Clinical Allergology – Short Communication

International Archives of Allergy and Immunology

Int Arch Allergy Immunol
DOI: 10.1159/000488142

Received: May 23, 2017
Accepted after revision: March 1, 2018
Published online: April 26, 2018

Diamine Oxidase Supplementation in Chronic Spontaneous Urticaria: A Randomized, Double-Blind Placebo-Controlled Study

Mona-Rita Yacoub[a, b] Giuseppe A. Ramirez[a, b] Alvise Berti[a, b] Giuseppe Mercurio[a, b] Daniela Breda[c] Nicoletta Saporiti[a] Samuele Burastero[c] Lorenzo Dagna[a, b] Giselda Colombo[a, b]

[a]Unit of Immunology, Rheumatology, Allergy and Rare Diseases, IRCCS Ospedale San Raffaele, Milan, Italy; [b]Università Vita-Salute San Raffaele, Milan, Italy; [c]Unit of Cellular and Molecular Allergology, IRCCS Ospedale San Raffaele, Milan, Italy

Keywords
Diamine oxidase · Chronic spontaneous urticaria · Histamine · Histamine intolerance

Abstract
Introduction: Diamine oxidase (DAO) catabolizes and inactivates histamine, a key player in a wide range of invalidating conditions, such as migraine and chronic spontaneous urticaria (CSU). The highest expression of DAO occurs in the gastrointestinal tract, possibly to control the burden of histamine intake from food. ***Methods:*** Here, we tested the hypothesis that a 30-day oral supplementation with DAO (1 capsule b.i.d., 15 min before a meal) could reduce the severity of CSU as estimated by the 7-Day Urticaria Activity Score (UAS-7). The study was designed as a double-blind, placebo-controlled, crossover investigation of 22 patients with CSU incompletely controlled by first-line antihistamine therapy. ***Results:*** Twenty patients completed the study. Supplemental therapy with DAO caused a 3.8 ± 1.2 point mean ± SEM UAS-7 score reduction in patients with low serum DAO levels at time 0 (p = 0.041 compared to placebo). The degree of UAS-7 improvement was inversely correlated with the levels of basal DAO (p = 0.019). Patients receiving DAO supplementation were able to slightly reduce their daily antihistamine dose (p = 0.049). ***Conclusion:*** These data suggest that DAO may be involved in the pathogenic cascade of CSU and that DAO supplementation could be effective for symptom relief in patients with low DAO levels in serum.

Introduction

Chronic spontaneous urticaria (CSU) is a heterogeneous mast-cell-related disease characterized by recurrent flares of wheals and/or angioedema for more than 6 weeks, usually in the absence of clear offending triggers. CSU is highly prevalent in the general population and severely affects patients' quality of life [1]. Histamine lies downstream the pathogenic cascade that leads to the development of itch and wheals in CSU as well as in most

M.-R.Y. and G.A.R. contributed equally to this work.
Edited by: T. Schwarz, Kiel.

KARGER

E-Mail karger@karger.com
www.karger.com/iaa

Correspondence to: Dr. Mona-Rita Yacoub
Unit of Immunology, Rheumatology, Allergy and Rare Diseases
IRCCS Ospedale San Raffaele
Via Olgettina 60, IT–20132 Milan (Italy)
E-Mail yacoub.monarita@hsr.it

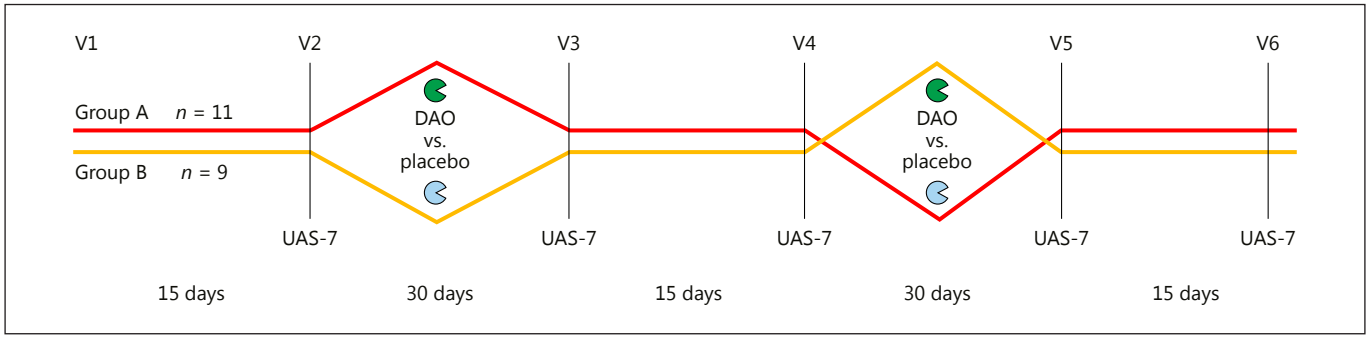

Fig. 1. Flowchart showing the study design and the duration of each study phase. Of the 22 patients recruited, 20 completed the study and were considered for the analysis. Eleven of them had been assigned to group A, 9 to group B. DAO, diamine oxidase; UAS-7, 7-Day Urticaria Activity Score; V1–6, study visits 1–6.

immediate-type hypersensitivity reactions, since it prompts vasodilation and increased vascular permeability [2]. Furthermore, histamine-related events have been implicated in the pathogenesis of other diseases such as atopic dermatitis and migraine [3]. Systemic and local levels of histamine are influenced by mast cell release, gastrointestinal absorption and catabolic efficiency. In particular, diamine oxidase (DAO) catabolizes histamine, thus neutralizing its biological effects [4, 5]. In mammals, DAO is mainly expressed at the level of the intestinal villi, in accordance with a peak enzymatic efficiency at pH 6.3 [6, 7]. Circulating levels of DAO may reflect DAO expression in the gastrointestinal tract [8]. Imbalances in the control of histamine levels due to inefficient DAO-related histamine clearance may lead to histamine accumulation and to the development of undesired histamine-related manifestations such as urticaria, accelerated gastrointestinal transit, or headache [2]. DAO supplementation has the potential to reverse these clinical manifestations [9], but data regarding its efficacy in CSU are lacking. We thus tested the hypothesis that circulating levels of DAO may reflect disease activity and that DAO supplementation may prompt symptom relief in patients with CSU.

Methods

We performed a double-blind, placebo-controlled, crossover study to assess the efficacy of a DAO supplement (Daosin®, AET-Pharma, Italy) in CSU. The study was approved by our local ethics committee. We enrolled 22 consecutive patients with CSU, lasting from an average of 6.8 years (0.25–17 years), unresponsive to a single daily dose of antihistamine and a 21-day histamine-free diet [1], diagnosed according to WAO guidelines at the San Raffaele Research Hospital, Milan, Italy [1]. Patients with concomitant gastrointestinal disorders, systemic autoimmune diseases, symptomatic IgE-mediated hypersensitivity to food allergens, previous severe anaphylaxis and/or elevated basal tryptase levels, other skin diseases, neoplasia, severe cardiopulmonary diseases or psychiatric illnesses, pregnant or lactating patients as well as patients with symptoms suggesting histamine intolerance other than urticaria were excluded. All enrolled patients were randomly allocated 1:1 to either group A or group B. The study lasted 105 days including (1) a first-entry, observational phase of 15 days; (2) a 30-day treatment phase in which group A received Daosin® 1 capsule 15 min before lunch and dinner, while group B received placebo; (3) a 15-day washout phase; (4) a 30-day treatment phase in which group A received placebo and group B the active treatment; (5) a final 15-day washout phase (Fig. 1). There were no specific dietary or drug restrictions during the study. Antihistamines in particular were administered, titrated, or tapered as per usual clinical practice. Serum levels of DAO were assessed at time 0 with a radioextraction assay using a commercial kit (DAOREA, AET Pharma, Italy) according to the manufacturer's instructions. The investigators were blinded with respect to basal DAO levels until the study was over. CSU severity as estimated by the 7-Day Urticaria Activity Score (UAS-7) and frequency of antihistamine use were measured on days 15, 45, 60, 90, and 105. Of 22 enrolled patients, 20 (11 in group A, 9 in group B) completed the study. Dropout was due to consent retreat in 1 case and change of residence in the other. After checking for normal distribution by the Kolmogorov-Smirnov test, UAS-7 variations among different study phases were compared between groups by employing the Student *t* test. Correlations between nonnormally distributed variables were analyzed by the Spearman test or Kruskall-Wallis test.

Results

Table 1 reports a summary of the patients' demographics. The mean ± standard error of mean (SEM) levels of tryptase and total serum IgE were 4.97 ± 0.66 μg/L (n = 14) and 170.01 ± 34.05 (n = 16), respectively. Five out of 20 patients had low (<10 U/mL) DAO levels at baseline

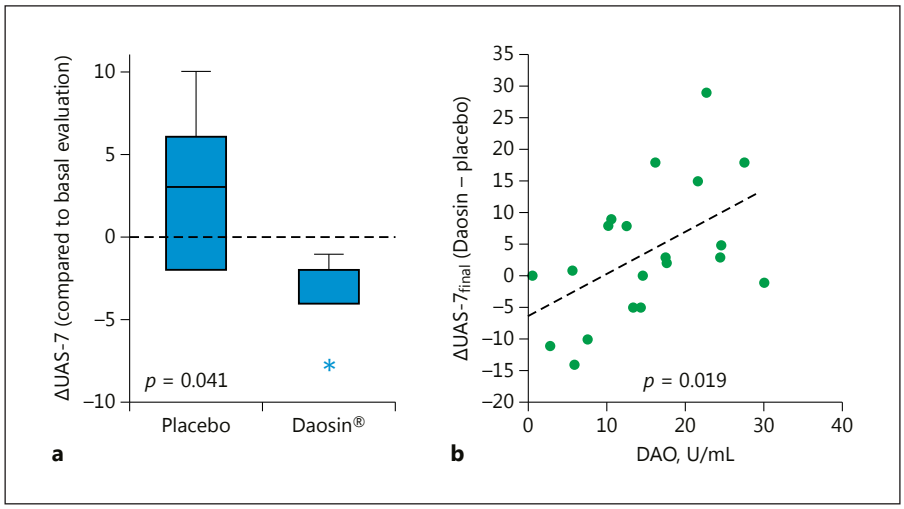

Fig. 2. a Reduction of 7-Day Urticaria Activity Score (UAS-7) with diamine oxidase (DAO) supplementation (Daosin®) and placebo in comparison to the basal UAS-7 in patients with low basal DAO. All patients with low basal DAO responded to the treatment. **b** Correlation between DAO levels in serum at time 0 and the net reduction of UAS-7 after treatment ($\Delta UAS\text{-}7_{final} = \Delta UAS\text{-}7_{Daosin} - \Delta UAS\text{-}7_{placebo}$).

Table 1. General demographics

Patient No.	Group	Gender	Age, years	Disease duration, years	Antithyroid immunity	Other autoantibodies
1	A	F	28	8	negative	no
2	A	F	24	1	negative	no
3	A	M	35	9	positive	no
4	A	F	19	0.25	negative	no
5	A	F	64	4	negative	no
6	A	F	59	0.5	positive	no
7	A	F	23	14	positive	no
8	A	F	41	6	positive	no
9	A	F	49	0.5	no	ANA
10	A	F	64	0.75	no	no
11	A	F	34	2.5	positive	no
12	B	F	59	6	no	no
13	B	M	60	0.16	no	no
14	B	F	48	1	no	no
15	B	F	70	17	no	no
16	B	M	55	0.33	no	ANA
17	B	M	35	0.5	no	no
18	B	F	45	0.5	no	no
19	B	F	59	0.5	positive	no
20	B	M	22	2	no	cold agglutinins

ANA, antinuclear antibodies.

[9]. When all patients' data were considered as a whole, there was no significant correlation between DAO basal levels and UAS-7 values at enrollment. The UAS-7 did not change significantly in either group during the treatment period, and no significant differences were detected in terms of disease severity between the placebo and the active group in both treatment phases. However, all patients with low basal DAO experienced a significant reduction of the UAS-7 score with Daosin®, when compared to placebo (mean ± SEM $\Delta UAS\text{-}7_{Daosin} = -3.8 \pm 1.2$ points vs. $\Delta UAS\text{-}7_{placebo} = 3.0 \pm 2.3$ points; $p = 0.041$; Fig. 2a). Basal levels of DAO correlated with the net effect of Daosin®, after adjusting for the placebo effect ($\Delta UAS\text{-}7_{final} = \Delta UAS\text{-}7_{Daosin} - \Delta UAS\text{-}7_{placebo}$; $p = 0.019$; Fig. 2b). The number of patients on antihistamines did not change significantly during DAO supplementation, when com-

Table 2. Antihistamine use throughout the study

	Baseline	DAO	Placebo	*p*
Patients regularly taking antihistamines, *n*	19/20	15/20	14/20	ns
Mean ± SEM daily dose of antihistamines, number of tablets	1.04±0.11	0.69±0.11	0.81±0.13	0.049

pared to the baseline evaluation or the phase in which patients took placebo. However, we observed a slight but significant ($p = 0.049$) reduction in the average daily dose of antihistamines when patients received DAO supplementation (Table 2). No other clinical features predicted a clinical response to DAO supplementation.

Discussion

In this small signal-seeking study, we aimed at finding clinical clues supporting a role of DAO as a disease modifier in CSU. Previous works suggest that imbalances in histamine degradation capacity may associate with the development of migraine and atopic dermatitis [3, 4], but little is known on the role of DAO in CSU. Taken together, our data suggest that DAO activity could be a cofactor in the development of CSU clinical manifestations and that DAO supplementation could contribute to a better disease control. Patients with low basal levels of DAO in serum may experience the most significant benefits. We acknowledge that the small sample size of our cohort as well as the lack of data regarding histamine levels and circulating levels of DAO after supplementation constitute potential limits of our study. In addition, by excluding patients with other potential symptoms of histamine intolerance in this trial setting, we might have sacrificed a wider understanding of the interactions between DAO and histamine in the real world to a clearer evidence about CSU. Larger studies are thus required to confirm our observations and possibly to extend the spectrum of DAO supplementation to other histamine-related manifestations.

Statement of Ethics

The authors declare that all patients involved in this study gave their written informed consent before being enrolled in the investigation. The study was approved by the ethics committee of the IRCCS Ospedale San Raffaele, Milan, Italy.

Disclosure Statement

The authors declare that there is no conflict of interest in connection with this work.

Funding Sources

The study was supported by AETPharma S.r.l., Inveruno (MI), Italy, which also provided the placebo/investigational product kits as well as the reagents for DAO measurement.

References

1 Zuberbier T, Aberer W, Asero R, Bindslev-Jensen C, Brzoza Z, Canonica GW, et al: The EAACI/GA(2) LEN/EDF/WAO guideline for the definition, classification, diagnosis, and management of urticaria: the 2013 revision and update. Allergy 2014;69:868–887.
2 Wernersson S, Pejler G: Mast cell secretory granules: armed for battle. Nat Rev Immunol 2014;14:478–494.
3 Garcia-Martin E, Martinez C, Serrador M, Alonso-Navarro H, Ayuso P, Navacerrada F, Agundez JA, Jimenez-Jimenez FJ: Diamine oxidase rs10156191 and rs2052129 variants are associated with the risk for migraine. Headache 2015;55:276–286.
4 Maintz L, Benfadal S, Allam JP, Hagemann T, Fimmers R, Novak N: Evidence for a reduced histamine degradation capacity in a subgroup of patients with atopic eczema. J Allergy Clin Immunol 2006;117:1106–1112.
5 Shamji MH, Layhadi JA, Scadding GW, Cheung DK, Calderon MA, Turka LA, Phippard D, Durham SR: Basophil expression of diamine oxidase: a novel biomarker of allergen immunotherapy response. J Allergy Clin Immunol 2015;135:913–921. e919.
6 Naila A, Flint S, Fletcher GC, Bremer PJ, Meerdink G, Morton RH: Prediction of the amount and rate of histamine degradation by diamine oxidase (DAO). Food Chem 2012; 135:2650–2660.
7 Stein J, Scheuermann EH, Yazdi R, Lembcke B, Caspary WF: Reduced postheparin plasma diamine oxidase activity in patients with chronic renal failure. Z Gastroenterol 1994; 32:236–239.
8 Maintz L, Yu CF, Rodriguez E, Baurecht H, Bieber T, Illig T, Weidinger S, Novak N: Association of single nucleotide polymorphisms in the diamine oxidase gene with diamine oxidase serum activities. Allergy 2011;66:893–902.
9 Manzotti G, Breda D, Di Gioacchino M, Burastero SE: Serum diamine oxidase activity in patients with histamine intolerance. Int J Immunopathol Pharmacol 2016;29:105–111.