RESEARCH LETTER

Immunologic skin signatures in SARS-CoV-2-negative patients with chilblain-like lesions

To the Editor: Several articles have reported chilblain-like lesions (CBL) as a possible cutaneous sign of SARS-CoV-2 infection, mostly in patients with negative polymerase chain reaction for viral RNA detection.[1] The majority of patients with CBL arising during the COVID-19 pandemic are young, in good health, without risk factors associated with classic chilblains.[1] It has been hypothesized that these subjects may develop an antiviral immune response preventing the onset of significant COVID-19 symptoms.[1,2] Therefore, our aim was to elucidate the immunologic response at skin level in patients with CBL by analyzing the expression profiles of cytokines and SARS-CoV-2 proteins.

From March 2020 to June 2020, patients under 18 years of age with CBLs were consecutively recruited by the dermatology and pediatric outpatient clinics of the University of Naples Federico II, Italy. Informed consent was obtained from their parents. Rhinopharyngeal and oropharyngeal swabs collected from all patients were used for SARS-CoV-2 RNA detection through polymerase chain reaction and a serology test (immunoglobulin M and IgG) for SARS-CoV-2. A 3-mm punch skin biopsy on CBL was performed according to clinical and dermatoscopic disease activity.[2] We performed gene expression analysis of monocyte chemoattractant protein-1 (MCP-1), C-X-C motif chemokine ligand 10 (CXCL10), interferon alfa (IFN-α), and SARS-CoV-2 nucleoprotein and spike protein using polymerase chain reaction. Moreover, SARS-CoV-2 spike and envelope proteins were assessed using immunofluorescence.

Fifteen patients were enrolled (6 girls and 9 boys; mean age, 13 years ± 2.08 SD) in the study. Hospital admission occurred on day 57 ± 43 SD since the lesions' onset. CBL appeared as erythematous-purpuric papules and macules, sometimes with bullous and crusty evolution or digital swelling (Fig 1). Rhinopharyngeal and oropharyngeal swab tests were negative and IgG and immunoglobulin M antibodies to SARS-CoV-2 were absent in all patients, although 3 of the 15 patients (20.0%) had established contacts with COVID-19 subjects, and 6 patients (40.0%) reported a history of mild COVID-19-like symptoms in the previous months. Laboratory tests excluded any autoimmune or infectious diseases. In the skin biopsy of all patients, we observed a strong increase in the gene expression of IFN-α, MCP-1, and CXCL10 compared with that in the skin of healthy subjects, whereas no mRNA expression was found for SARS-CoV-2 nucleoprotein and spike protein (Fig 2). Immunofluorescence was performed in 10 of the 15 patients; no SARS-CoV-2 spike and envelope proteins were detected.

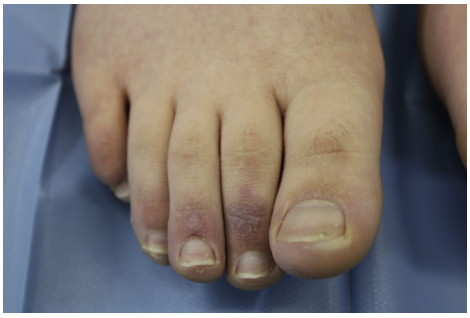

Fig 1. Chilblain-like lesions appearing as erythematous-purpuric papules and macules.

Chemokines, such as MCP-1 and CXCL10, are low-molecular weight cytokines that are able to chemoattract leukocytes and other immune cells at the site of damage or infection.[3] Interestingly, MCP-1, as well as CXCL10, has a well-established role in COVID-19-related cytokine storm and acute respiratory distress syndrome.[3,4] On the other hand, type I IFNs (IFN-α and IFN-β) are key drivers of antiviral immunity. A sustained early response of type I IFN seems to be associated with early SARS-CoV-2 control and mild disease. Conversely, an inadequate response of type I IFN may be accompanied by progression to more severe disease.[5] Our data reveal the development of a high tissue type I IFN response in patients with CBL. Thus, CBL could represent the clinical expression of the exposure to SARS-CoV-2 in patients mounting a robust innate immune type I IFN response, which neutralizes the virus, not requiring the production of antibodies. Moreover, CBLs may be a late manifestation of SARS-CoV-2 infection, thus accounting for the negativity of rhinopharyngeal and oropharyngeal swab tests and the absence of SARS-CoV-2 proteins in the skin.

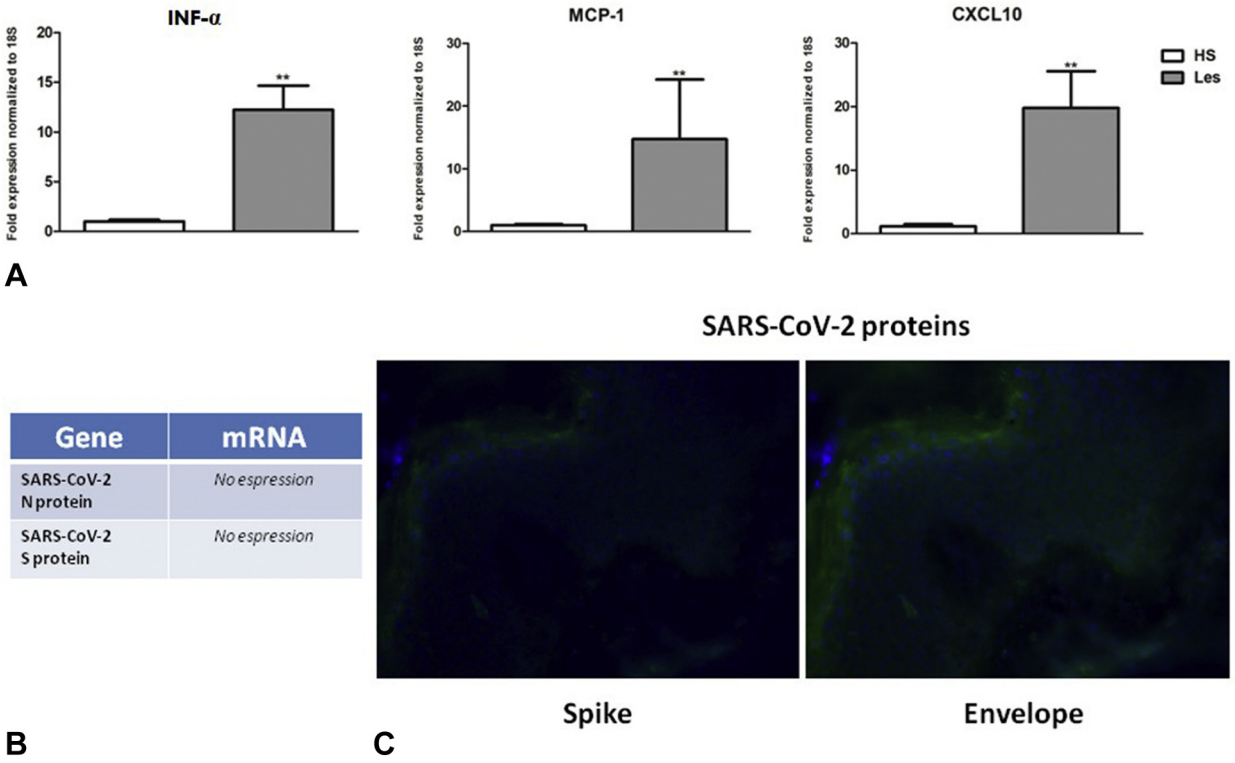

Fig 2. **A**, IFN-α, MCP-1, and CXCL10 gene expressions in the skin of healthy subjects (HS) and lesional (Les) skin of patients with chilblains. Values are normalized to the housekeeping gene 18S and expressed as mean ± SD. Statistical significance was assessed using the Mann-Whitney U test for comparison between Les and HS (**$P < .01$). **B,** Gene expression of SARS-CoV-2 nucleoprotein and spike protein in lesional skin of patients with chilblains. **C,** SARS-CoV-2 envelop and spike protein levels in lesional skin of patients with chilblains. *HS,* Skin of healthy subjects; *Les,* lesional skin of patients with chilblains; *N,* nucleoprotein; *S,* spike protein. (Original magnification: **C,** ×20.)

Maria Vastarella, MD,[a] *Angela Patrì, MD,*[a] *Giuseppina Caiazzo, MD,*[a] *Paola Nappa, MD,*[a] *Francesca Gaudiello, MD,*[a] *Luca Pierri, MD,*[b] *Andrea Catzola, MD,*[b] *Maria Alessio, MD,*[b] *Alfredo Guarino, MD,*[b] *and Gabriella Fabbrocini, MD*[a]

From the Department of Clinical Medicine and Surgery, Section of Dermatology[a] *and Department of Translational Medicine, Section of Pediatrics, University of Naples Federico II, Naples, Italy.*[b]

Drs Guarino and Fabbrocini contributed equally to this article.

Funding sources: None.

IRB approval status: Not applicable.

Correspondence to: Gabriella Fabbrocini, MD, Department of Clinical Medicine and Surgery, Section of Dermatology and Venereology, University of Naples Federico II, Via Pansini 5, Naples, NA 80131, Italy

E-mail: gafabbro@unina.it

Conflicts of interest

None disclosed.

REFERENCES

1. Hubiche T, Le Duff F, Chiaverini C, Giordanengo V, Passeron T. Negative SARS-CoV-2 PCR in patients with chilblain-like lesions. *Lancet Infect Dis*. 2021;21(3):315-316.
2. Fabbrocini G, Vastarella M, Nappa P, et al. A new dermoscopic pattern for chilblain-COVID-19—like skin lesions in adolescents. *JAAD Case Rep*. 2020;6(12):1271-1274.
3. Oliviero A, de Castro F, Coperchini F, Chiovato L, Rotondi M. COVID-19 pulmonary and olfactory dysfunctions: is the chemokine CXCL10 the common denominator? *Neuroscientist*. 2021;27(3):214-221.
4. Chen Y, Wang J, Liu C, et al. IP-10 and MCP-1 as biomarkers associated with disease severity of COVID-19. *Mol Med*. 2020;26(1):97.
5. Damsky W, Peterson D, King B. When interferon tiptoes through COVID-19: Pernio-like lesions and their prognostic implications during SARS-CoV-2 infection. *J Am Acad Dermatol*. 2020;83(3):e269-e270.

https://doi.org/10.1016/j.jdin.2021.05.005