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Human Keratinocyte Toll-like Receptors Promote Distinct Immune Responses

Lloyd S. Miller[1] and Robert L. Modlin[1,2]

It has been well established that Toll-like receptors (TLRs) are expressed by keratinocytes and respond to their respective ligands to initiate immune responses. However, it appears that keratinocytes, via differential activation of TLRs, may play a key role in determining the type of subsequent cutaneous immune response generated against a particular pathogen.

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Human Toll-like receptors (TLRs, numbered 1–10) are found on a variety of different cell types and can recognize various components of microorganisms, subsequently initiating signaling pathways important in the generation of cytokines, chemokines, antimicrobial peptides, and upregulation of adhesion and costimulatory molecules involved in innate and acquired immune responses (Kaisho and Akira, 2006). Previous studies have demonstrated that human keratinocytes express TLRs 1–6 and 9 (Kawai *et al.*, 2002; Mempel *et al.*, 2003; Song *et al.*, 2002; Pivarcsi *et al.*, 2003; Baker *et al.*, 2003; Miller *et al.*, 2005; Kollisch *et al.*, 2005). In addition, some of these studies have demonstrated that TLRs on keratinocytes are functional and respond to their respective ligands to produce cytokines, and chemokines, and to activate NF-κB. For example, several studies have reported that TLR2 and TLR4 are expressed by human keratinocytes and can be activated by their ligands, bacterial lipopeptides and lipopolysaccharide, respectively (Kawai *et al.*, 2002; Mempel *et al.*, 2003; Song *et al.*, 2002; Pivarcsi *et al.*, 2003; Kollisch *et al.*, 2005). Furthermore, additional studies have demonstrated that TLR3 and TLR5 are also expressed by human keratinocytes and can be activated by their ligands, double-stranded RNA (poly-I:C) and bacterial flagellin, respectively (Dai *et al.*, 2006; Baker *et al.*, 2003; Miller *et al.*, 2005; Kollisch *et al.*, 2005). Lastly, previous studies have also identified that human keratinocytes express TLR9 and can respond to CpG motifs of bacterial DNA (Mempel *et al.*, 2003; Miller *et al.*, 2005).

Lebre *et al.* (2007, this issue) confirm previous reports demonstrating that human keratinocytes express certain TLRs, including TLRs 1–6, 9, and 10. A particular strength of this study is that the authors evaluated TLR expression and activation on cutaneous keratinocytes derived from plastic surgery skin specimens and not mucosa-derived keratinocytes from human foreskin specimens. Taken together, these data provide additional evidence that keratinocytes not only act as a barrier to infectious microorganisms but also detect components of these organisms and initiate immune responses via activation of TLRs. However, the authors also demonstrate that certain immune responses generated by activation of TLRs 3, 4, 5, and 9 on keratinocytes are indeed distinct (summarized in Table 1). Although activation of TLRs 3, 4, 5, and 9 all induced expression of the proinflammatory cytokine tumor necrosis factor-α, the neutrophil chemotactic factor IL-8 (CXCL8), the monocyte and basophil chemokine CCL2, and macrophage inflammatory protein-3 (CCL20), of particular interest was the differential production of chemokines by these TLRs. Activation of TLR3 and TLR5 selectively induced CCL27, a chemokine that promotes memory T-

> TLRs on keratinocytes may participate in immune responses and host defense against viruses and bacteria.

[1]*Division of Dermatology, David Geffen School of Medicine at UCLA, Los Angeles, California, USA; and* [2]*Department of Microbiology, Immunology and Molecular Genetics, David Geffen School of Medicine at UCLA, Los Angeles, California, USA*

Correspondence: Robert L. Modlin, Department of Microbiology, Immunology and Molecular Genetics, David Geffen School of Medicine at UCLA, 52-121 CHS, 10833 Le Conte Avenue, Los Angeles, California 90095, USA. E-mail: rmodlin@mednet.ucla.edu

Table 1. Activation of keratinocyte Toll-like receptors 3, 4, 5, and 9 results in differential production of cytokines and chemokines, and upregulation of adhesion, activation, and costimulatory molecules.

	TLR3	TLR4	TLR5	TLR9
Innate immune cytokines and chemokines				
TNF-α	+	+	+	+
IL-8 (CXCL8)	+	+	+	+
CCL2	+	+	+	+
CCL20/MIP-3α	+	+	+	+
Memory T-cell chemotaxis				
CCL27/CTACK	+	–	+	–
CXCL9/CXCL10	+	–	–	+
Type I IFN	+	–	–	+
Upregulation of ICAM-1, HLA-DR, HLA-ABC, FasR, and CD40	+	+	+	–
Translocation of NF-κBp65	+	+	+	+

FasR, Fas receptor; MIP-3α, macrophage inflammatory protein-3α; TLR, Toll-like receptor; TNF-α, tumor necrosis factor-α.

cell recruitment specifically to the skin. Activation of TLR3 and TLR9 selectively induced production of CXCL9 and CXCL10, which predominantly activate memory T cells and are associated with generation of T-helper type 1 immune responses. In addition, activation of TLR3 and TLR9 also induced production of type I interferons (IFN-α and IFN-β), which are known to be important in antiviral immune responses (Iwasaki and Medzhitov, 2004).

Lebre *et al.* (2007) discuss that the production of these chemokines demonstrates that TLRs on keratinocytes may participate in immune responses and host defense against viruses and bacteria. However, they do not discuss why the different TLRs might elicit different production of chemokines. It is known that the signaling pathways of TLRs are not identical. For example, TLR1 and TLR6 can heterodimerize with TLR2 to activate an adaptor molecule called MyD88 to initiate signaling (Kaisho and Akira, 2006). In addition, TLRs 4, 5, 6, 7, 8, and 9 also use MyD88 to initiate signaling (Kaisho and Akira, 2006). On the other hand, TLR4, in addition to using MyD88, also uses a different adaptor molecule called TRIF to initiate signaling (Kaisho and Akira, 2006). TLR3 only uses TRIF (and not MyD88) to initiate signaling (Kaisho and Akira, 2006). Unlike MyD88, TRIF activates a signaling pathway involving IRF3, resulting in direct production of type I IFN (Kaisho and Akira, 2006). Perhaps the different adaptor molecules used by these TLRs contribute to the different chemokines produced by keratinocytes. An alternative hypothesis is that the distinct immune responses generated by the different TLRs could be dependent on the cellular location of the TLRs. TLR4 and TLR5 are located on the surface of cells, whereas TLR3 and TLR9 are found in endosomes within the cytoplasm of cells (Kaisho and Akira, 2006). Therefore the activation of these TLRs from these different cellular compartments may lead to distinct signaling pathways, resulting in differential cytokine/chemokine production. In particular, in various cell types, the intracellular location of TLR3 and TLR9 has implicated these receptors as important in defense against intracellular pathogens such as viruses and intracellular bacterial infections (Kaisho and Akira, 2006; Iwasaki and Medzhitov, 2004). Lebre *et al.* (2007) have demonstrated that activation of these receptors by human keratinocytes leads to a predominant T-helper type 1 immune response and to the production of type I IFN, which have both been implicated in eliciting cell-mediated immunity against these infections.

Overall, this study by Lebre *et al.* (2007) further enhances our knowledge about the expression and function of keratinocyte TLRs and suggests that the immune responses generated by different keratinocyte TLRs are distinct. These distinct immune responses are likely to be critical in promoting different host defense mechanisms and inflammatory responses in the skin.

CONFLICT OF INTEREST

The authors state no conflict of interest.

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