pyrin domain containing 1; NLRP3, NOD-like receptor family, pyrin domain containing 3; pro-Casp1, procaspase-1; PYD, pyrin domain; ROS, reactive oxygen species; ssRNA, single stranded ribonucleic acid.

B. The ligand binding scaffold subtypes bind their cognate agonist (DAMPs or PAMPs) which promotes heptameric oligomerization, multimeric assembly and ultimately activation of an active caspase-1 platform. The active caspase-1 platform is required to process pro-IL-1β and pro-IL-18 into their mature forms and secrete them into extracellular space. The inflammasome is also associated with unconventional secretion of alarmins IL-1α, IL-33 and HMGB-1.

Figure 2.

Downstream effects of UV on inflammasome assembly and secretion of cytokines. UV activates many alarmins which in turn activate the assembly of these inflammasome complexes and required for pro-inflammatory cytokine secretion.

Figure 3.

The role of IL-1 family cytokines in UVR-induced inflammation and immunosuppression. These cytokines affect different cell subtypes of innate and adaptive immunity and skew the immune system to favor activation, inflammation or tolerance.