Received: 19 April 2022 | Revised: 20 June 2022 | Accepted: 11 July 2022

DOI: 10.1002/cia2.12271

Journal of Cutaneous Immunology and Allergy

WILEY

CORRESPONDENCE

Role of pigment stem cells in hair follicles in the treatment of vitiligo with a 308-nm excimer laser: An immunohistochemical study of human cases

Pigment regeneration in vitiligo is often observed perifollicular following ultraviolet (UV) therapy, suggesting that a reservoir of melanocytes is present in the hair follicles move to the epidermis and differentiate into melanocytes.[1] While the 308-nm excimer laser is effective for vitiligo, the mechanisms underlying pigment regeneration are not yet clear. To investigate the relationship between pigment regeneration and pigment stem cells in vitiligo, we obtained biopsy specimens from a patient treated with the XTRAC Velocity 7™ (Strata Science). Immunohistochemical examination was performed with anti-cytokeratin 15 (anti-CK15 mouse monoclonal antibody, Clone: LKH15; Gene Tex Inc.) and anti-microphthalmia transcription factor (anti-MITF mouse monoclonal antibody, clone: D5; Neomarkers). CK15 is a marker of stem cells in the hair follicle bulge lesion,[2] and MITF is a melanocyte lineage marker expressed in melanoblasts and melanocytes.[3] This study aimed to determine the association between excimer laser irradiation and MITF-positive cells in the bulge region indicated as CK15-positive cells. CK15- and MITF-positive cells in the bulge lesion were considered melanoblasts. Biopsy specimens were obtained from the lower thigh of a male patient in his 70s, from a normal non-irradiated area, a pigment regeneration area with a follicular pattern, and a non-pigment regeneration area in vitiligo after 20 sessions of treatment. In samples from the normal non-irradiated area, bulge lesion in the follicle were indicated as CK15-positive cell (Figure 1A). MITF-positive cells were observed in the epidermis and hair follicles (Figure 1B). MITF-positive cells observed in the bulge lesions indicated CK15-positive cells were melanoblasts (Figure 1C). Similarly in samples from the pigment-regenerated area, pigment stem cells were indicated by CK15-positive cell (Figure 1D) and MITF-positive cells were observed in the epidermis and hair follicles (Figure 1E). MITF- and CK15-positive cells were considered melanoblasts observed in bulge lesion (Figure 1F). In contrast, although a few CK15-positive cells were observed in hair follicles (Figure 1G), MITF-positive cells were not observed in the bulge region and the epidermis in the non-pigment-regenerated area (Figure 1H). This result suggested that excimer laser irradiation may induce the differentiation of melanoblasts and melanocytes from bulge stem cells. The differentiation of melanocyte stem cells into melanoblasts in hair follicles is followed by the induction of epidermal melanocyte differentiation by repetitive UVB irradiation. Melanocyte stem cell differentiation is triggered by Wnt7a through β-catenin activation.[4] Excimer laser stimulates melanogenesis by acting on the Wnt/β-catenin signaling pathway in B16 cells.[5] The rate of UVB irradiation (mW/cm^2) plays a more important role than the dose of irradiation (mJ/cm^2) in determining light absorption by DNA and intracytoplasmic photoreceptors, as well as subsequent differentiation of immature pigment cells,[6] confirming the effect of excimer laser on vitiligo.

In conclusion, our study helps to elucidate the mechanism by which pigment stem cells in the bulge region differentiate into melanocytes and migrate to the epidermis in vitiligo following excimer

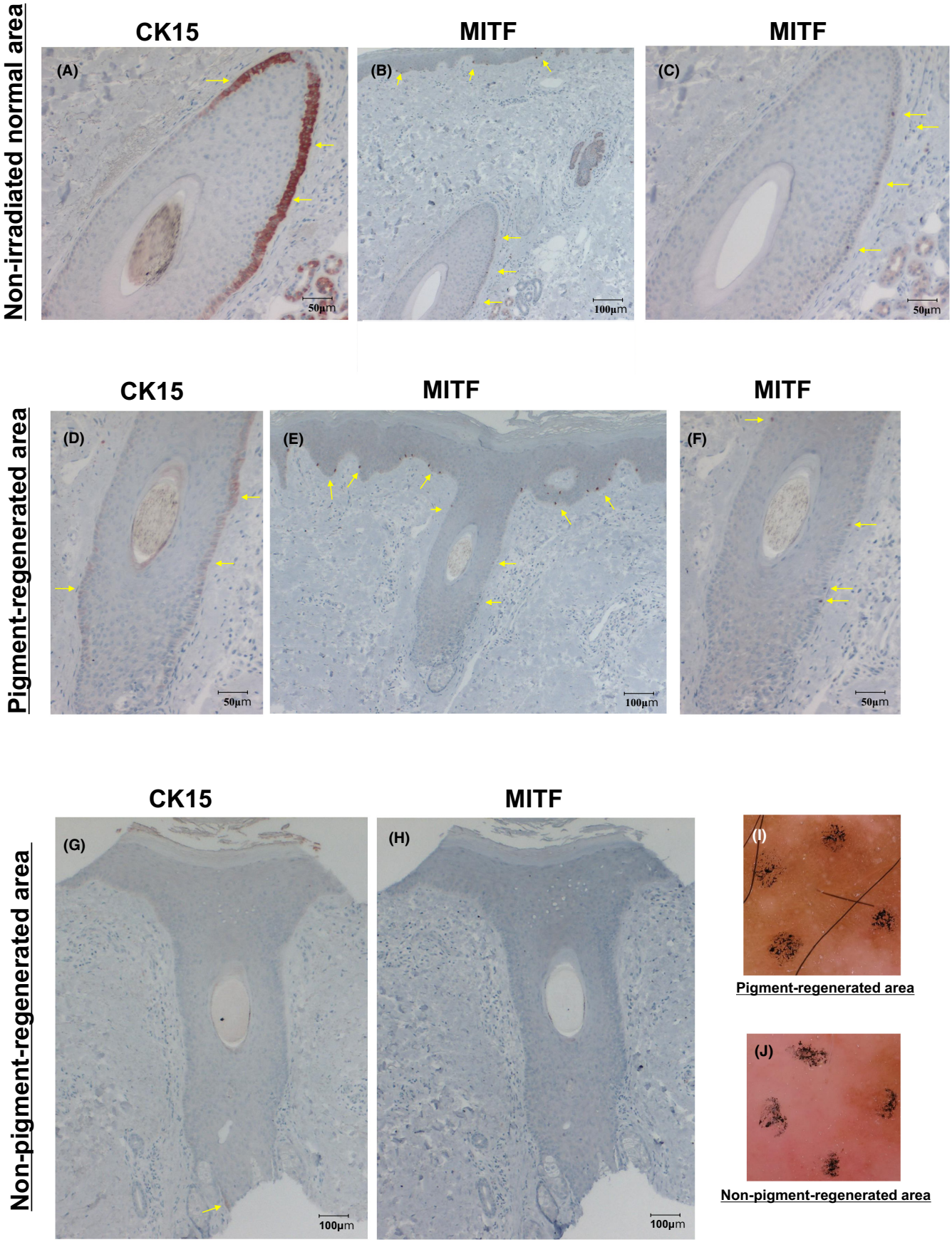
Non-irradiated normal area
CK15
MITF
MITF
(A)
(B)
(C)
50μm
100μm
50μm
Pigment-regenerated area
CK15
MITF
MITF
(D)
(E)
(F)
50μm
100μm
50μm
Non-pigment-regenerated area
CK15
MITF
(G)
(H)
100μm
100μm
(I)
Pigment-regenerated area
(J)
Non-pigment-regenerated area

FIGURE 1 Biopsy specimens obtained from the lower thigh of a male patient in his 70s. (A) CK15+ cells in the hair follicle bulge lesion in the sample from the non-irradiated normal area. (B) MITF+ cells in the epidermis and hair follicle bulge lesion in the sample from the non-irradiated normal area. (C) MITF+ cells in the hair follicle bulge lesion (high magnification). The yellow arrows indicate MITF+ cell. (D) CK15+ cells in the hair follicles in the sample from the pigment-regenerated area. (E) MITF+ cells in the epidermis and hair follicle in the sample from the pigment-regenerated area. (F) MITF+ cells in the hair follicle (high magnification). The yellow arrows indicate MITF+ cell. (G) CK15+ cells in the sample from the non-pigment-regenerated area after irradiation with the excimer laser. (H) MITF+ cells in the sample from the non-pigment-regenerated area after irradiation with the excimer laser. Dermoscopy: (I) Pigment-regenerated area, (J) non-pigment-regenerated area. Original magnification: (A, B, D, E, G, H) ×40; (C, F) ×100

laser irradiation. To the best of our knowledge, immunohistochemical examination of these factors in human cases of vitiligo has not yet been reported.

DECLARATION SECTION

Approval of the research protocol: This clinical study was approved by the Ethics Committee of Kansai Medical University Kori Hospital. Informed Consent: Informed consent was obtained from all patients. Registry and the Registration No.: This study was registered at the University Hospital Medical Information Network (UMIN ID: UMIN00032165).
Animal Studies: N/A.

CONFLICT OF INTEREST

The authors declare no conflict of interest. Dr. Akimichi Morita is a member of the Journal of Cutaneous Immunology and Allergy Editorial Board. The management of the peer-review process, and all editorial decision making, for this article, was undertaken by Editor-in-Chief.

Reiko Noborio MD, PhD[1]
Yuki Nomura MD[1]
Kahori Nakatani MD[1]
Takahiro Kiyohara MD, PhD[2]
Emi Nishida MD, PhD[3]
Motoki Nakamura MD, PhD[4]
Akimichi Morita MD, PhD[4]

[1]*Department of Dermatology, Kansai Medical University Kori Hospital, Neyagawa, Japan*
[2]*Department of Dermatology, Kansai Medical University Medical Centre, Moriguchi, Japan*
[3]*Department of Dermatology, Okazaki City Hospital, Okazaki, Japan*
[4]*Department of Geriatric and Environmental Dermatology, Nagoya City University Graduate School of Medical Sciences, Nagoya, Japan*

Correspondence
Reiko Noborio, MD, PhD, Department of Dermatology, Kansai Medical University Kori Hospital, 8-45 Kori Hondori-cho, Neyagawa, Osaka 572-8551, Japan.
Email: reikonobo@yahoo.co.jp

ORCID

Reiko Noborio https://orcid.org/0000-0002-7589-3478

REFERENCES

1. Cui J, Shen LY, Wang GC. Role of hair follicles on the repigmentation of vitiligo. J Invest Dermatol. 1991;97:410–6.
2. Lyle S, Liu Y, Christofidou-Solomidou M. Human hair bulge cells are biochemically distinct and possess an epithelial stem cell phenotype. Invest Dermatol. 1999;4:296–301.
3. Yamada T, Hasegawa S, Inoue Y. Comprehensive analysis of melanogenesis and proliferation potential of melanocyte lineage in solar lentigines. J Dermatol Sci. 2014;73:251–7.
4. Yamada T, Hasegawa S, Inoue Y, Date Y, Yamamoto N, Mizutani H, et al. Wnt/β-catenin and kit signaling sequentially regulate melanocyte stem cell differentiation in UVB-induced epidermal pigmentation. J Invest Dermatol. 2013;133:2753–62.
5. Li L, Liang Y, Zhang D, Wang C, Pan N, Hong J, et al. The 308-nm excimer laser stimulates melanogenesis via the Wnt/β-catenin signaling pathway in B16 cells. J Dermatol Treat. 2019;30:826–30.
6. Lan CC, Yu HS, Lu JH, Wu CS, Lai HC. Irradiance, but not fluence, plays a crucial role in UVB- induced immature pigment cell development: new insights for efficient UVB phototherapy. Pigment Cell Melanoma Res. 2013;26:367–76.