Severe cold urticaria can point to an underlying clonal mast cell disorder

To the Editor,

Gain-of-function mutations of KIT, most frequently D816V, cause clonal mast cell disorders (CMCD) and drive severe anaphylaxis in patients with systemic mastocytosis (SM). The connection between CMCD and chronic inducible urticaria (CIudU) has not been investigated before.

Cold urticaria (ColdU) is characterized by the appearance of wheals and/or angioedema in response to body cooling and rewarming. In cold-induced anaphylaxis (ColdA), there is a sudden cold-induced onset of at least two of the following: (a) involvement of the skin and/or mucosal tissue, (b) respiratory involvement, (c) reduced blood pressure or associated symptoms, or (d) gastrointestinal
symptoms. Clinical presentations of ColdU depend on the potency and duration of exposure to different cold triggers (air, fluids, surfaces), individual cold sensitivity thresholds and other yet to be defined factors. The diagnosis of ColdU relies on the patient’s history and positive cold stimulation testing (CST) with an ice cube and/or TempTest® or other less commonly used methods. Critical temperature threshold (CTT) is the highest TempTest® temperature, and the critical stimulation time threshold (CSTT) is the shortest time required to induce a wheal in CST.1

Here, we describe detailed characteristics of a 65-year-old female patient who presented with a 19-year history of ColdU with increasing severity two years before evaluation and compare them to characteristics of 35 other subsequent ColdU patients comprehensively evaluated at the Urticaria Center of Reference and Excellence (UCARE) at the University Clinic Golnik.2 The following data indicated a severe ColdU (Table 1): (a) wheals induced by air temperatures of 24°C or lower, (b) an unusual 5-minute episode of dyspnea after drinking cold water, and (c) high disease activity with a 30-second CSTT and a 28°C CTT, with ice cube- and TempTest®-induced pseudopodial whealing, and whealing beyond the TempTest®-stimulated skin area (Figure 1A,B). Because these findings pointed to unusually severe ColdU as compared to 35 control ColdU patients (Table 1), we determined the patient’s basal serum tryptase levels, which were elevated twice in a four-month period (17.3, 23.0 ng/mL). An underlying CMCD was suspected and confirmed by a positive KIT D816V mutation test in circulating blood leukocytes (1.0% mutational burden) and in the bone marrow. No cytological features of SM were found in the bone marrow.

The diagnosis of SM is established if: (a) at least a major and one minor or (b) three minor criteria defined by the World Health Organization are fulfilled.3 Our patient did not meet these criteria. Since she presented with symptoms of mast cell activation, the KIT D816V mutation and lacked signs of cutaneous mastocytosis, she was diagnosed with a monoclonal mast cell activation syndrome (MMAS).4 MMAS has not been previously described in patients with ColdU, probably because they are hardly ever assessed for a CMCD.

Our findings suggest that severe reactivity to cold triggers, in patients with ColdU, can point to comorbid MMAS and the possibility that MMAS is a risk factor for severe reactions. Our patient’s reaction to drinking cold water, a single, short-lived episode of dyspnea with spontaneous resolution, may have been ColdA or, more likely, a severe local response. Screening ClndU patients with severe disease for an elevated tryptase and the presence of the KIT D816V mutation could help to clarify the rates and role of activating KIT mutations as a potential driver for severe reactions. It could also improve our approach to the management and treatment of patients with severe KIT D816V-positive ColdU, for example, by the use of selective tyrosine-kinase inhibitors, such as avapritinib or other KIT-targeting therapies, and of omalizumab, which has been shown to benefit patients with ClndU and effectively prevent anaphylaxis in patients with MMAS and SM.5,6

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KEYWORDS
basal serum tryptase, clonal mast cell disorder, cold urticaria, systemic mastocytosis

CONFLICT OF INTEREST
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# TABLE 1 Characteristics of study participants

| Characteristics                                      | Described patient | Comparative group (N = 35) |
|------------------------------------------------------|-------------------|---------------------------|
| **A. Demographics and baseline characteristics**     |                   |                           |
| Age (years)                                          | 65                | Median (range): 40 (18–73)|
| Female gender                                        | Yes               | 66% (23/35)               |
| BMI (kg/m²)                                          | 24.0              | Median (range): 24.9 (18.0–36.0)|
| **B. Patient history data**                          |                   |                           |
| Age at onset of ColdU (years)                        | 46                | Median (range): 33 (9–60)|
| Early onset of ColdU (≤18 years old)                 | No                | 9% (3/35)                 |
| Duration of ColdU before evaluation (months)         | 228f              | Median (range): 60 (2–384)|
| Wheals induced by cold                                | Yes               | 100% (35/35)              |
| Induced by air temperatures of ≤20°C                  | Yes               | 66% (23/35)               |
| Induced by air temperatures of >20°C                  | Yesf              | 11% (4/35)                |
| Induced by contact with solid surfaces                | Yes               | 37% (13/35)               |
| Induced by localized contact with cold liquids        | Yes               | 43% (15/35)               |
| Maximal duration of wheals (min)                      | 45                | Median (range): 60 (5–720)|
| Itch induced by immersion in the sea                  | Yes               | 63% (22/35)               |
| The shortest time to onset of itch (minute/s)         | 1f                | Median (range): 5 (1–20), N=18|
| Angioedema induced by cold                            | Yes               | 34% (12/35)               |
| Maximal duration of angioedema (minutes)              | 45                | Median (range): 60 (20–720), N=11|
| Oropharyngeal or laryngeal symptoms induced by cold   | Yesf              | 31% (11/35)               |
| Gastrointestinal symptoms induced by cold             | No                | 14% (5/35)                |
| Respiratory symptoms induced by cold                  | Yes               | 37% (13/35)               |
| Induced by ingestion of cold drinks                   | Yesf              | 0% (0/35)                 |
| Induced by air temperatures of ≤20°C                  | No                | 9% (3/35)                 |
| Induced by air temperatures of ≤20°C, no asthma       | No                | 3% (1/35)                 |
| Induced by immersion in the sea                       | No                | 11% (4/35)                |
| Symptoms of reduced blood pressure induced by cold    | No                | 29% (10/35)               |
| Wind is an aggravating factor                         | Yes               | 60% (21/35)               |
| Increased humidity is an aggravating factor           | Yes               | 49% (17/35)               |
| Asthma                                               | No                | 34% (12/35)               |
| At least one: asthma, allergic rhinitis, allergic conjunctivitis, or atopic dermatitis | No | 40% (14/35) |
| Thyroid disease                                      | No                | 17% (6/35)                |
| Systemic reaction to a Hymenoptera sting             | No                | 3% (1/35)                 |
| Positive family history of ColdU                     | No                | 6% (2/35)                 |
| **C. Cold stimulation testing (CST) results**         |                   |                           |
| Positive ice cube test                                | Yes               | 66% (23/35)               |
| The shortest CSTT ever determined (seconds)           | 30f               | Median (range): 300 (30–300), N=22|
| Ice cube-induced pseudopodial whealing                | Yesf              | 9% (2/23)                 |
| Positive TempTest®                                    | Yes               | 43% (15/35)               |
| The highest CTT ever determined (°C)                  | 28f               | Median (range): 17 (6–25), N=15|
| TempTest®-induced wheal diameter                      | 10f               | Median (range): 9 (5–15), N=11|
| TempTest®-induced pseudopodial whealing               | Yesf              | 0% (0/11)                 |
| Whealing beyond stimulated skin area                  | Yesf              | 0% (0/11)                 |
| **D. Patient-reported outcome**                       |                   |                           |
| ACUSI score (5–18)                                   | 13f               | Median (range): 10 (6–17) |

(Continues)
TABLE 1 (continued)

| Characteristics                                                                 | Described patient | Comparative group (N = 35) |
|---------------------------------------------------------------------------------|-------------------|-----------------------------|
| E. Diagnosis                                                                    |                   |                             |
| Cold contact urticaria<sup>b</sup>                                               | Yes               | 60% (21/35)                 |
| Ice cube and TempTest<sup>®</sup> positive                                       | Yes               | 43% (15/35)                 |
| Ice cube positive at least once, TempTest<sup>®</sup> never positive             | No                | 17% (6/35)                  |
| Ice cube never positive, TempTest<sup>®</sup> positive at least once             | No                | 0% (0/35)                   |
| Localized ColdU                                                                 | No                | 3% (1/35)                   |
| Localized cold reflex urticaria                                                  | No                | 6% (2/35)                   |
| Systemic atypical ColdU suspected (negative local CST)                           | No                | 31% (11/35)                 |
| ColdA                                                                           | No                | 34% (12/35)                 |
| Comorbid other form of ClndU                                                    | No                | 17% (6/35)                  |
| Comorbid CSU                                                                     | No                | 17% (6/35)                  |
|                                                                                   |                   |                             |
| F. Laboratory findings                                                           |                   |                             |
| Total IgE level (IU/mL)                                                         | 113               | Median (range): 97 (9–2050), N=30 |
| Basal serum tryptase (ng/mL)<sup>c</sup>                                         | 23.0              | Median (range): 5.5 (2.9–15.9), N=33 |
| Elevated basal serum tryptase (>11.4 ng/mL)                                     | Yes               | 15% (5/33)                  |
| Basal serum tryptase >8.0 ng/mL<sup>d</sup>                                      | Yes               | 18% (6/33)                  |
| KIT D816V mutation in circulating blood leukocytes                               | Yes               | 0% (0/3)                    |
| KIT D816V mutation in the bone marrow                                            | Yes               | 0% (0/2)                    |
| Positive cryoglobulin test                                                       | No                | 28% (9/32)                  |
| Positive cold agglutinin test at 4°C                                             | Yes               | 44% (15/34)                 |
| Cold agglutinin titer at 4°C<sup>e</sup>                                         | 2                 | Mean ±SD: 1.4 ± 2.9, N=34   |

Note: Comparative group: A total of 35 cold urticaria patients represent a comparative group. Results are shown as percentage (%) of total for categorical variables and median (range) or mean ±SD for numerical variables.

Abbreviations: ACUSI, Acquired Cold Urticaria Severity Index; BMI, body mass index; ClndU, chronic inducible urticaria; ColdA, cold-induced anaphylaxis; ColdU, cold urticaria; CST, cold stimulation testing; CSTT, critical stimulation time threshold; CSU, chronic spontaneous urticaria; CTT, critical temperature threshold; IgE, immunoglobulin E; N, number of patients; SD, standard deviation.

All patients:

Values exceeding the range of a comparative group are in bold.
<sup>a</sup>CST results from past evaluations were also obtained.
<sup>b</sup>Whealing on cold-stimulated area on the forearm is obligatory for diagnosis.
<sup>c</sup>The highest level of basal serum tryptase ever determined.
<sup>d</sup>Cutoff value for likelihood of hyper-alpha-tryptasemia.
<sup>e</sup>The numbers are a reciprocal expression of cold agglutinin titers (ie, 2 = agglutination at 1:2 dilution).
<sup>f</sup>Described patient: Values indicating a more severe disease.

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GPR109A deficiency promotes IL-33 overproduction and type 2 immune response in food allergy in mice

To the Editor,

Food allergy (FA) is an uncontrolled hyperreactive immune response resulting in loss of tolerance to food allergens. Defects in epithelial barrier frequently precede the development of allergic responses to food allergens. The G-protein-coupled receptor GPR109A (GPR109A) is expressed by epithelium and innate immune cells in the intestine. Earlier, GPR109A has been reported to mediate anti-lipolytic or anti-inflammatory effects as a receptor for butyrate and niacin. Here, we sought to determine the unique role of GPR109A in FA by employing the oral ovalbumin (OVA) sensitization-induced FA model in wild-type (WT) and Gpr109a−/− mice.

Compared with OVA-sensitized WT mice, OVA-sensitized Gpr109a−/− mice exhibited more severe clinical features of FA, as evidenced by reduced body weight and temperature, and increased diarrhea and intestinal permeability (Figure 1A–D). Pathological markers of FA, serum OVA-IgE and mucosal mast cell protease-1 (mMCP-1) levels were also the greatest in OVA-sensitized Gpr109a−/− mice (Figure 1E,F). Intestinal barrier damage is generally manifested as decreased tight junction protein expression. Compared with the WT mice, OVA-sensitized Gpr109a−/− mice expressed reduced occludin, claudin-1, and zona occludens (ZO)-1, but not ZO-2 (Figure 1G). Jejunum histological examination confirmed an exacerbated FA phenotype in OVA-sensitized Gpr109a−/− mice, evidencing by damaged intestinal villi (Figure 1H). No difference in the above FA markers was observed between untreated Gpr109a−/− and WT mice (Figure S1).

These data indicate a role for GPR109A in the modulation of FA development and epithelial integrity.

GPR109A is expressed in both immune cells and epithelial cells in the intestine. We further investigated whether GPR109A deficiency in the hematopoietic or nonhematopoietic cell compartment plays a more important role in FA by bone marrow chimera (BMC) transfer experiments. Four groups of BMC mice, WT → WT/OVA, Gpr109a−/− → Gpr109a−/−/OVA, WT → Gpr109a−/−/OVA, Gpr109a−/− → WT/OVA, were monitored for FA development. Symptoms of FA were most severely observed in the Gpr109a−/− → Gpr109a−/−+OVA group (Figure S2A-E), followed by WT → Gpr109a−/−/OVA group, which lacked GPR109A in nonimmune cells and Gpr109a−/− → WT/OVA group, which lacked GPR109A in immune cells, suggesting that in addition to its role in nonimmune cells, GPR109A expression in immune cells also plays an important role in FA development.

To decipher the mechanism linking GPR109A deficiency-induced epithelial dysfunction and allergic immune cell responses, we analyzed the epithelial-derived alarmin cytokine IL-33 that promotes a type 2 immune response and mediates FA propagation. The RT-qPCR analysis revealed increased IL-33 expression in jejunum of OVA-sensitized Gpr109a−/− mice compared with OVA-sensitized WT mice (Figure 2A). Western blot analyses further showed that OVA-sensitized Gpr109a−/− mice had increased production of full-length IL-33 and the cleaved form with enhanced activity, compared with OVA-sensitized WT group (Figure 2B). A similar increase of IL-33 was observed in ex vivo isolated jejunal epithelia (Figure S3).

BMC experiments suggest that GPR109A deficiency on immune cells plays a role in promoting disease. Thus, we analyzed...