Research letter

T-cell responses against 4-*tert*-butylphenol-exposed pigmented cells in a patient with occupational vitiligo

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Dear Editor, Several case studies on contact or occupational vitiligo after skin contact with 4-*tert*-butylphenol (4-TBP) have been reported.[1,2] Biochemical and cellular effects of 4-TBP on melanocytes have been shown *in vitro*,[3–6] but immunological analyses explaining the immunizing ability of 4-TBP leading to widespread vitiligo lesions beyond areas of primary contact are still lacking. We report here the presence of a systemic T-cell response to 4-TBP-exposed pigmented cells in a patient with chemically induced vitiligo.

A 54-year-old man with Fitzpatrick skin type V presented with vitiligo after contact of the hands and arms with the two-component adhesive Armaflex, containing 4-TBP, during work as an insulation installer at a shipyard (Fig. 1). He first developed dermatitis at the contact sites, which depigmented 4 weeks later. Although further contact with Armaflex adhesive was avoided, he developed white patches on the dorsal site of his feet after wearing plastic sandals. After 3 months the depigmented lesions were still spreading beyond the contact sites and new white macules occurred spontaneously on his chest (Fig. 1). The patient had a family history of type 1 diabetes and early hair greying, but no family history of vitiligo or thyroid disease.

Histopathology of a depigmented lesional skin biopsy showed a normal epidermis with a dermal perivascular lymphocytic infiltrate, pigment-containing macrophages and the absence of melanocytes. Patch tests with Armaflex and the European Standard Series showed contact allergy to Armaflex and to 4-TBP, followed by depigmentation of both patch test areas 7 weeks later, with similar histopathology. Considering the negative family history of vitiligo or thyroid disease, a Koebner reaction by 4-TBP that reactivated a pre-existent occult vitiligo seems unlikely. Three months of treatment with topical tacrolimus followed by narrowband ultraviolet B therapy for 6 months resulted in repigmentation of his face, but not of the extremities. Afterwards, the vitiligo remained stable for 3 years and was successfully treated by autologous punch grafting. Nine years after initial contact with 4-TBP and the onset of vitiligo, the disease was still stable, and blood samples for immunological analyses were taken upon informed consent.

Our *in vitro* analyses confirmed that 4-TBP inhibited melanin synthesis in pigmented cells (data not shown).[3] To investigate the presence of systemic immunity against TBP-exposed melanocytes, we stimulated peripheral blood T cells with autologous dendritic cells loaded with overnight 4-TBP-exposed pigmented melanoma cells, as described previously for monobenzone.[7] Increased $CD8^+$ T-cell reactivity against 4-TBP-exposed cells was found in the patient, compared with a panel of healthy donors (data available on request). In addition, moderate reactivity against unexposed cells was seen in the patient, which may mediate the systemic spread of the vitiligo to distant, unexposed body sites. The moderate levels probably reflect the stable disease activity at the time of blood sampling. The $CD8^+$ T-cell response of the patient predominantly produced interferon-γ upon 4-TBP stimulation, corresponding to the T-cell responses found in vitiligo. No $CD4^+$ T-cell reactivity was observed in the patient. Both $CD8^+$ and $CD4^+$ T-cell reactivity to 4-TBP-exposed cells could be induced in a healthy donor blood during 7 days of culture.

These T-cell responses were also reactive with the keratinocyte cell line HaCaT exposed to 4-TBP, indicating reactivity towards 4-TBP as a hapten in a contact dermatitis–delayed-type hypersensitivity reaction (DTH),[8] rather than being pigment cell specific. DTH reactions are predominantly mediated by $CD4^+$ and $CD8^+$ T-cell responses, found locally in sensitized skin, followed by dampening of the response by regulatory T cells producing interleukin (IL)-10. The depigmentation of the 4-TBP patch test site in the patient suggests that contact sensitization by 4-TBP can increase the immunogenicity of pigmented cells leading to autoimmunity and

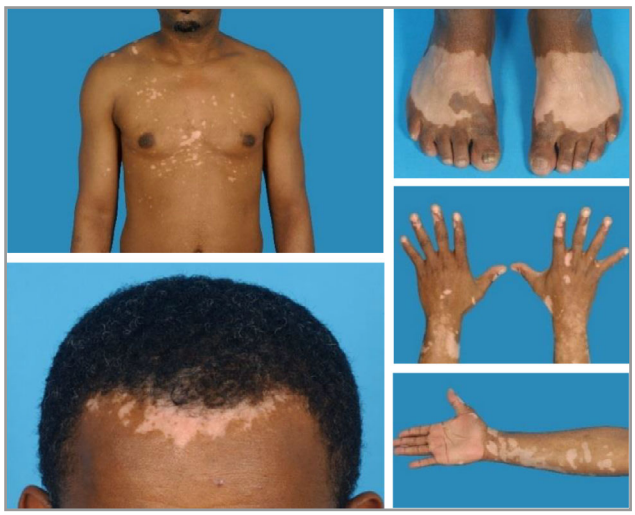

Fig 1. The patient with 4-*tert*-butylphenol-induced contact vitiligo.

vitiligo. The potential TBP reactivity in patients with stable vitiligo without a history of TBP exposure was not tested, as occult exposure to (similar) phenols cannot be ruled out, thereby obscuring conclusions on specificity.

The mechanism by which 4-TBP can induce vitiligo may involve oxidative stress in melanocytes and apoptotic cell death,[3,4] resulting in the release of melanocyte antigens that are taken up by dendritic cells. 4-TBP also activates the production of IL-6, IL-8 and heat shock protein 70, key molecules in the pathogenesis of vitiligo and other autoimmune diseases.[5,6] Presentation of melanocyte autoantigens by activated dendritic cells can trigger a cytotoxic $CD8^+$ T-cell response against melanocytes, leading to vitiligo.

In conclusion, we have shown increased $CD8^+$ T-cell reactivity against pigmented cells upon 4-TBP exposure in a patient with chemical-induced vitiligo. Our study adds to the knowledge of 4-TBP as a provoking factor for vitiligo. These findings emphasize the immunizing ability of 4-TBP against pigmented cells and the risk of developing vitiligo upon 4-TBP exposure in susceptible individuals.

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Conflicts of interest: none to declare.