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Antinuclear antibodies are common and linked to poor response to omalizumab treatment in patients with CSU

To the Editor,

Chronic spontaneous urticaria (CSU) is a frequent disorder that is strongly associated with autoimmunity.[1] This is supported by high rates of autoimmune comorbidities, the age of onset and the female preponderance.[1,2] Also, Patients with CSU have been shown to exhibit functional and relevant mast cell-activating IgG and IgE autoantibodies.[1] These autoantibodies are held to be responsible for the development of the urticarial signs and symptoms of most patients with CSU.[3] Autoimmune CSU (aiCSU) is linked to more severe disease and poor treatment responses to omalizumab.[4,5] Clinical markers of aiCSU and predictors of treatment responses are needed.

Patients with CSU commonly have antithyroid autoantibodies, and antinuclear antibodies (ANAs) have also been reported to be elevated in CSU.[5] As of now, however, it is unclear how common and relevant increased levels of ANAs are in CSU.

Here, we analyzed a large population of 447 patients with CSU treated at the Urticaria Center of Reference and Excellence (UCARE)[6] of Kayseri City Hospital for the prevalence of increased ANA levels. Also, we compared ANA-positive (ANA^+) and ANA-negative (ANA^-) patients for differences in their disease characteristics and response to treatment.

All patients had wheals with or without angioedema; patients with angioedema and without wheals were excluded from the study. This study was approved by the Institutional Review Board and the local ethics committee. Serum samples were analyzed for ANAs by traditional fluorescence microscopy using HEp-2 slides (EUROIMMUN, AG). The cutoff for ANA positivity was set at an antibody titer of 1/100, as per our center's laboratory standard.[7] We also assessed demographic and clinical features, erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), total IgE, vitamin B12, and anti-TPO (Table 1). Overall disease activity was assessed by a physician at baseline and week 12 of omalizumab treatment (300 mg/4 weeks) in 171 patients, by a 10-cm visual analog scale (VAS). Patients who had a more than 80% improvement and a 20%-80% improvement at week 12 were considered complete and partial responders, respectively (nonresponders = VAS improvement < 20% at week 12). More information on the rationale and methods of this study is in the (File S1).

Of 447 patients with CSU, 107 (23.9%) were ANA^+, and 37% of them showed titers of 1/320 or higher. The rate of ANA positivity was higher in female patients ($P < .001$) (Table 1). ANA^+ patients with CSU exhibited angioedema more often than ANA^- patients (70% vs 53%; $P = .002$) (Table 1). Also, rheumatoid arthritis (10.3% vs 4.4%; $P = .025$) and Sjögren disease (4.7% vs 0.6%; $P = .003$) were seen more often in ANA^+ patients (Table 1). In contrast, ANA^+ and ANA^- patients with CSU did not differ in their age, age at onset of CSU, or their disease activity, control, or duration, or any of the other demographic or clinical features assessed (Table 1).

ANA^+ patients with CSU had markedly lower total IgE serum levels than ANA^- patients, 110 vs 64 IU/mL on average ($P < .001$). Also, more ANA^+ patients had low IgE levels, that is, total IgE < 40 IU/mL (38% vs 23%, $P = .003$), and more ANA^- patients had elevated IgE levels, that is, >100 IU/mL (53% vs 39%, $P = .014$). There was a statistically significant, albeit weak negative correlation between ANA titers and IgE levels ($P < .001$, $r = -.170$). ANA^+ patients had higher serum levels of B12 ($P = .003$, Table 1), and there was a statistically significant weak positive correlation between ANA titers and B12 levels ($P = .002$, $r = .175$). ANA^+ patients also were positive for anti-TPO more often than ANA^- patients (27% vs 18%, $P = .049$, Table 1). In contrast, ANA^+ and ANA^- patients with CSU showed similar blood levels of markers for inflammation (CRP, ESR).

Of 171 patients treated, 28 (16%) were resistant to omalizumab. More than half of these nonresponders (54%) were ANA^+, and ANA^+ patients had markedly higher rates of nonresponse (45% vs 9%, in ANA^- patients, $P < .001$) (Table 2). After 12 weeks of omalizumab treatment, ANA^- patients showed a mean reduction in disease activity of 95% as assessed by VAS, compared with 33% in ANA^+ patients. Also, ANA titers and treatment responses to omalizumab showed a significant negative correlation ($P < .001$, $r = -.345$). When we used 1:320 as the cutoff for ANA positivity, 1 of 2 ANA^+ patients were nonresponders, significantly more than in the ANA^- group ($P = .01$). In contrast, ANA^+ and ANA^- patients with CSU showed similar rates of resistance to antihistamine treatment (ie, VAS scores ≥ 7), and antihistamine resistant and nonresistant patients were similar in their rates of ANA positivity (Table 2).

In addition to confirming the high prevalence of ANA in CSU, we show that these autoantibodies may be relevant for patients with CSU and importantly linked to their disease. ANA^+ patients with CSU are more likely to have angioedema and comorbid rheumatological autoimmune diseases. A previous study showed that rheumatoid arthritis and Sjögren disease are more common in patients with CSU.[8] Our study suggests that this is indeed the case and that patients with CSU can be screened for both of these conditions by assessing ANAs.

TABLE 1 Clinical and laboratory characteristics of CSU patients who do or do not have positive ANA titers

	Patients with CSU		
	ANA+	ANA−	*P*
Age in years[a], mean (±SD)	39.48 (±16.58)	38.6 (±14.3)	.607[k]
Age at onset of disease in years[a], mean (±SD)	33.9 (±16.4)	34.2 (±14.5)	.877[k]
Duration of disease in months[a], median (IQR)	24 (8-96)	24 (9-60)	.860[m]
Female[a], n (%)	92 (86%)	223 (66%)	**<.001**[l]
Angioedema (in addition to wheals)[a], n (%)	75 (70%)	181 (53%)	**.002**[l]
Atopy (positive history)[b], n (%)	46 (43%)	147 (43%)	.977[l]
Smoking (positive history)[c], n (%)	26 (25%)	101 (31%)	.187[l]
Asthma (comorbidity)[b], n (%)	21 (20%)	58 (17%)	.552[l]
Hypertension (comorbidity)[a], n (%)	10 (9%)	37 (11%)	.651[l]
Diabetes mellitus (comorbidity)[a], n (%)	8 (8%)	46 (14%)	.094[l]
Thyroid disease (comorbidity) [a], n (%)	26 (24%)	60 (18%)	.128[l]
Psychological disorder (positive history)[d], n (%)	37 (35%)	99 (29%)	.301[l]
History of drug allergy (positive history)[b], n (%)	43 (40%)	104 (31%)	.068[l]
History of food allergy (positive history)[e], n (%)	34 (33%)	102 (33%)	1.000[l]
Infection as a trigger of CSU[e], n (%)	5 (5%)	35 (11%)	.055[l]
Positive family history of CU[f], n (%)	23 (22%)	65 (20%)	.717[l]
Nocturnal pruritus with disturbed sleep[f], n (%)	63 (59%)	185 (57%)	.673[l]
Rheumatoid arthritis[a], n (%)	11 (10.3%)	15 (4.4%)	**.025**[l]
Sjögren[a], n (%)	5 (4.7%)	2 (0.6%)	**.003**[l]
Total IgE in (IU/mL), median (IQR)[g]	64 (21-160)	110 (44-236)	**.001**[k]
ESR (mm/h), median (IQR)[h]	11 (5-18)	8 (4-16)	.128[k]
CRP (mg/L), median (IQR)[i]	3.3 (3.0-7.5)	3.3 (3.0-6.8)	.765[k]
B12 level (pg/mL), median (IQR)[j]	246 (183-338)	199 (155-294)	**.003**[k]
IgG-anti-TPO (IU/mL) (+)[a]	27 (27%)	58 (18%)	**.049**[l]

Abbreviations: ANA, antinuclear antibody; CSU, ESR erythrocyte sedimentation, chronic spontaneous urticaria; IQR, interquartile range (lower quartile-upper quartile); SD, standard deviation.
Bold numbers show "statistically significant results: $p<.05$"
Number of patients for whom information was available:
[a]n = 447.
[b]n = 446.
[c]n = 429.
[d]n = 445.
[e]n = 416.
[f]n = 430.
[g]n = 427.
[h]n = 406.
[i]n = 401.
[j]n = 308.
[k]*t* Test for parametric variables.
[l]Chi-square test.
[m]Mann-Whitney *U* test.

Our finding that ANA$^+$ patients have low IgE fits the notion that CSU is driven by IgG autoantibodies in some patients, that is, patients with aiCSU.[1] ANA, together with anti-TPO, which we found to be linked to ANA, may help to identify patients with aiCSU.[5] Our study also shows that being ANA$^+$ and being ANA$^-$ does make a difference when it comes to the effects of omalizumab on CSU. This finding is backed by several recent reports that CSU patients with autoimmune features show less favorable responses to omalizumab.[9]

This is a monocentric study, and our patients had severe and difficult to treat CSU. One in three patients was refractory to antihistamine treatment. This may be the reason of the higher rate of ANA positivity as compared to previous studies. For additional considerations on the interpretation and relevance of our findings, please refer to the (File S1).

In conclusion, patients with CSU often have ANAs, and this is linked to differences in clinical features, laboratory markers, and treatment responses to omalizumab. Our findings support further

TABLE 2 In CSU, ANAs are linked to poor therapeutic responses in patients treated with omalizumab, but not antihistamines

	Treatment response to omalizumab*			
	CR (n = 110)	PR (n = 33)	NR (n = 28)	*P*
ANA (+) patients, n = 33	12 (36%)	6 (18%)	15 (45%)	**<.001**[a]
ANA (−) patients, n = 138	98 (71%)	27 (20%)	13 (9%)	
	Treatment response to antihistamine therapy**			
	Responder (n = 278)	Nonresponder (n = 173)	*P*	
ANA (+) patients, n = 107	72 (67.3%)	35 (32.7%)	.145[a]	
ANA (−) patients, n = 340	202 (59.4%)	138 (40.6%)		

Abbreviations: CR, complete responder; CSU, chronic spontaneous urticaria; NR, nonresponder; PR, partial responder.
[a]Chi-square test.
*Response to omalizumab treatment was assessed by measuring disease activity at baseline and week 12 of omalizumab treatment by visual analog scale (VAS). Patients who had a more than 80% improvement and a 20%-80% improvement at week 12 were considered complete and partial responders, respectively (nonresponders = VAS improvement < 20% at week12).
**Resistance to antihistamine was assessed by use of the VAS post-treatment with VAS scores ≥ 7 taken to reflect resistance to treatment.

studies of ANA as a biomarker and to assess these autoantibodies for their relevance in CSU.

CONFLICTS OF INTEREST

None.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.