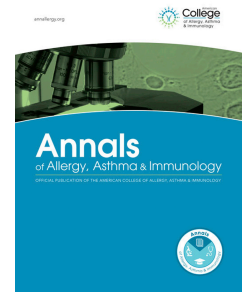


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Keywords: YKL-40, chronic spontaneous urticarial (CSU), HDMECs, permeability, VE-cadherin,

Abbreviations:

CSU: chronic spontaneous urticarial

HMC-1: human mast cell line

HDMECs: human dermal microvascular endothelial cells

VE-cadherin: vascular endothelial cadherin

VEGF: vascular endothelial growth factor

PMA: phorbol 12- myristate 13-acetate

ELISA: enzyme-linked immunosorbent assay

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Chronic spontaneous urticaria (CSU), a common vascular skin disease with high incidence (approximately 0.5% to 1%) and recurrent attacks, is one of the skin diseases that seriously affect the quality of patients' life¹. Some CSU patients have resistance to treatment. About 45%-60% of patients could be effectively treated by antihistamines. Studies demonstrate that histamine and other vasoactive substances, released by mast cell degranulation, can ultimately lead to increase the permeability of human dermal microvascular endothelial cells (HDMECs)², which is considered to be the central link in the pathogenesis of CSU. Bossi et al had reported that the supernatant of human mast cell line (HMC-1), which treated with serum from CSU patients, could increase the permeability of HDMECs². The stability of vascular permeability depends on the normal function of vascular endothelial barrier. As an important intercellular adhesion molecule, vascular endothelial cadherin (VE-cadherin) plays an important role in the changes of vascular permeability

17 induced by various vasoactive substances (including histamine)³. The function of
18 endothelial barrier depends on the correct location and function of VE-cadherin.
19 VE-cadherin at the endothelial junction stabilizes the function of endothelial barrier
20 under the action of integrin $\beta 1$ of endothelial cells⁴. YKL-40, also described with
21 human cartilage glycoprotein-39 and chitinase-3-like-1, can be secreted and expressed
22 by various inflammatory-related cells. YKL-40 plays an important role in cell
23 proliferation, microangiogenesis and acute or chronic inflammation. Clinical studies
24 show that the serum level of YKL-40 is increased in many allergic diseases, such as
25 atopic dermatitis⁵ and asthma⁶, and is positively related to the severity of these
26 diseases. YKL-40 can be secreted by a series of inflammatory-related cells, including
27 tumor cells, vascular smooth muscle cells, mature neutrophils, articular synovial cells.
28 Recent studies have found that YKL-40 can up-regulate the expression of vascular
29 endothelial growth factor (VEGF) in vascular endothelial cells by inducing the
30 coordination of membrane-bound receptor syndecan-1 and integrin $\alpha v \beta 3$, thus
31 promoting the formation of microvessels in vascular endothelial cells⁷. Therefore,
32 YKL-40 may be currently considered to be an important marker of endothelial

dysfunction. Although YKL-40 is highly expressed in many diseases, the expression and effects of YKL-40 in CSU has not yet been elucidated.

To explore the role of YKL-40 in the pathogenic mechanism of CSU, this study investigated the serum levels of YKL-40 in 30 patients with CSU and 30 age and sex matched healthy controls. All patients and healthy subjects were received no any medicines two weeks before this study. We detected the serum levels of YKL-40 in CSU patients and healthy controls. And then, 20 patients with CSU were treated with oral H1 antihistamine (cetirizine or fexofenadine) at the licensed dose for 2 weeks. And then, we also detected the serum levels of YKL-40 in 20 CSU patients which before and after treatment. Serum YKL-40 levels in all groups were measured by Human Chitinase 3-like 1/YKL-40 ELISA Kit (Boster Biological Technology, Wuhan, China. Catalog #EK0974). As shown in Figure 1, patients with CSU have significantly higher YKL-40 serum levels than healthy controls ($p=0.0121$). Moreover, 20 CSU patients had increased serum YKL-40 levels when compared with after treatment ($P=0.0064$). All the results are indicated that YKL-40 may play an important role in the pathogenesis of CSU. Mast cells, as one of the major effector

49 cells in CSU, play an integral role in the inflammatory response by accumulating at
50 sites of inflammation and mediating the production of inflammatory cytokines⁸. To
51 explore the origin of YKL-40 in pathogenesis of CSU, we also assayed mRNA
52 expression of YKL-40 in HMC-1 stimulated by phorbol 12- myristate 13-acetate
53 (PMA) plus 1 μ M of A23187 (calcium ionophore). We found the mRNA levels of
54 YKL-40 in HMC-1 stimulated by PMA and A23187 were markedly increased when
55 compared with PBS controls (data not shown). Abundant evidence had indicated that
56 increased vascular permeability play an important role in the occurrence and
57 pathogenesis of CSU. Recent studies have found that YKL-40 can up-regulate the
58 expression of vascular endothelial growth factor (VEGF) in vascular endothelial cells
59 by inducing the coordination of membrane-bound receptor syndecan-1 and integrin
60 α v β 3, thus promoting the formation of microvessels in vascular endothelial cells⁷. It is
61 suggested that YKL-40-related receptors is expressed in vascular endothelial cells.
62 VE-cadherin is an important intercellular adhesion molecule, which plays an
63 important role in maintaining and restoring the function of endothelial barrier. In vitro,

64 we also found YKL-40 significantly promoted the permeability changes and
65 VE-cadherin release in HDMECs induced by histamine (data not shown).

66 Taken together, this study provides first observations on the association of YKL-40
67 and CSU, and showed the increased YKL-40 serum levels in CSU patients and
68 decreased after treatment. Therefore, we suggest that YKL-40 may play a role in the
69 pathogenesis of CSU. This study may help to further elucidate the pathogenesis of
70 CSU and provide a new target for the development of anti-histamine resistance drugs
71 for CSU.

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1 **Figure legends**

2 **Figure 1. Increased serum YKL-40 levels in CSU patients.**

3 Serum levels of YKL-40 in patients with CSU and healthy controls were determined
4 by ELISA. Serum YKL-40 levels in patients with CSU in acute stage were
5 significantly higher than those in healthy control subjects. P values are based on the
6 Mann-Whitney test. Wilcoxon Signed Ranks Test for paired data showed that serum
7 YKL-40 levels of 20 patients with CSU in convalescent stage were significantly
8 lower than those in acute stage.

