addition, Tc17 cells do not produce IL-17 exclusively, as most CD8+IL-17+ cells also secrete TNF-α and about half produce IFN-γ along with IL-17 (Ortega *et al.*, 2009), a feature that can also be attributed to MAIT cells.

In the light of the information presented here, it is important to determine whether the IL-17+CD8+ T cells in psoriatic skin are conventional MHC class I–restricted T cells or belong to the MAIT subset. In this paper Teunissen *et al.* (2014) demonstrate that, although MAIT cells are found in psoriatic skin, they represent only a proportion of IL-17A-expressing CD8+ T cells and that the majority of Tc17 clones derived from psoriatic epidermis are conventional T cells. In addition, the investigators demonstrate that the frequency of Tc17 cells in peripheral blood correlates with disease severity, although they did not take the next step of determining whether the cells belong to the MAIT subset rather than being conventional CD8+ T cells. Taken together, these data suggest that MAIT cells are unlikely to have a major role in the pathogenesis of psoriasis and, instead, focus the spotlight back on conventional Tc17 cells as the critical CD8+ T-cell population in psoriasis pathogenesis. This, along with other evidence supporting the role of CD8+ T cells in psoriasis (Elder *et al.*, 2010), indicates that these cells may have been either selectively recruited to or expanded in the epidermis of psoriatic skin. Expansion might occur after recognition of antigens in the context of MHC class I, with HLA-Cw*0602 being most likely the restriction element in a large proportion of patients with psoriasis (Nair *et al.*, 2006). This is a relatively unexplored avenue of research in psoriasis, but addressing the nature of the antigen specificity of conventional CD8+ T cells in psoriasis is very attractive. It holds considerable promise because it is the most likely approach to eventually ''cure'' psoriasis.

CONFLICT OF INTEREST

The authors state no conflict of interest.

REFERENCES

Baker BS, Swain AF, Fry L *et al.* (1984) Epidermal T lymphocytes and HLA-DR expression in psoriasis. *Br J Dermatol* 110:555–64

Bonish B, Jullien D, Dutronc Y *et al.* (2000) Overexpression of CD1d by keratinocytes in psoriasis and CD1d-dependent IFN-gamma production by NK-T cells. *J Immunol* 165:4076–85

Conrad C, Boyman O, Tonel G *et al.* (2007) Alpha1beta1 integrin is crucial for accumulation of epidermal T cells and the development of psoriasis. *Nat Med* 13:836–42

Dusseaux M, Martin E, Serriari N *et al.* (2011) Human MAIT cells are xenobiotic-resistant, tissue-targeted, CD161hi IL-17-secreting T cells. *Blood* 117:1250–9

Elder JT, Bruce AT, Gudjonsson JE *et al.* (2010) Molecular dissection of psoriasis: integrating genetics and biology. *J Invest Dermatol* 130:1213–26

Kryczek I, Bruce AT, Gudjonsson JE *et al.* (2008) Induction of IL-17+ T cell trafficking and development by IFN-gamma: mechanism and pathological relevance in psoriasis. *J Immunol* 181:4733–41

Laggner U, Di Meglio P, Perera GK *et al.* (2011) Identification of a novel proinflammatory human skin-homing Vgamma9Vdelta2 T cell subset with a potential role in psoriasis. *J Immunol* 187:2783–93

Le Bourhis L, Guerri L, Dusseaux M *et al.* (2011) Mucosal-associated invariant T cells: unconventional development and function. *Trends Immunol* 32:212–8

Lin AM, Rubin CJ, Khandpur R *et al.* (2011) Mast cells and neutrophils release IL-17 through extracellular trap formation in psoriasis. *J Immunol* 187:490–500

Nair RP, Stuart PE, Nistor I *et al.* (2006) Sequence and haplotype analysis supports HLA-C as the psoriasis susceptibility 1 gene. *Am J Hum Genet* 78:827–51

Ortega C, Fernández-A S, Carrillo JM *et al.* (2009) IL-17-producing CD8+ T lymphocytes from psoriasis skin plaques are cytotoxic effector cells that secrete Th17-related cytokines. *J Leukoc Biol* 86:435–43

Res PC, Piskin G, de Boer OJ *et al.* (2010) Overrepresentation of IL-17A and IL-22 producing CD8 T cells in lesional skin suggests their involvement in the pathogenesis of psoriasis. *PLoS ONE* 5:e14108

Teunissen MB, Yeremenko NG, Baeten DL *et al.* (2014) The IL-17A-producing CD8 T cell population in psoriatic lesional skin comprises mucosa-associated invariant T cells and conventional T cells. *J Invest Dermatol* 134:2898–907

Valdimarsson H, Thorleifsdottir RH, Sigurdardottir SL *et al.* (2009) Psoriasis—as an autoimmune disease caused by molecular mimicry. *Trends Immunol* 30:494–501

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Manipulated Microenvironment in Human Papilloma Virus–Infected Epithelial Cells: Is the CD40–CD154 Pathway Beneficial for Host or Virus?

Takatoshi Shimauchi[1] and Vincent Piguet[1]

In this issue, Tummers *et al.* (2014) demonstrate that high-risk human papilloma viruses (hrHPVs) attenuate the magnitude of responses to CD40 ligation and the epithelial cells' (ECs) capacity to attract leukocytes. These results suggest that hrHPVs can escape from host immune surveillance by modulating pro-inflammatory responses in infected ECs, resulting in persistent infections and potential carcinogenesis.

Journal of Investigative Dermatology (2014) **134,** 2866–2868; doi:10.1038/jid.2014.357

The human papilloma virus (HPV) is a non-enveloped double-stranded DNA virus that infects human epithelial tissues, including skin, anogenital epithelia, and oral cavity mucosa. In particular, hrHPVs, including HPV16 and

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Clinical Implications

- Activation of epithelial cells (ECs) via CD40 induces immune-related, anti-proliferative, and anti-apoptotic genes, and the capacity to cause the migration of immune cells.
- High-risk human papilloma viruses (hrHPVs) attenuate the magnitude of responses to CD40 ligation, including a reduced capacity of ECs to attract immune cells.
- The CD40–CD154 pathway has an important role in protective epithelial immune responses to infection.

HPV18, are associated with cervical cancer, which is the second most common cancer in women.

To eliminate viral invasion, skin-resident cells, including keratinocytes, Langerhans cells (LCs), dermal dendritic cells (DCs), and macrophages, express many pattern-recognition receptors such as C-type lectin receptors, Toll-like receptors (TLRs), nucleotide-binding oligomerization domain–like receptors, retinoic acid–inducible gene I–like receptors (RLRs), and cytosolic DNA sensors, which can detect pathogen-associated molecular patterns of the invading viruses, which, in turn, activate anti-viral immune responses (Kawamura *et al.*, 2014). Because keratinocytes in the basal layer of the epithelia are the major targets of HPV, the innate and adaptive immune responses by ECs have important roles in eliminating HPV. For instance, ECs express several TLRs on their cell surfaces (TLR-1, TLR-2, TLR-5, TLR-6, and TLR-10) or in endosomes (TLR-3 and TLR-9), and their activation initiates signaling pathways that result in both innate and adaptive immune responses. In particular, TLR-9 recognizes double-stranded CpG-rich DNA, and the activation of TLR-9 in ECs results in the production of cytokines and chemokines such as tumor necrosis factor-α, IL-8, MCP-1, MIP-3α, MIG, and IFN-1(Lebre *et al.*, 2007). The consequent pro-inflammatory microenvironments within the epithelia lead to the recruitment of DCs, LCs, T cells, and Natural killer cells, which, in turn, help eliminate HPV-infected keratinocytes.

However, HPV can also escape from host immune surveillance by downregulating this pro-inflammatory response in ECs using two principal oncoproteins E6 and E7 (Li *et al.*, 1999; Um *et al.*, 2002). In recent studies, investigators in this same laboratory have demonstrated that hrHPV dampens a network of genes encoding chemotactic, pro-inflammatory, and anti-microbial cytokines in ECs that is activated by TLR-3, protein kinase R, RLRs, and melanoma differentiation–associated gene 5 (Karim *et al.*, 2011). They have also demonstrated mechanisms by which hrHPV inhibits these responses in ECs. hrHPV can upregulate a cellular protein called ubiquitin carboxyl–terminal hydrolase L1 (UCHL1), which suppresses activation of signals downstream of the pathogen-recognition receptor, leading to reduced

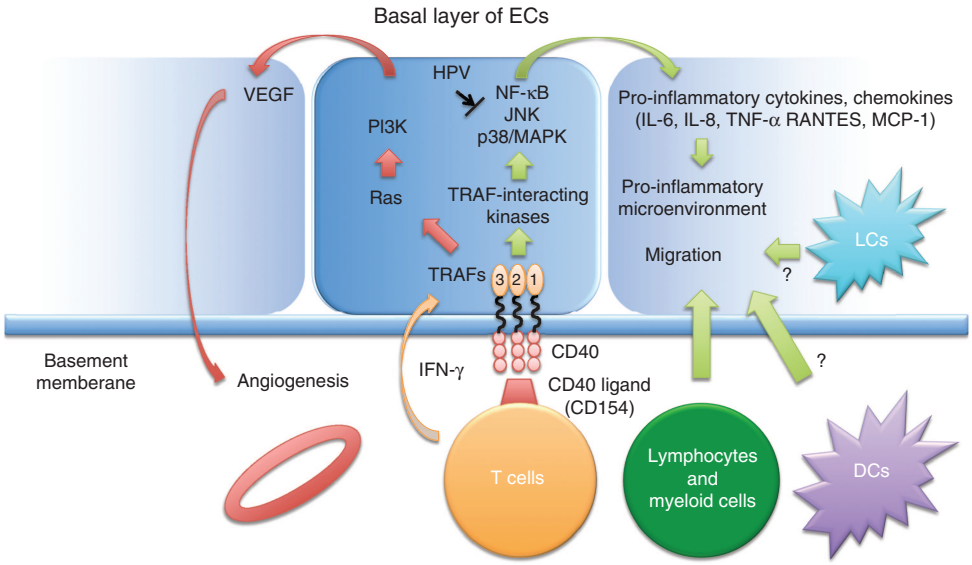

Figure 1. Potential immune responses in endothelial cells (ECs) via CD40–CD154 activation. INF-γ-induced CD40-expressing ECs can interact with activated, CD40 ligand (CD154)–expressing T lymphocytes. Engagement of CD40 causes conformational changes that recruit adapter molecules known as TRAFs. TRAFs recruit TRAF-interacting kinases and activate the NF-κB, p38/MAPK, and JNK pathways. CD40 signaling induces the production of pro-inflammatory cytokines and chemokines required for the migration of immune cells such as LCs, DCs, and leukocytes, resulting in the establishment of a pro-inflammatory microenvironment. The high-risk human papilloma virus (hrHPV) interferes with CD40 signaling and attenuates these responses. CD40 ligation also induces the activation of Ras and PI3K, leading to the production of VEGF, which can promote angiogenesis in the dermis. DCs, dendritic cells; JNK, c-JUN-NH_2-kinase; LCs, Langerhans cells; NF-κB, nuclear factor-κB; p38/MAPK, p38/mitogen-activated protein kinase; PI3K, phosphatidylinositol 3-kinase; TRAF, tumor necrosis factor receptor associated factors; VEGF, vascular endothelial growth factor.

transcription factor activation and downstream gene expression (Karim *et al.*, 2013). In addition, these findings suggest an important role for ECs as initiators of immune responses against HPV and as a link to the adaptive immune response.

In the current issue of *JID*, Tummers *et al.* (2014) demonstrate that IFN-γ-induced CD40-expressing ECs react in a coordinated way to CD40 ligation with the induction of immune-related genes by using genome-wide expression profiling. They also confirm that IL-8 and RANTES, produced by CD40-ligated ECs, have a capacity to cause immune cell migration. hrHPVs attenuate the magnitude of the response to CD40 ligation and the ECs' capacity to attract immune cells. UCHL1 may explain the mechanisms of impaired CD40 signaling in rhHPV-infected ECs.

A similar *in vitro* study has shown previously that CD40 ligation can induce the production of RANTES, MCP-3, and IP-10 in both nonmalignant and malignant HPV-positive cervix cell lines when stimulated with IFN-γ (Altenburg *et al.*, 1999). In this regard, a key finding in Tummers *et al.* (2014) is that CD40 ligation of ECs can recruit lymphocytes, as well as myeloid cells, and this process is attenuated by hrHPV infection (Figure 1). This is an important finding because it was previously unclear whether HPV infection could inhibit the migration of immune cells into the ECs in a human model.

CD40 is expressed on ECs in a wide variety of tissues. Its expression and function in these cells contribute to their role as immune effectors within local microenvironments (Dugger *et al.*, 2009). Engagement of epithelial CD40 activates these cells and results in the release of pro- and anti-inflammatory mediators (Dugger *et al.*, 2009). However, CD40 is also found in some tumor cells, and engagement via the CD40 ligand can lead to the production of vascular endothelial growth factor (VEGF) and fibroblast growth factor, resulting in the promotion of angiogenesis (Bergmann and Pandolfi, 2006; Figure 1). These two functions of CD40, promoting immune responses and promoting angiogenesis, have opposite effects on the growth of tumors (Bergmann and Pandolfi, 2006). *In vivo* studies have shown that CD40 expression is significantly higher in cervical carcinomas than in cervicitis and in the normal cervix, and CD40 ligand was detected on a subpopulation of tumor-infiltrating lymphocytes (Altenburg *et al.*, 1999; Huang *et al.*, 2011). In addition, the expression of CD40 is also correlated significantly with HPV, VEGF expressions, and microvessel density (Huang *et al.*, 2011). Thus, in the case of HPV-infected cervical carcinoma, there is a possibility that the CD40–CD154 pathway contributes to angiogenesis via VEGF production, resulting in tumor progression and metastasis. Therefore, the situation *in vivo* is even more complex, and it is intriguing to question whether activation of the CD40–CD154 pathway in HPV-infected ECs is beneficial for host immune surveillance and/or whether it promotes HPV carcinogenesis.

Two prophylactic HPV vaccines (Gardasil and Cervarix) are currently available and have been shown in clinical trials to have a high efficacy for the prevention of infection and associated diseases attributed to HPV types targeted by the vaccine. However, both vaccines provide no protection for women already infected with HPV16 and 18 serotypes (Nikolic and Piguet, 2010). The administration of adjuvants such as TLR agonists, including lipopolysaccharide (TLR-4 agonist), 3 M-002 (TLR-8 agonist), resiquimod (TLR-7 and 8 agonist), and oligodeoxinucleotide (TLR-9 agonist), shows promise for the elimination of HPV from ECs in *in vitro* studies or in a mouse model (Amador-Molina *et al.*, 2013). However, as shown by the investigators and their research groups, pro-inflammatory immune responses against HPV are downregulated in HPV-infected ECs (Karim *et al.*, 2011; Karim *et al.*, 2013; Tummers *et al.*, 2014). Thus, new adjuvants or other molecules that can improve the microenvironment surrounding HPV infection will be beneficial in treating HPV. In order to confirm a potential use for CD40 ligand, either alone or in combination with other adjuvants to treat HPV, preclinical animal models will be required. Results from such studies may have important implications for the treatment of HPV-associated diseases.

CONFLICT OF INTEREST

The authors state no conflict of interest.

ACKNOWLEDGMENTS

This work was supported by Cardiff University, a Wellcome Trust Institutional Strategic Support Fund to Cardiff University (506398), and a Gates Foundation Grant to VP.

REFERENCES

Altenburg A, Baldus SE, Smola H *et al.* (1999) CD40 ligand-CD40 interaction induces chemokines in cervical carcinoma cells in synergism with IFN-gamma. *J Immunol* 162:4140–7

Amador-Molina A, Hernandez-Valencia JF, Lamoyi E *et al.* (2013) Role of innate immunity against human papillomavirus (HPV) infections and effect of adjuvants in promoting specific immune response. *Viruses* 5:2624–42

Bergmann S, Pandolfi PP (2006) Giving blood: a new role for CD40 in tumorigenesis. *J Exp Med* 203:2409–12

Dugger K, Lowder TW, Tucker TA *et al.* (2009) Epithelial cells as immune effector cells: the role of CD40. *Semin Immunol* 21:289–92

Huang Q, Qu QX, Xie F *et al.* (2011) CD40 is overexpressed by HPV16/18-E6 positive cervical carcinoma and correlated with clinical parameters and vascular density. *Cancer Epidemiol* 35:388–92

Karim R, Meyers C, Backendorf C *et al.* (2011) Human papillomavirus deregulates the response of a cellular network comprising of chemotactic and proinflammatory genes. *PLoS One* 6:e17848

Karim R, Tummers B, Meyers C *et al.* (2013) Human papillomavirus (HPV) upregulates the cellular deubiquitinase UCHL1 to suppress the keratinocyte's innate immune response. *PLoS Pathog* 9:e1003384

Kawamura T, Ogawa Y, Aoki R *et al.* (2014) Innate and intrinsic antiviral immunity in skin. *J Dermatol Sci* doi:10.1016/j.jdermsci.2014.05.004 (e-pub ahead of print)

Lebre MC, van der Aar AM, van Baarsen L *et al.* (2007) Human keratinocytes express functional Toll-like receptor 3, 4, 5, and 9. *J Invest Dermatol* 127:331–41

Li S, Labrecque S, Gauzzi MC *et al.* (1999) The human papilloma virus (HPV)-18 E6 oncoprotein physically associates with Tyk2 and impairs Jak-STAT activation by interferon-alpha. *Oncogene* 18:5727–37

Nikolic DS, Piguet V (2010) Vaccines and microbicides preventing HIV-1, HSV-2, and HPV mucosal transmission. *J Invest Dermatol* 130:352–61

Tummers B, Goedemans R, Jha V *et al.* (2014) CD40-mediated amplification of local immunity by epithelial cells is impaired by HPV. *J Invest Dermatol* 134:2918–27

Um SJ, Rhyu JW, Kim EJ *et al.* (2002) Abrogation of IRF-1 response by high-risk HPV E7 protein *in vivo*. *Cancer Lett* 179:205–12