Psoriasis Triggered by Toll-like Receptor 7 Agonist Imiquimod in the Presence of Dermal Plasmacytoid Dendritic Cell Precursors

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Background: It has been proposed that the innate immune system plays a central role in driving the autoimmune T-cell cascade leading to psoriasis; however, there is no direct evidence for this.

Observations: We observed aggravation and spreading of a psoriatic plaque when treated topically with the toll-like receptor (TLR) 7 agonist imiquimod. The exacerbation of psoriasis was accompanied by a massive induction of lesional type I interferon activity, detected by MxA expression after imiquimod therapy. Since imiquimod induces large amounts of type I interferon production from TLR7-expressing plasmacytoid dendritic cell precursors (PDCs), the natural interferon-producing cells of the peripheral blood, we asked whether PDCs are present in psoriatic skin. We identified high numbers of PDCs in psoriatic skin lesions (up to 16% of the total dermal infiltrate) based on their coexpression of BDCA2 and CD123. By contrast, PDCs were present at very low levels in atopic dermatitis and not detected in normal human skin.

Conclusions: This study shows that psoriasis can be driven by the innate immune system through TLR ligation. Furthermore, our finding that large numbers of PDCs infiltrate psoriatic skin suggests a role of lesional PDCs as type I interferon-producing targets for the TLR7 agonist imiquimod.

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ducer of type I IFNs in response to imiquimod is represented by plasmacytoid dendritic cell precursors (PDCs), a novel subset of lymphoid-related cells selectively expressing high levels of TLR7 and TLR9. Plasmacytoid dendritic cell precursors are key players in innate antiviral immunity owing to their unique ability to produce large amounts of type I IFN. In addition, on viral stimulation, PDCs differentiate into dendritic cells with the ability to stimulate T-cell–mediated adaptive immunity. The unique link between innate and adaptive immunity has recently generated great interest in the use of imiquimod and other synthetic TLR agonists as adjuvants in cancer immunotherapy strategies. However, the observation that, in murine models, TLR activation may also trigger T-cell–mediated autoimmune disease recommends caution.

We report herein a case of psoriasis exacerbated by topical imiquimod therapy, identify large numbers of PDCs in the dermal infiltrate of psoriasis, and suggest that, as IFN-producing targets of TLR-ligands such as imiquimod, PDCs may represent a crucial link between the innate and adaptive immune responses ultimately leading to psoriasis.

**REPORT OF A CASE**

A 58-year-old white man presented with a 3-month history of an erythematous lesion on his back. Physical examination revealed a solitary, well-demarcated erythematous plaque measuring 3.5 × 3.0 cm with an irregular border and surface scaling (Figure 1A). The finding of parakeratosis with some degree of disorganization of the epidermal architecture on histological examination led initially to the diagnosis of Bowenoid keratosis. Because surgical excision was not suitable as a first-line treatment owing to the size of the lesion, daily application of 5% imiquimod cream (Aldara) was initiated. After 1 week of treatment, the lesion became erosive. Thereafter, the frequency of therapy was reduced to 2 to 3 times weekly. Following this, the lesion became scaly and increased in size. At week 5 of treatment, the erythematous plaques measuring 5.0 × 3.0 cm (Figure 1B), after 10 weeks the enlarged lesion measuring 7.0 × 7.0 cm was surrounded by small erythematous satellite lesions (Figure 1C) and distant, dropletlike lesions appeared disseminated throughout the trunk and the lower extremities reminiscent of a psoriasis guttata. Histological examination at week 10 showed a fully fledged psoriatic phenotype with parakeratosis, acanthosis, focal loss of the granular layer, and papillomatosis (Figure 2B). Furthermore, the diagnosis of psoriasis was confirmed by prompt regression of the lesion by treatment with topical calcipotriol cream (Daivonex; Leo Pharma Nordic, Malmo, Sweden) twice a day in combination with 311-nm UV-B therapy 3 times a week. Histological reevaluation of the initial lesion by 2 independent investigators did
not confirm the diagnosis of bowenoid keratosis, but was rather compatible with the diagnosis of psoriasis (Figure 2A). Hence, imiquimod treatment of a primary psoriasis lesion induced (1) a fully fledged histological psoriasis phenotype with a significant increase in acanthosis and papillomatosis (Figure 2C), (2) a greater than 4-fold enlargement of the surface of the psoriatic lesion, and (3) the spreading of psoriatic lesions to the immediate surroundings as well as to distant sites, with the clinical aspect of a psoriasis guttata.

**IMMUNOHISTOLOGICAL ANALYSIS**

Cryostat sections, prepared from frozen tissue specimen, were fixed in acetone and subsequently stained using a standard alkaline phosphatase anti–alkaline phosphatase technique. Briefly, after blocking of nonspecific binding sites with normal rabbit serum, tissue sections were incubated with an excess of primary antibodies, followed by 3 cycles of sequential incubations with rabbit anti–mouse IgG xenoantibodies conjugated to alkaline-phosphatase and anti–alkaline phosphatase–staining for MxA and counterstain with hematoxylin as described in the “Methods” section.

**RESULTS**

Imiquimod is known to exert its immunomodulatory properties principally through the induction of type I IFN. To detect specific changes induced by imiquimod, we investigated lesional type I IFN activity by immunohistochemical examination for MxA, an adenosine triphosphatase selectively induced in response to IFN-α/β and thus representing a surrogate marker for lesional type I IFN activity. While MxA expression was absent before imiquimod treatment (Figure 3A), a massive dermal and epidermal expression of MxA was detected throughout the treated lesion at week 10 (Figure 3B). Given that peripheral blood PDCs express high levels of TLR7 and represent the principal source of type I IFN in response to imiquimod, we sought to investigate whether PDCs are present in psoriatic skin lesions by immunohistochemical examination for BDCA2, a specific marker for blood PDCs. In human lymph node sections, the specificity of BDCA2 for tissue PDC was shown by the typical location of BDCA2-stained cells around high endothelial venules in the T-cell areas and not B-cell areas (Figure 4A). Staining of chronic plaque psoriasis samples revealed high numbers of BDCA2-positive cells among the dermal cellular infiltrate (range, 2.3%-16.9% of the cellular infiltrate; mean 8.0%; n=8) (Figure 4B and Figure 5A). An unequivocal PDC phenotype was demonstrated by the co-staining of BDCA2-positive cells with CD123 (interleukin 3 receptor α-chain), a marker that is highly expressed on blood PDCs, but not with the T-cell marker CD3 in 3-color confocal microscopy (Figure 4C). Among the samples tested, we did not detect any significant epidermotropism of PDCs. By contrast, PDCs were undetectable in normal human skin (n=5) and were present at significantly lower levels in atopic dermatitis (range, 0.6%-1.8%; mean 0.9%; n=5) (Figure 5A), an inflammatory skin disease with a comparable amount of infiltrate to psoriasis (Figure 5B). Interestingly, psoriatic skin, normal skin, and atopic dermatitis all had comparable percentages of CD1c+ dermal myeloid dendritic cells (Figure 5C), suggesting that the presence of PDCs is characteristic of psoriatic skin and may result from a specific recruitment. Since anti-
BDCA2 antibody is not suitable for staining of paraffin-embedded tissue sections, staining of imiquimod-triggered psoriasis was performed by using anti-CD123 mAb. CD123 was expressed in both PDCs and endothelial cells in control lymph node sections (Figure 6A) and psoriatic skin samples. However, we were able to identify large numbers PDCs in imiquimod-treated psoriatic skin based on their strong CD123 expression and plasmacytoid morphology, which allowed clear distinction from the elongated CD123dim endothelial cells (Figure 6B).

**COMMENT**

Psoriasis is a chronic-relapsing T-cell–mediated autoimmune disease, in which type 1 cytokine secretion by T cells induces keratinocyte hyperproliferation in geneti-
There is growing interest in the role of the innate immune system in the pathogenesis of psoriasis. The recent finding that TLR7 expression on MDCs can be increased by IFN-α has suggested that, in vivo, there might be a coordinated response to imiquimod between PDCs and MDCs. Plasmacytoid dendritic cell precursors represent the primary target of imiquimod and induce thereafter, through their secretion of IFN-α, a broader immune response by up-regulating TLR7. It is not known whether other cell types in the human skin represent primary targets of imiquimod through the expression of TLR7. In situ analysis of TLR7 expression in human skin will require the availability of a TLR7-specific antibody suitable for immunohistochemical examination.

Our present report provides direct evidence for the ability of the innate immune system to drive psoriasis through the TLR7 agonist imiquimod. To our knowledge, this represents the first description of an autoimmune-related disorder triggered by defined TLR activation in humans. The induction of a strong lesional type I IFN activity and our finding of large numbers of PDCs infiltrating psoriatic skin lesions suggest that PDCs might represent targets for the TLR7 agonist imiquimod in psoriasis. Whether PDCs represent key cellular mediators of innate immune responses driving the pathogenic events leading to psoriasis needs to be determined in future studies. The experimental proof of this model may provide the basis for new therapeutic approaches targeting upstream events in the pathogenesis of psoriasis.

CONCLUSIONS

Figure 6. Large numbers of plasmacytoid dendritic cell precursors (PDCs) in the dermal infiltrate of imiquimod-triggered psoriasis. A, CD123 staining of cryopreserved human lymph node sections shows in addition to PDC staining of endothelial cells (arrows) in the T-cell area (b indicates B-cell areas of the lymph node; t, T-cell areas of the lymph node). B, Immunohistochemical staining of paraffin-embedded tissue specimen of imiquimod-triggered psoriasis at week 10 stained for CD123 shows a large number of CD123+ PDCs in the dermal papillae. A clear distinction from endothelial cells (arrows) can be made by the strength of CD123 positivity and their typical plasmacytoid morphology. (Alkaline phosphatase anti–alkaline phosphatase–staining for CD123; original magnification x400; scale bars correspond to 40 µm.)

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