Received: 9 August 2023 | Revised: 15 September 2023 | Accepted: 20 September 2023

DOI: 10.1002/cia2.12332

Journal of Cutaneous Immunology and Allergy

WILEY

RESEARCH ARTICLE

Decreased CXCL14 expression in psoriasis recovered by narrow-band ultraviolet B therapy

Kaori Nakajima MD[1] | Tomomitsu Miyagaki MD, PhD[1] | Miho Tanaka MD[1] | Reo Komaki MD[1] | Tatsuro Okano MD, PhD[1] | Sora Takeuchi MD, PhD[1] | Hidenori Watabe MD, PhD[1,2] | Takafumi Kadono MD, PhD[1]

[1]Department of Dermatology, St. Marianna University School of Medicine, Kawasaki, Japan

[2]Haruhino Dermatology Clinic, Kawasaki, Japan

Correspondence
Tomomitsu Miyagaki, Department of Dermatology, St. Marianna University School of Medicine, 2-16-1 Sugao, Miyamae-ku, Kawasaki 216-0015, Japan.
Email: asahikari1979@gmail.com

Funding information
Ministry of Education, Culture, Sports, Science and Technology, Grant/Award Number: 20K08683 and 23K07797

Abstract

Background: CXCL14 is a member of CXC chemokine family, constitutively expressed in various normal tissues unlike many other chemokines. Other than the capacity to recruit natural killer cells, macrophages, and dendritic cells, CXCL14 suppresses CXCL12-CXCR4 interactions by inducing CXCR4 internalization. Thus, CXCL14 can both promote and hinder immune responses. Psoriasis is a chronic skin inflammatory disorder in which various chemokines play an important role.

Methods: To investigate possible roles of CXCL14 in psoriasis, we examined CXCL14 expression in lesional skin by immunohistochemistry and measured serum CXCL14 levels in psoriasis. We also assessed the effect of ultraviolet irradiation, one of the main therapies for psoriasis, on CXCL14 expression by HaCaT cells.

Results: CXCL14 expression was decreased in epidermal keratinocytes in lesional skin and serum CXCL14 levels were negatively correlated with Psoriasis Area and Severity Index scores in psoriasis patients. Serum CXCL14 levels were increased in nbUVB-treated psoriasis patients and UVB irradiation induced CXCL14 mRNA expression from HaCaT cells.

Conclusion: Our results suggest that decreased CXCL14 expression may contribute to the exacerbation of psoriasis and that the amplification of CXCL14 can be a therapeutic option for psoriasis. One of the mechanisms of the efficacy of nbUVB therapy in psoriasis may be the upregulation of CXCL14.

KEYWORDS
CXCL14, epidermal keratinocyte, psoriasis, psoriasis area severity index, ultraviolet B therapy

1 | INTRODUCTION

Psoriasis is a dendritic-cell (DC) and T-cell-mediated immunologic skin disease with a complex pathogenesis where both genetic and environmental factors are involved.[1–3] Clinically, psoriasis is characterized by well-demarcated reddish plaques with thick scale on various sites including the scalp, face, trunk, and extremities. Based on the recent advances in molecular-targeted therapies, crosstalk between the innate and adaptive immune system mediated by tumor necrosis factor (TNF)-α and IL-23 and preceding

activation of Th17 cells are considered to play an essential role in the onset and exacerbation of psoriasis.[3–5] In addition to such cytokines, various chemokines are also involved in the development of psoriasis via their capacities to induce chemotaxis of immune cells.[6] IL-17A induces neutrophil-attracting chemokines, such as CXCL1, CXCL2, and CXCL8, by epidermal keratinocytes and exacerbates skin inflammation in psoriasis.[7] IL-17A also upregulates the expression of CCL20, an essential chemoattractant of CCR6-positive Th17 cells, by epidermal keratinocytes, resulting in the augmentation of Th17-mediated responses.[8,9] Interactions between chemokines and chemokine receptors can also be therapeutic targets for psoriasis.

CXCL14, a member of CXC chemokine family, is constitutively expressed in various normal tissues, especially in the epithelia, unlike many other chemokines.[10,11] CXCL14 has the capacity to recruit natural killer cells, macrophages, and dendritic cells, but not naïve and activated T cells.[10,11] Other than chemotaxis of immune cells, CXCL14 has the capacity to suppress angiogenesis by hindering chemotaxis of endothelial cells, and exert antimicrobial activity.[12,13] More importantly, CXCL14 binds to CXCR4 with high affinity and inactivates CXCR4 through internalization, leading to the inhibition of CXCL12/CXCR4-mediated biological activities.[14] CXCL14 has been mostly investigated in malignant diseases since its discovery, because CXCL14 expression is decreased or completely lost in many human cancer specimens and cancerous cell lines.[15] Reflecting the multi-faced function of CXCL14, contradictory results have been reported on whether CXCL14 promotes or suppresses tumors depending on the types of malignancies, but CXCL14 is regarded as a therapeutic target in some malignant tumors.[10] On the other hand, CXCL14 involvement in inflammatory disorders has been scarcely investigated. In this study, we examined CXCL14 expression in psoriasis patients and its association with therapies for psoriasis to determine CXCL14 involvement in psoriasis.

2 | MATERIALS AND METHODS

2.1 | Patients

Twenty-nine untreated or topically treated psoriasis patients (mean ± SEM age; 64.3 ± 3.0 years, 16 males and 13 females), 16 psoriasis patients treated with biologics including anti-TNFα antibodies, anti-IL-12/23 p19 antibodies, anti-IL-17 antibodies, and anti-IL-23 p40 antibodies (mean ± SEM age; 54.6 ± 4.5 years, 10 males and 6 females), 13 psoriasis patients treated with oral systemic therapies including cyclosporin, etretinate, methotrexate, and apremilast (mean ± SEM age; 60.1 ± 3.7 years, 9 males and 4 females), 4 psoriasis patients treated with narrow-band ultraviolet B (nbUVB) (mean ± SEM age; 49.8 ± 6.6 years, 4 males and 0 females) and 23 healthy controls subjects (mean ± SEM age; 49.0 ± 3.1 years, 10 males and 13 females) were enrolled in this study. All psoriasis patients were diagnosed as psoriasis vulgaris and patients with pustular psoriasis and psoriasis arthritis were not included in this study. In 24 untreated or topically treated psoriasis patients, the disease severity was determined by the Psoriasis Area and Severity Index (PASI). The 23 healthy controls had no history of cutaneous inflammatory diseases. The medical ethical committee of St. Marianna University School of Medicine approved all described studies, and the study was conducted according to the Declaration of Helsinki Principles. All patients were provided written informed consent.

2.2 | Enzyme-linked immunosorbent assay

Serum and supernatant CXCL14 levels were quantified using Human CXCL14 ELISA (RayBiotech Life) according to the manufacturer's instructions. These assays employ the quantitative sandwich enzyme immunoassay technique.

2.3 | Immunohistochemistry

We performed immunohistochemical staining for CXCL14 using lesional skin of psoriasis ($n = 10$) and healthy skin ($n = 10$). These sections were stained with rabbit anti-human CXCL14 polyclonal antibody (Abcam) or isotype-matched control antibodies followed by ABC staining (Vector Lab). Diaminobenzidine was used for visualizing the staining, and counterstaining with Mayer hematoxylin was performed, according to the manufacturer's instructions. The CXCL14 immunostaining intensity was evaluated according to the following scores: score 0 (absent), score 1 (weak expression), score 2 (moderate expression), and score 3 (strong expression).

2.4 | In vitro experiments

HaCaT cells, a cell line of immortalized epidermal keratinocytes, (kindly provided by Dr Toshio Kuroki, Institute of Molecular Oncology, Showa University, Tokyo, Japan) were cultured in Eagle's minimum essential medium (Sigma-Aldrich) containing 10% fetal bovine serum, penicillin G sodium, streptomycin sulfate, and amphotericin B. When 80% confluence was achieved, the cells were trypsinized, washed, and resuspended in the medium at 2.5×10^6 cells/mL, and 280 μL was added to each well of the 6-well plates (Becton Dickinson Labware). When the cells reached confluence, they were irradiated with 312 nm UVB light (1.0 J/cm^2) using the BIO-LINK® crosslinker (Vilber). Total mRNA was obtained from cells with RNeasy Mini Kit (QIAGEN), 3 or 6 h after UVB irradiation. Complementary DNA was synthesized using ReverTra Ace qPCR RT Master Mix (TOYOBO). The mRNA levels were analyzed using quantitative RT-PCR with TaqMan™ Fast Advanced Master Mix (Thermo Fisher Scientific) or THUNDERBIRD SYBR qPCR Mix (TOYOBO). The mRNA levels were normalized to those of the GAPDH gene. The relative change in the levels of genes of interest was determined by the $2^{-\Delta\Delta C_T}$ method. Primers for human CXCL14 were purchased from Thermo Fisher Scientific. Primers for GAPDH were as follows: 5′-ACC CAC TCC TCC

ACC TTT GA-3′ as the forward primer and 5′-CAT ACC AGG AAA TGA GCT TGA CAA-3′ as the reverse primer.

2.5 | Statistical analysis

Statistical analysis was performed using the Mann–Whitney's *U*-test for comparison of two groups. For multiple comparison, Kruskal–Wallis test followed by Mann–Whitney's *U*-test was used. Correlation coefficients were determined using the Spearman's rank correlation test. *p*-values of <.05 were considered statistically significant.

3 | RESULTS

3.1 | Decreased CXCL14 expression in epidermal keratinocytes in psoriasis

We first examined CXCL14 expression in lesional skin of psoriasis patients by immunohistochemistry. We found that epidermal keratinocytes mainly expressed CXCL14 in healthy skin (Figure 1A) consistent with the previous report.[16] In psoriasis lesional skin, CXCL14 expression was decreased compared to healthy skin (Figure 1B). The intensity of CXCL14 staining in epidermal keratinocytes was significantly lower in psoriasis skin compared to healthy skin (Figure 1C).

3.2 | Negative correlation between serum CXCL14 levels and PASI scores in psoriasis

We next measured serum CXCL14 levels in patients with psoriasis and healthy controls. Median serum CXCL14 levels in untreated or topically treated psoriasis patients were 2796.1 [1974.0–4558.0] pg/mL, which was lower than those in healthy individuals (4118.2 [2747.1–4465.2] pg/mL), whereas significant difference was not found (Figure 2A). Interestingly, serum CXCL14 levels were significantly negatively correlated with PASI scores in psoriasis patients (Figure 2B), indicating that a decrease in CXCL14 expression may contribute to the progression of psoriasis.

3.3 | Increased serum CXCL14 levels in nbUVB-treated psoriasis patients

To examine the effect of therapies for psoriasis on CXCL14 expression, we compared serum CXCL14 levels between patients treated with biologics, oral systemic therapies, and nbUVB therapy. We found that serum CXCL14 levels in patients treated with nbUVB therapy were significantly higher than those with biologics or oral systemic therapies (Figure 2C).

3.4 | Upregulated CXCL14 expression in UVB-irradiated HaCaT cells

Based on the results above, we hypothesized that UVB irradiation has the capacity to upregulate CXCL14 expression in the skin. To clarify that, we examined CXCL14 expression on UVB-irradiated HaCaT cells. We found that CXCL14 mRNA expression was increased in UVB-irradiated HaCaT cells at 6 h after UVB irradiation compared to nonirradiated cells (Figure 3). On the other hand, supernatant CXCL14 protein levels were below the detection limit of ELISA Kit, and the increase in protein levels was not confirmed. Thus, UVB irradiation can induce CXCL14 mRNA expression in epidermal keratinocytes.

4 | DISCUSSION

In this report, we first found that CXCL14 expression in epidermal keratinocytes was significantly decreased in psoriasis skin. Similar

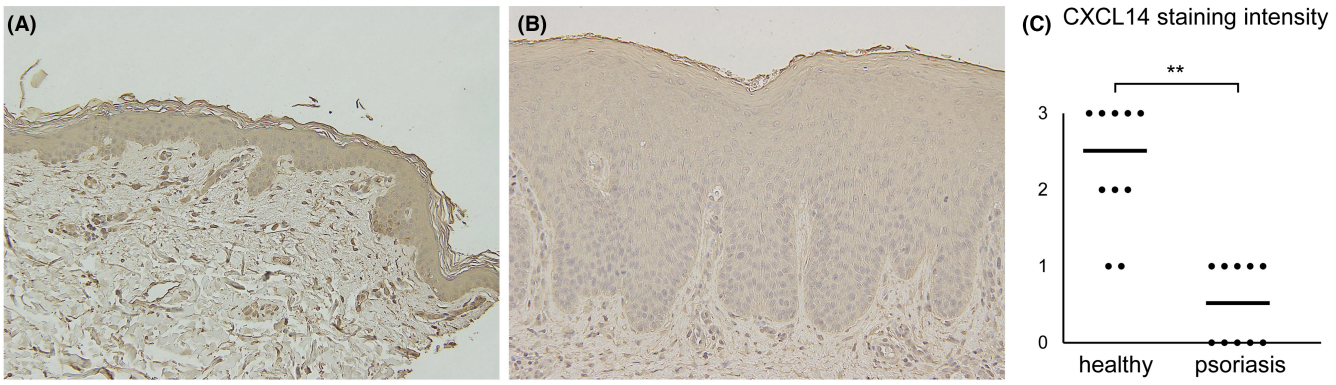

FIGURE 1 (A, B) Immunohistochemical staining for CXCL14 in healthy skin (A) and lesional skin of psoriasis (B; original magnification × 200). The representative images of each 10 cases were shown. (C) CXCL14 staining intensity in epidermal keratinocytes in healthy skin and lesional skin of psoriasis is shown. The measured values from individual patients are plotted by dots. The bar indicates the median. $^{**}p < .01$.

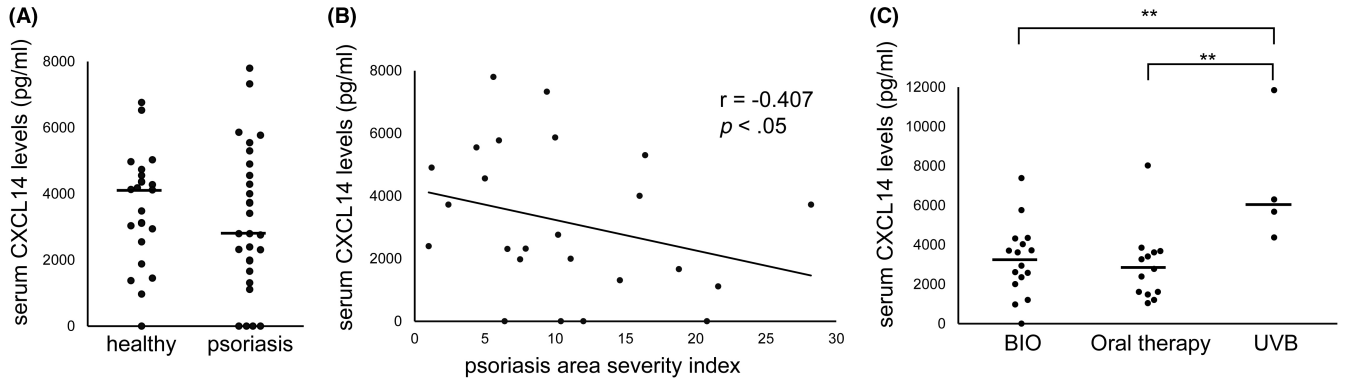

FIGURE 2 (A) Serum CXCL14 levels in patients with psoriasis ($n=29$) and healthy controls ($n=23$). (B) The correlation between serum CXCL14 levels and the Psoriasis Area and Severity Index (PASI) scores ($n=24$). (C) Serum CXCL14 levels in psoriasis patients treated with BIO (biologics; $n=16$), oral systemic therapies ($n=13$), and narrow band ultraviolet B (nbUVB) therapy ($n=4$). The measured values from individual patients are plotted by dots. The bar indicates median. $^{**}p<.01$.

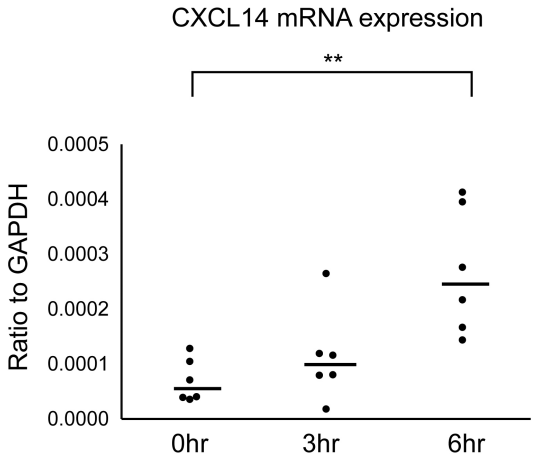

FIGURE 3 HaCaT cells were irradiated with 312 nm ultraviolet B (UVB) light (1.0 J/cm^2) and CXCL14 mRNA expression was measured by quantitative RT-PCR at 3 and 6 h after UVB irradiation. One representative result from three independent experiments with similar results. The measured values from individual samples are plotted by dots ($n=6$). The bar indicates median. $^{**}p<.01$.

to our results, a decrease in CXCL14 expression has been reported in several autoimmune and inflammatory diseases. CXCL14 mRNA expression was significantly decreased in the large colon of the mouse model of ulcerative colitis.[17] In patients with systemic lupus erythematosus, CXCL14 mRNA expression was decreased in peripheral blood mononuclear cells and negatively correlated with albuminuria.[18] Decreased serum CXCL14 levels were observed in systemic sclerosis patients and associated with digital ulcers.[19] Our results together with these previous reports lead to the possibility that a decrease in CXCL14 may be associated with the development of various inflammatory diseases. Actually, we found that serum CXCL14 levels were negatively correlated with PASI scores, the clinical disease severity marker in psoriasis. On the other hand, serum CXCL14 levels tended to decrease in psoriasis patients compared to healthy controls, whereas the difference was not statistically significant in our cohort. Although we did not examine CXCL14 expression in the nonlesional skin of psoriasis patients, abundant CXCL14 produced in nonlesional skin might explain the lack of statistical difference.

As described above, CXCL14 can hamper the function of CXCL12. Expression levels of CXCL12 and its receptor CXCR4 are increased in psoriasis skin and the inhibition of CXCL12/CXCR4 axis by AMD3100, a specific CXCR4 antagonist, reduced the inflammatory cell accumulation in psoriasis mouse models.[20–22] Given that, the decrease in CXCL14 expression in psoriasis skin may be followed by the augmentation of CXCL12/CXCR4 axis, leading to the exacerbation of skin inflammation. The amplification of CXCL14 expression may be a therapeutic option for psoriasis.

We next examined the effect of psoriasis therapies on CXCL14 expression and found that serum CXCL14 levels were increased in patients treated with nbUVB compared to those with biologics or oral systemic therapies. Moreover, UVB irradiation upregulated CXCL14 mRNA expression from HaCaT cells. Although the increase of CXCL14 protein levels in the supernatant of HaCaT cells by UVB irradiation was not confirmed, considering that UVB therapy affects almost all epidermal keratinocytes on the whole body, the accumulation of the trace amounts of CXCL14 induced from each keratinocyte may lead to increased CXCL14 levels in the patients' serum. NbUVB therapy has been widely used for inflammatory skin diseases including psoriasis, and the effect is thought to be because of inducing apoptosis in T cells and cytokine synthesis implicated in immune suppression.[23,24] Our results suggest that CXCL14 upregulation in epidermal keratinocytes in addition to such mechanisms may contribute to the therapeutic effect of nbUVB therapy for psoriasis. In the context of CXCL14 modulation, the combination of nbUVB therapy and other systemic therapies may be reasonable and have an additive effect to systemic therapies alone. Actually, the efficacy of the combination of nbUVB therapy and several systemic therapies has been demonstrated in clinical settings.[25–28]

In summary, CXCL14 expression was decreased in epidermal keratinocytes in lesional skin, and serum CXCL14 levels were negatively correlated with PASI scores in psoriasis patients. Serum CXCL14 levels were increased in nbUVB-treated psoriasis patients, and UVB irradiation induced CXCL14 expression from HaCaT cells. Our results suggest that decreased CXCL14 expression may contribute to the exacerbation of psoriasis and that the amplification of CXCL14 can be a therapeutic option for psoriasis. One of the mechanisms of the efficacy of nbUVB therapy in psoriasis may be the upregulation of CXCL14.

ACKNOWLEDGMENTS

This study was supported by grants from the Ministry of Education, Culture, Sports and Technology in Japan (20K08683 and 23K07797).

CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

ETHICS STATEMENT

Approval of the research protocol: The medical ethical committee of St. Marianna University School of Medicine approved all described studies, and the study was conducted according to the Declaration of Helsinki Principles (approval number: 5666 and 5970).
Informed Consent: For blood samples, all patients were provided written informed consent. For skin samples, informed consent was obtained in the form of opt-out on the web-site.
Registry and the Registration No.: N/A.
Animal Studies: N/A.

ORCID

Tomomitsu Miyagaki https://orcid.org/0000-0003-1879-3128

REFERENCES

1. Lowes MA, Bowcock AM, Krueger JG. Pathogenesis and therapy of psoriasis. Nature. 2007;445:866–73.
2. Nestle FO, Kaplan DH, Barker J. Psoriasis. N Engl J Med. 2009;361:496–509.
3. Boehncke WH, Schön MP. Psoriasis. Lancet. 2015;386:983–94.
4. Honma M, Hayashi K. Psoriasis: recent progress in molecular-targeted therapies. J Dermatol. 2021;48:761–77.
5. Furue K, Ito T, Furue M. Differential efficacy of biologic treatments targeting the TNF-α/IL-23/IL-17 axis in psoriasis and psoriatic arthritis. Cytokine. 2018;111:182–8.
6. Zdanowska N, Kasprowicz-Furmańczyk M, Placek W, Owczarczyk-Saczonek A. The role of chemokines in psoriasis – an overview. Medicina (Kaunas). 2021;57:754.
7. Furue M, Furue K, Tsuji G, Nakahara T. Interleukin-17A and keratinocytes in psoriasis. Int J Mol Sci. 2020;21:1275.
8. Hirota K, Yoshitomi H, Hashimoto M, Maeda S, Teradaira S, Sugimoto N, et al. Preferential recruitment of CCR6-expressing Th17 cells to inflamed joints via CCL20 in rheumatoid arthritis and its animal model. J Exp Med. 2007;204:2803–12.
9. Homey B, Dieu-Nosjean MC, Wiesenborn A, Massacrier C, Pin JJ, Oldham E, et al. Up-regulation of macrophage inflammatory protein-3 alpha/CCL20 and CC chemokine receptor 6 in psoriasis. J Immunol. 2000;164:6621–32.
10. Westrich JA, Vermeer DW, Colbert PL, Spanos WC, Pyeon D. The multifarious roles of the chemokine CXCL14 in cancer progression and immune responses. Mol Carcinog. 2020;59:794–806.
11. Lu J, Chatterjee M, Schmid H, Beck S, Gawaz M. CXCL14 as an emerging immune and inflammatory modulator. J Inflamm (Lond). 2016;13:1.
12. Shellenberger TD, Wang M, Gujrati M, Jayakumar A, Strieter RM, Burdick MD, et al. BRAK/CXCL14 is a potent inhibitor of angiogenesis and a chemotactic factor for immature dendritic cells. Cancer Res. 2004;64:8262–70.
13. Dai C, Basilico P, Cremona TP, Collins P, Moser B, Benarafa C, et al. CXCL14 displays antimicrobial activity against respiratory tract bacteria and contributes to clearance of *Streptococcus pneumoniae* pulmonary infection. J Immunol. 2015;194:5980–9.
14. Hara T, Tanegashima K. CXCL14 antagonizes the CXCL12-CXCR4 signaling axis. Biomol Concepts. 2014;5:167–73.
15. Hromas R, Broxmeyer HE, Kim C, Nakshatri H, Christopherson K 2nd, Azam M, et al. Cloning of BRAK, a novel divergent CXC chemokine preferentially expressed in normal versus malignant cells. Biochem Biophys Res Commun. 1999;255:703–6.
16. Meuter S, Moser B. Constitutive expression of CXCL14 in healthy human and murine epithelial tissues. Cytokine. 2008;44:248–55.
17. Boshagh MA, Foroutan P, Moloudi MR, Fakhari S, Malakouti P, Nikkhoo B, et al. ELR positive CXCL chemokines are highly expressed in an animal model of ulcerative colitis. J Inflamm Res. 2019;12:167–74.
18. Wang JD, Huang QQ, Ye LL, Fan CF, Guo GQ, Xu LJ, et al. Expression and its promoter methylation of chemokine CXC ligand 14 in peripheral blood mononuclear cells from patients with lupus. Zhongguo Ying Yong Sheng Li Xue Za Zhi. 2017;33:197–201.
19. Fukui Y, Miyagawa T, Hirabayashi M, Yamashita T, Saigusa R, Miura S, et al. Possible association of decreased serum CXCL14 levels with digital ulcers in patients with systemic sclerosis. J Dermatol. 2019;46:584–9.
20. Hänsel A, Günther C, Ingwersen J, Starke J, Schmitz M, Bachmann M, et al. Human slan (6-sulfo LacNAc) dendritic cells are inflammatory dermal dendritic cells in psoriasis and drive strong TH17/TH1 T-cell responses. J Allergy Clin Immunol. 2011;127:787–94. e1–9.
21. Quan C, Cho MK, Shao Y, Mianecki LE, Liao E, Perry D, et al. Dermal fibroblast expression of stromal cell-derived factor-1 (SDF-1) promotes epidermal keratinocyte proliferation in normal and diseased skin. Protein Cell. 2015;6:890–903.
22. Zgraggen S, Huggenberger R, Kerl K, Detmar M. An important role of the SDF-1/CXCR4 axis in chronic skin inflammation. PLoS One. 2014;9:e93665.
23. Wong T, Hsu L, Liao W. Phototherapy in psoriasis: a review of mechanisms of action. J Cutan Med Surg. 2013;17:6–12.
24. Bulat V, Situm M, Dediol I, Ljubicić I, Bradić L. The mechanisms of action of phototherapy in the treatment of the most common dermatoses. Coll Antropol. 2011;35(Suppl 2):147–51.
25. Farahnik B, Patel V, Beroukhim K, Zhu TH, Abrouk M, Nakamura M, et al. Combining biologic and phototherapy treatments for psoriasis: safety, efficacy, and patient acceptability. Psoriasis (Auckl). 2016;6:105–11.
26. Bagel J, Nelson E, Keegan BR. Apremilast and narrowband ultraviolet-B combination therapy for treating moderate-to-severe plaque psoriasis. J Drugs Dermatol. 2017;16:957–62.

27. Spuls PI, Rozenblit M, Lebwohl M. Retrospective study of the efficacy of narrowband UVB and acitretin. J Dermatolog Treat. 2003;14(Suppl 2):17–20.
28. Mahajan R, Kaur I, Kanwar AJ. Methotrexate/narrowband UVB phototherapy combination vs. narrowband UVB phototherapy in the treatment of chronic plaque-type psoriasis – a randomized single-blinded placebo-controlled study. J Eur Acad Dermatol Venereol. 2010;24:595–600.

How to cite this article: Nakajima K, Miyagaki T, Tanaka M, Komaki R, Okano T, Takeuchi S, et al. Decreased CXCL14 expression in psoriasis recovered by narrow-band ultraviolet B therapy. J Cutan Immunol Allergy. 2023;6:225–230. https://doi.org/10.1002/cia2.12332