Trappin-2/Elafin and Clusterin serum levels in pemphigus vulgaris and correlation with the severity score: a case–control study

Badran Y. Aya[a], Shehata Refaat Rofaida[a], Abd-Elkader S. Alaa[b], Kamel A. Amira[c], Abd-Elsamea S. Fatma[d], Gomaa S. Ahmed[a]

[a]Department of Dermatology, Venereology and Andrology, Faculty of Medicine, Assiut University, Assiut, Egypt, [b]Department of Clinical Pathology, Assiut University, Assiut, Egypt, [c]Department of Medical Biochemistry, Assiut University, Assiut, Egypt, [d]Department of Medical Microbiology and Immunology, Assiut University, Assiut, Egypt

Correspondence to Aya Y. Badran, Department of Dermatology, Venereology and Andrology, Faculty of Medicine, Assiut university, Assiut, 71511, Egypt. Tel: 01013244819; fax: 0882080616;
e-mail: Ayabadran@aun.edu.eg, Aya_badran@yahoo.com

Received: 4 March 2021
Revised: 19 March 2021
Accepted: 2 April 2021
Published: 17 December 2021

Egyptian Journal of Dermatology and Venereology 2021, 42:53–59

Downloaded from http://journals.lww.com/ejdv by BhDMf5ePHKav1zEoum1tQfN4a+kJLhEZgbsIHo4XMi0hCywCX1AWnYQp/IlQrHD3i3D0OdRyi7TvSFl4Cf3VC4/OAVpDDa8KKGKV0Ymy+78= on 12/24/2024

Background
Trappin-2/Elafin is a 9.9 KDa molecule released from its precursor Preproelafin that exists principally in immune cells, skin, the lungs, the vagina, and other organs. Clusterin is a heterodimeric glycoprotein that plays a major role in many biological processes such as interaction with lipids, apoptosis regulation, weakening of complement activation, toxin removal, response to damage, and stress as well as autoimmune damage.
Both Trappin-2/Elafin and Clusterin serum levels have been studied in various immunologically mediated dermatological and nondermatological diseases. However, it still unknown whether their circulating levels are altered in pemphigus vulgaris (PV) and whether they play a role in this disease.
Objective
This study aimed to elucidate the potential link between both Trappin-2/Elafin and Clusterin levels and PV through a quantitative assessment of their serum levels by enzyme-linked immunosorbent assay and also to detect their possible correlations with PV severity using the pemphigus disease area index.
Patients and methods
Fifty patients with PV and 40 matched healthy controls were enrolled in this study. After a full assessment of history and complete dermatological examination, the severity score was calculated using pemphigus disease area index, and then serum samples were collected and subjected to quantitative measurements of serum Trappin-2/Elafin and Clusterin levels by enzyme-linked immunosorbent assay.
Results
Serum levels of both Trappin-2/Elafin and Clusterin were significantly higher in the patients than in the controls ($P<0.001$); still, their levels were not correlated with the severity of the disease.
Conclusion
The finding indicates that both Trappin-2/Elafin and Clusterin serum levels become elevated in patients with PV; however, the increase is not specific for the disease. None of the markers are correlated with the severity score of PV. Increased Trappin-2/Elafin levels indicate the existence of chronic inflammation, autoimmunity and skin or other system damage. Increased Clusterin levels suggest autoimmune damage, stress or transforming growth factor stimulation.

Keywords:
clusterin, pemphigus disease area index, pemphigus vulgaris, skin-derived antileukopeptidase, Trappin-2/Elafin

Egypt J Dermatol Venereol 42:53–59
© 2021 The Egyptian Society of Dermatology and Venereal Diseases
1110-6530

Introduction

Trappin-2/Elafin (also known as skin-derived antileukopeptidase (SKALP) or elastase-specific inhibitor), is one of the Chelonians, released by proteolysis from its precursor Preproelafin. At the ultrastructural level, Trappin-2/Elafin contains N-terminal transglutaminase motifs as well as the antiproteinase domain. This 9.9 KDa molecule exists principally in immune cells, skin, the lungs, the vagina and the gastrointestinal system [1].

The release of Trappin-2/Elafin is mediated through tumor necrosis factor-α (TNF-α), interleukin-1 (IL-1) and less probably by IL-6 [2]. It has the capacity to inhibit neutrophilic elastase, proteinase-3, and endogenous vascular elastase as well. Surprisingly, Elafin is cleaved by its target neutrophil elastase [3].

Trappin-2/Elafin is considered a key mediator in innate immune response. Moreover, it integrates both innate and adaptive immunity systems [3].

This is an open access journal, and articles are distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 License, which allows others to remix, tweak, and build upon the work non-commercially, as long as appropriate credit is given and the new creations are licensed under the identical terms.

Downloaded from http://journals.lww.com/ejdv by BhDMf5ePHKav1zEoum1tQfN4a+kJLhEZgbsIHo4XMi0hCywCX1AWnYQp/IlQrHD3i3D0OdRyi7TvSFl4Cf3VC4/OAVpDDa8KKGKV0Ymy+78= on 12/24/2024

In normal human epidermis, Trappin-2/Elafin is nondetectable; however, it becomes upregulated in inflammatory dermatoses as psoriasis [4], Neutrophilic dermatoses as dermatitis herpetiformis (DH) [5] and in various types of epidermal tumors [6,7].

The heterodimeric glycoprotein Clusterin is a well-known apolipoprotein J with a molecular weight of about 70–80 kDa. This glycoprotein has both nuclear and secreted isoforms. The secreted isoform is the most common one [8].

Structurally, Clusterin is composed of two chains linked together through disulfide bonds. In nearly all mammalian cells, Clusterin is manufactured either in response to stress or transforming growth factor (TGF) stimulation [9]. It plays a major role in many biological processes such as interaction with lipids, apoptosis regulation, weakening of complement activation, toxin removal, response to damage, and stress and autoimmune damage [10].

The intracellular isoform of Clusterin is either present inside the nucleus or in the cytosol, while the secreted isoform is either secreted into the extracellular space or into the biological fluids. The role of the intracellular form is to control DNA repair and to eliminate the misfolded proteins [11].

Interestingly, the nonglycated isoform in the nucleus has a proapoptotic effect, while the cytosolic form has an anti-apoptotic effect. This anti-apoptotic effect is mediated through its interaction with mitochondrial Bax, reducing the mitochondrial fission and production of reactive oxygen species [12]

At the extracellular level, the role of Clusterin is to eliminate the protein aggregates, and cellular debris, to promote degeneration of amyloid protein and to prevent its accumulation. It plays a non-negligible role in cellular proliferation and immune system regulation as it has both proinflammatory and both anti-inflammatory effects [13].

Its immunomodulatory role is mediated through inhibition of nuclear factor-B translocation, inhibition of complement system activation and through macrophage chemoattraction [14].

Autoimmune bullous disorders assimilate a group of diseases linked by the absence of tolerance to normal protein components of the epithelial cells of skin and mucous membranes. They has two main groups, 'Pemphigus' and 'Pemphigoid', and they represent major causes of considerable morbidity and mortality [15].

Intraepithelial clefting and acantholysis are the main features of the pemphigus group, which has multiple subtypes. Pemphigus vulgaris (PV) and pemphigus foliaceus are its most common subtypes. Each subtype has dissimilar clinical features. Considerable research has been carried out on the role of different immunological and inflammatory mediators in the pathogenesis of Pemphigus, although their results are not consistent [16].

PV is one of the classical examples of antigen-mediated autoimmune diseases, in which autoantibodies against adhesion molecules of the epidermis are formed. This in turn leads to loss of their adhesive function, which manifests clinically as blisters and erosions of the skin and mucous membranes. It has been verified that cytokine dysregulation and proinflammatory cytokines play the main role in the pathogenesis of PV; however, the exact mechanism is still being researched [17].

To date, the role of both Trappin-2/Elafin and Clusterin in the pathogenesis of PV has not been determined, and their levels have not been measured. Thus, we designed this study to investigate their role in this disease throughby measuring their serum level in patients with active PV. Furthermore, we aimed to detect their possible correlation with PV activity using the pemphigus disease area index (PDAI).

Patients and methods

We designed a case–control study on 50 PV patients and 40 matched healthy controls. The study was carried out at the department of Dermatology at Assiut University Hospital in the period from June 2020 to February 2021. The study protocol was approved by the Assiut Medical School Ethical Review Board (Ethical Committee N: IRB 17300554). A written informed consent was obtained from all participants. The control participants had no dermatological or chronic systemic diseases. We included only adult patients with active PV. (The active stage of PV was defined by Lee *et al.* [17], as the de-novo development of blisters on healthy, either unaffected or healed, skin or mucous membrane). Exclusion criteria were patients on regular treatment with systemic corticosteroids (or other immunosuppressive drugs) in the last 1 month and patients with other concomitant dermatological or chronic systemic diseases.

Downloaded from http://journals.lww.com/ejdv by BhDMf5ePHKav1zEoum1tQfN4a+kJLhEZgbsIHo4XMi0hCywCX1AWnYQp/IlQrHD3i3D0OdRyi7TvSFl4Cf3VC4/OAVpDDa8KKGKV0Ymy+78= on 12/24/2024

The diagnosis of each patient depended on clinical examination, histopathological examination (acantholysis and intraepidermal splitting) and direct immunofluorescence.

For every patient, a full assessment of history was performed including age, sex, education, residence, special habits, age at onset, and family history. A detailed cutaneous examination was performed including disease severity assessment by the PDAI. The PDAI ranges from 0 to 263, it measures effects on the skin and mucosa: 120 points for skin activity, 120 points for mucosal activity, 10 points for scalp activity and 13 points for disease damage. The body and the mucous membranes are divided into 12 parts each, while the scalp is considered as one area with four quadrants [18]. We used the classification by Shimizu *et al.* [19] (Mild <8; Moderate, from 9 to 24; and severe, ≥25).

Laboratory analysis

According to all study participants, 5 ml of aseptically withdrawn peripheral blood samples were separated and the sera were stored at −20°C for laboratory analysis. Human Clusterin ELISA kit (Bioassay Technology Laboratory Co., Ltd., Shanghai, China Catalog No: E1189 Hu), with detection limit range (0.5–300 ng/ml & Human Elafin ELISA kit (Bioassay Technology Laboratory Co., Ltd., Shanghai, China Catalog No: E4762 Hu), with detection limit range (5–1000 ng/L) were used for quantitative measurement of serum human Clusterin and Elafin levels. The assay was performed according to the manufacturer's instructions and protocols.

Statistical analysis

Data were analyzed using the Statistical Package for the Social Sciences (SPSS.20.0; SPSS Inc., Chicago, IL, USA) software program. Graphs were constructed using Graph Pad Prism 7 Software (San Diego, CA, USA). Data were described as mean, SD, range, and frequencies. A comparison of quantitative data among different groups was performed using a Student *t*-test and analysis of variance for the independent sample when normally distributed and the Mann–Whitney *U*-test for an independent sample when not normally distributed. Comparisons of the categorical variables were performed using the χ^2 test. A probability value (*P*) of less than 0.05 was considered statistically significant.

Results

We included 50 PV patients and 40 age-matched and sex-matched healthy volunteers in the study; 20 patients (40%) were males and 30 (60%) were females. Their ages ranged from 25 years to 70 years, with a mean±SD of 52±12.234 years. Family history was positive in only four (8%) of them. The total PDAI score in all patients ranged from 7 to 80, with a mean±SD of 39.78±26.615. The skin score ranged from 14 to 71, with a mean±SD of 37.74 ±16.939, while the mucous membrane score ranged from 5 to 22, with a mean±SD of 12.16±4.821. Other demographic and clinical data are illustrated in Table 1.

Table 1 Demographic and clinical data of pemphigus vulgaris patients

Characteristics	Mean±SD or *n* (%)
Age	
Mean±SD	52±12.234
Range	25–70
Sex	
Male	20 (40)
Female	30 (60)
Residence	
Rural	36 (72)
Urban	14 (28)
Smoking	
Yes	13 (26)
No	37 (74)
Education	
Yes	20 (40)
No	30 (60)
Age at onset (years)	
Mean±SD	50.74±12.938
Range	25–75
Total disease duration	
≤1 week	17 (34)
1–24 months	16 (32)
>24 months	17 (34)
Attack number	
First	17 (34)
Second	14 (28)
Third	3 (6)
Fourth	6 (12)
Fifth	3 (6)
Sixth	3 (6)
Seventh	2 (4)
Eighth	2 (4)
Family history	
Positive	4 (8)
Negative	46 (92)
Nikolisky's sign	
Positive	18 (36)
Negative	32 (64)
Skin score	
Mean±SD	37.74±16.939
Range (minimum–maximum)	14–71
Median (interquartile range)	38 (22.75–43.50)
Mucous membrane score	
Mean±SD	12.16±4.821
Range	5–22

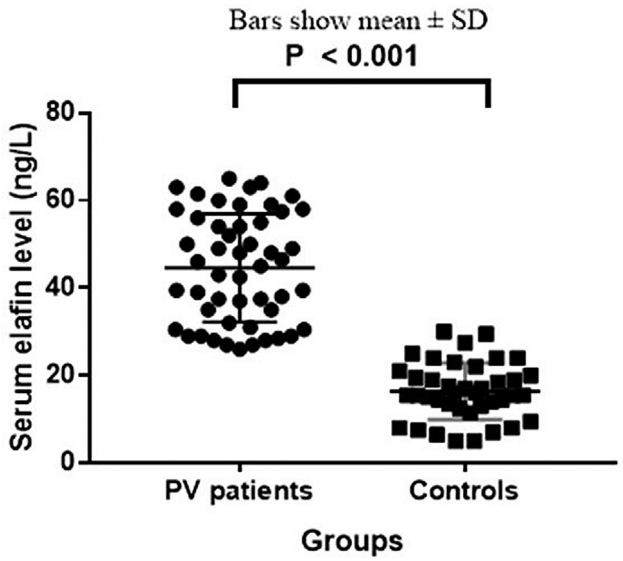
Downloaded from http://journals.lww.com/ejdv by BhDMf5ePHKav1zEoum1tQfN4a+kJLhEZgbsIHo4XMi0hCywCX1AWnYQp/IlQrHD3i3D0OdRyi7TvSFl4Cf3VC4/OAVpDDa8KKGKV0Ymy+78= on 12/24/2024

Table 2 Serum Trappin-2/Elafin and Clusterin levels in pemphigus vulgaris patients and controls

	Patients (*n*=50)	Controls (*n*=40)	*P*
Trappin-2/Elafin (ng/l)			
Mean±SD	44.61±12.389	16.412±6.519	<0.001***
Minimum–maximum	26–65	5–30	
Clusterin (ng/l)			
Mean±SD	101.44±8.313	84.262±7.815	<0.001***
Minimum–maximum	88–118	71–97	

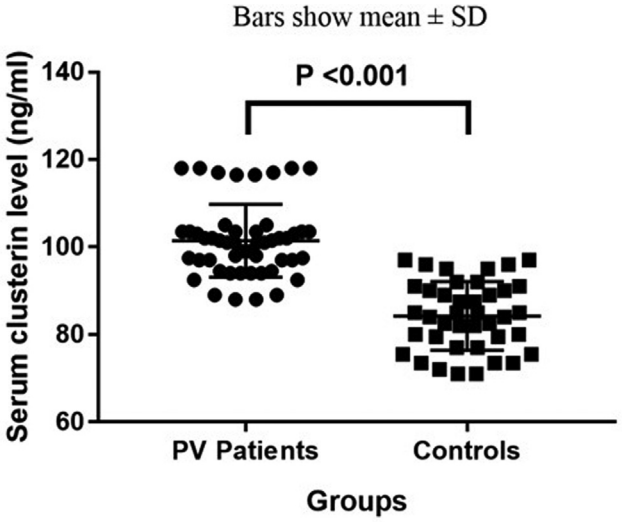
***Highly Significant difference between the level of serum Trappin-2/Elafin and Clusterin in patients and controls (*P* value 0.001).

Figure 1



Serum Trappin-2/Elafin levels in pemphigus vulgaris patients and controls.

Figure 2



Serum Clusterin levels in pemphigus vulgaris patients and controls.

A significant difference was present between the serum levels of both Trappin-2/Elafin and Clusterin in PV patients and controls ($P<0.001$ for both) (Table 2, Figs 1 and 2).

Table 3 shows the correlations of Trappin-2/Elafin and Clusterin serum levels and different tested variables in PV patients. All the correlations between the tested markers and age of the patients, age at onset, total PDAI, skin score and mucous membrane score were nonsignificant ($P>0.5$).

After dividing the patients according to their disease severity (using PDAI) into three groups (mild, moderate, and severe), we found that both Trappin-2/Elafin and Clusterin serum levels not differ significantly among patients ($P>0.5$) (Table 4).

Discussion

Several studies have proved that cytokines are key players in the pathogenesis of PV, as they coordinate both cellular and humoral responses [15]. The diversity of the dominant profiles of T helper (Th) cells, together with the diversity of cytokines and chemokines, could explain the heterogeneity of clinical manifestations of pemphigus. Thus, the description of a defined cell subset as the key regulator in the pathogenesis of pemphigus is not rational [17]. In the same way, the description of a defined cytokine as the key regulator in pemphigus pathogenesis is not rational.

Serum Trappin-2/Elafin levels have already been studied in various immunologically mediated dermatological and nondermatological diseases [3–7]. Also, serum Clusterin level has been evaluated in psoriasis [20–22], photoaged skin [23], malignant melanoma [24], and other malignancies [25].

In the present work, we explored Trappin-2/Elafin and Clusterin serum levels in 50 active PV patients and compared it with 40 matched healthy individuals. The mean serum level of both markers was significantly higher in patients than in the healthy control individuals ($P<0.001$). However, the measured levels were not correlated with the disease severity using PDAI; in addition, it was not correlated with other

Table 3 Correlations of Trappin-2/Elafin and Clusterin serum level and different tested variables in the pemphigus vulgaris patients

	Trappin-2/Elafin	Clusterin	Age	Age of onset	Total PDAI	Skin score	Mucous membrane score
Trappin-2/Elafin Elafin	1						
Clusterin	0.042	1					
	0.771						
Age	0.218	0.109	1				
	0.128	0.450					
Age of onset	0.107	0.049	0.907**	1			
	0.460	0.736	0.000				
Total PDAI	0.314	0.021	0.104	0.078	1		
	0.027	0.886	0.473	0.593			
Skin score	0.013	0.095	0.208	0.138	0.040	1	
	0.930	0.511	0.148	0.338	0.783		
Mucous membrane score	0.108	0.028	0.177	0.091	0.205	0.282*	1
	0.455	0.846	0.219	0.531	0.153	0.048	

PDAI, pemphigus disease area index. *Significant correlation between skin score and mucous membrane score (P value$<$ 0.1). **Highly Significant correlation between the age and the age of onset (P value$<$ 0.01).

Table 4 Serum Trappin-2/Elafin and Clusterin levels in pemphigus vulgaris patients classified according to disease severity measurement by pemphigus disease area index

	Mild (n=8)	Moderate (n=16)	Severe (n=26)	P^1	P^2	P^3	P^4
Trappin-2/Elafin (ng/l)							
Mean±SD	46.813±10.909	49.062±12.775	41.192±11.968	0.116	0.903	0.112	0.488
Minimum–maximum	28.5–65	26–63	27–64				
Clusterin (ng/l)							
Mean±SD	103.75±8.836	100.656±8.378	101.212±8.3261	0.686	0.675	0.977	0.738
Minimum–maximum	94.5–118	88–118	89–118				

P_1- value between the three groups (mild, moderate, and severe); P_2 value between the mild and moderate groups; P_3 value between the moderate and severe groups; P_4 value between the mild and severe groups.

Downloaded from http://journals.lww.com/ejdv by BhDMf5ePHKav1zEoum1tQfN4a+kJLhEZgbsIHo4XMi0hCywCX1AWnYQp/IlQrHD3i3D0OdRyi7TvSFl4Cf3VC4/OAVpDDa8KKGKV0Ymy+78= on 12/24/2024

variables such as age of the patients or age at onset of the disease.

We divided the included patients according to the disease severity grading into three groups (mild, moderate, and severe) and we found that the measured serum levels did not differ significantly among them (P>0.5).

The expression of Trappin-2/Elafin varied among different dermatological diseases; some authors have described its overexpression in disorders with a predominant neutrophilic infiltrate such as Sweet syndrome, Behcet syndrome, and other Neutrophilic vasculitis [5], while others reported the opposite result [3].

DH is an example of an immunologically mediated vesiculobullous disease. There have been debates on the role of Trappin-2/Elafin in this disorder. Ollague *et al.* [3] claimed that skin biopsies from patients with active DH showed minimal to absent epidermal appearance of Trappin-2/Elafin. They believed that the abnormal expression suggests the presence of a deficient innate immunity against the neutrophilic infiltrate.

It has been found that in acute and chronic inflammations (as in infections, graft rejection, diabetes, colitis, and inflammatory skin diseases), Trappin-2/Elafin defends tissue against harm. As chronic inflammatory processes are commonly accompanied by excessive neutrophil-released enzyme activity, Trappin-2/Elafin overwhelms the immune response; also, it shows antimicrobial activity against diverse types of pathogens [26].

High circulating serum Trappin-2/Elafin levels lead to higher risk of mortality among patients with graft-versus-host disease (GVHD). Paczesny *et al.* [27] verified that it has both diagnostic and prognostic values in GVHD Furthermore, skin rashes of GVHD can be differentiated from other skin rashes that develop after bone marrow transplantation by quantitatively assessing serum Trappin-2/Elafin level [28].

In addition, high serum Trappin-2/Elafin levels are present in other autoimmune diseases such as rheumatoid arthritis and systemic sclerosis and Psoriasis [4,29].

Downloaded from http://journals.lww.com/ejdv by BhDMf5ePHKav1zEoum1tQfN4a+kJLhEZgbsIHo4XMi0hCywCX1AWnYQp/IlQrHD3i3D0OdRyi7TvSFl4Cf3VC4/OAVpDDa8KKGKV0Ymy+78= on 12/24/2024

As TNF-α, IL-1, and IL-6 are considered stimulators for Elafin release [4,5], and as their levels increase in PV, we could explain the increase of Elafin levels on this basis.

In the current work, we could explain the increase in serum Trappin-2/Elafin level on 4 bases: first, Trappin-2/Elafin released from epidermal cells in response to damage produced by the disease itself [1]. Second, IL-1, IL-6, and TNF-α, which are common stimulators for Trappin-2/Elafin, are commonly elevated in PV patients [4,5]. Third, damage that occurs elsewhere during the course of PV as Trappin-2/Elafin could be caused by bronchial epithelium. Finally, Trappin-2/Elafin production might mirror a chronic inflammatory process [1,2].

Tavakolpour *et al.* [15] in their systematic review, studied the protective and pathogenic roles of diverse cytokines in PV. They found that the levels of TNF-α, TGF-β, IL-8, IL-10, IL-12, IL-17, and IL-21 were elevated, while levels of IL-2 and IL-23 were decreased. Furthermore, they found diverse studies showing the association of TNF-α and IL-6 with disease activity as well as values of autoantibodies. Studies revealed that the multifunctional stress-induced glycoprotein (Clusterin) is present in all body fluids; furthermore, it is expressed excessively in diseased and aged tissues [23]. In dermal connective tissue alterations such as solar elastosis and in degenerative diseases such as Alzheimer's disease [30], higher Clusterin levels have been detected.

In respect to dermatological disorders, the data on the relation between Clusterin and psoriasis have shown conflicting results. Some authors believed that Clusterin level is related to the presence and activity of Psoriasis [20], while others refute this role [21,22]. Clusterin might serve as either a protective or aggravating factor in the pathogenesis of psoriasis [31].

In rheumatoid arthritis [32] and atopic dermatitis [33], elevated serum Clusterin levels occur as a compensatory mechanism; accordingly, in these cases, Clusterin could play a protective role.

In terms of mycosis fungoides and Sézary syndrome, Tobisawa *et al.* [34], who performed immunostaining in diseased tissue, claimed that Clusterin expression could be used as a prognostic marker in early-stage mycosis fungoides. Furthermore, in systemic sclerosis, serum Clusterin levels were found to be higher than normal [35].

Even though Clusterin plays a principal role in many processes, including autoimmunity, inflammation, and immunological activities, it remains a mysterious protein [31].

In this work, we might elucidate the link between the elevated serum Clusterin level and the presence of PV on the basis that Clusterin is released in response to either stress, TGF stimulation or autoimmune damage.

Unfortunately, in the present work, we found only a clinical association between elevated Trappin-2/Elafin and Clusterin levels and the presence of PV and not causation. Therefore, we recommend further studies to explain this presumed relation.

The limitations of our study were as follows: the relatively small number of patients included, difference between the number of cases and controls and reliance on a single investigatory method (ELISA). Also, we didn't measure the markers level during remission.

Conclusion

Our finding indicates that both Trappin-2/Elafin and Clusterin serum levels become elevated in patients with PV; however, the increase is not specific for the disease. None of the markers are correlated with the severity score of PV. Elevated Trappin-2/Elafin levels indicate the presence of chronic inflammation, autoimmunity and skin or other system damage. Elevated Clusterin levels suggest Autoimmune damage, stress or TGF stimulation.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

References

1 Guyot N, Butler MW, McNally P, Weldon S, Greene CM, Levine RL, *et al.* Elafin, an elastase-specific inhibitor, is cleaved by its cognate enzyme neutrophil elastase in sputum from individuals with cystic fibrosis. J Biol Chem 2008; 283:32377–32385.

2 Williams SE, Brown TI, Roghanian A, *et al.* SLPI and elafin: one glove, many fingers. Clin Sci. 2006; 110:21–35.

3 Ollague JE, Nousari JE, Carlos H. Expression of elafin in dermatitis herpetiformis. Am J Dermatopathol 2018; 40:1-6.

4 Elgharib I, Khashaba SA, Elsaid HH, Sharaf MM. Serum elafin as a potential inflammatory marker in psoriasis. Int J Dermatol 2018; 58:205–209.

Downloaded from http://journals.lww.com/ejdv by BhDMf5ePHKav1zEoum1tQfN4a+kJLhEZgbsIHo4XMi0hCywCX1AWnYQp/IlQrHD3i3D0OdRyi7TvSFl4Cf3VC4/OAVpDDa8KKGKV0Ymy+78= on 12/24/2024

5 Tanaka N, Fujioka A, Tajima S, Ishibashi A, Hirose S. Elafin is induced in epidermis in skin disorders with dermal neutrophilic infiltration: Interleukin-1 and tumour necrosis factor-stimulate its secretion in vitro. Br J Dermatol 2000; 143:728–732.

6 Zhang M, Zou Z, Maass N, Sager R. Differential expression of elafin in human normal mammary epithelial cells and carcinomas is regulated at the transcriptional level. Cancer Res. 1995; 55:2537–2541.

7 Yu KS, Jo JY, Kim SJ, Lee Y, Bae JH, Chung YH, *et al.* Epigenetic regulation of the transcription factor Foxa2 directs differential elafin expression in melanocytes and melanoma cells. Biochem Biophys Res Commun. 2011; 408:160–166.

8 Aronow BJ, Lund SD, Brown TL, Harmony JA, Witte DP. Apolipoprotein J expression at fluid-tissue interfaces: poten tial role in barrier cytoprotection. Proc Natl Acad Sci USA. 1993; 90:725–729.

9 Michel D, Chatelain G, North S, Brun G. Stress-induced transcription of the clusterin/apoJ gene. Biochem J 1997; 328:45–50.

10 Jones SE, Jomary C. Clusterin. Int J Biochem Cell Biol 2002; 34:427–431.

11 Trougakos IP, Lourda M, Antonelou MH, Kletsas D, Gorgoulis VG, Papassideri IS, *et al.* Intracellular clusterin inhibits mitochondrial apoptosis by suppressing p53-activating stress signals and stabilizing the cytosolic Ku70-Bax protein complex. Clin Cancer Res 2009; 15:48–59.

12 Itakura E, Chiba M, Murata T, Matsuura A. Heparan sulfate is a clearance receptor for aberrant extracellular proteins. J Cell Biol 2020; 219: e201911126.

13 Essabbani A, Margottin-Goguet F, Chiocchia G. Identification of clusterin domain involved in NF-B pathway regulation. J Biol Chem 2009; 285:4273–4277.

14 Shim Y-J, Kang B-H, Choi B-K, Park I-S, Min B-H. Clusterin induces the secretion of TNF- and the chemotactic migration of macrophages. Biochem Biophys Res Commun 2012; 422:200–205.

15 Tavakolpour S, Mahmoudi H, Mirzazadeh A, *et al.* Pathogenic and protective roles of cytokines in pemphigus: a systematic review. Cytokine 2020; 129:155026.

16 Kridin K. Pemphigus group: overview, epidemiology, mortality, and comorbidities. Immunol Res. 2018; 66:255–270.

17 Lee SH, Hong WJ, Kim SC. Analysis of serum cytokine profile in pemphigus. Ann Dermatol. 2017; 29:438–445.

18 Murrell DF, Dick S, Ahmed AR, *et al.* Consensus statement on definitions of disease, end points, and therapeutic response for pemphigus. J Am Acad Dermatol. 2008; 58:1043–1046.

19 Shimizu T, Takebayashi T, Sato Y, *et al.* Grading criteria for disease severity by pemphigus disease area index. J Dermatol. 2014; 41:969–973.

20 Buquicchio R, Foti C, Loconsole F, Polimeno L, Ventura MT. Clusterin serum level: How does it affect psoriatic patients? J Biol Regul Homeost Agents 2017; 31:785–789.

21 Ghazy A, Salem A, Abdel-hameed A, Tahoun A. Serum clusterin in psoriasis: relation to metabolic syndrome. AIMJ 2020; 1:64–68.

22 Garcia-Rodriguez S, Arias-antiago S, Perandres-Lopez R, Orgaz-Molina J, Castellote L, Buendía-Eisman A, *et al.* Decreased plasma levels of clusterin in patients with psoriasis. Actas Dermosifiliogr. 2013; 104:497–503.

23 Janig E, Haslbeck M, Aigelsreiter A, Braun N, Unterthor D, Wolf P, *et al.* Clusterin associates with altered elastic fibers in human photoaged skin and prevents elastin from ultraviolet-induced aggregation in vitro. Am J Pathol. 2007; 171:1474–1482.

24 Shannan B, Seifert M, Leskov K, *et al.* Clusterin (CLU) and melanoma growth: CLU is expressed in malignant melanoma and 1, 25-dihydroxyvitamin D3 modulates expression of CLU in melanoma cell lines in vitro. Anticancer Res. 2006; 26:2707–2716.

25 Pucci S, Bonanno E, Pichiorri F, Angeloni C, Spagnoli LG. Modulation of different clusterin isoforms in human colon tumorigenesis. Oncogene 2004; 23:2298–2304.

26 Simpson AJ, Maxwell A, Govan J, Haslett C, Sallenave J-M. Elafin (elastase-specific inhibitor) has anti-microbial activity against Gram-positive and Gram-negative respiratory pathogens. FEBS Lett. 1999; 452:309–313.

27 Paczesny S, Braun TM, Levine JE, *et al.* Elafin is a biomarker of graft-versus-host disease of the skin. Sci Transl Med. 2010; 2:13ra2.

28 Antin JH, Deeg HJ. Clinical spectrum and pathophysiology of acute graft-vs.-host disease. In Ferrara J, Cooke K, Deeg HJ, eds. Graft-vs-Host Disease. New York, NY: Marcel Dekker Inc; 2005. 369–381

29 Alkemade H, Kerkhof P van de, Schalkqijk J. Demonstration of skin-derived antileukoproteinase (SKALP) in urine of psoriatic patients. J Invest Dermatol. 1992; 99:3–7.

30 Foster EM, Dangla-Valls A, Lovestone S, Ribe EM, Buckley NJ. Clusterin in Alzheimer's disease: mechanisms, genetics, and lessons from other pathologies. Front Neurosci. 2019; 13:164.

31 Ataseven A, Kesli R, Kurtipek GS, *et al.* Assessment of lipocalin 2, clusterin, soluble tumor necrosis factor receptor-1, interleukin-6, homocysteine, and uric acid levels in patients with psoriasis. Dis Markers. 2014; 2014:541709.

32 Devauchelle V, Essabbani A, De Pinieux G, Germain S, Tourneur L, Mistou S, *et al.* Characterization and functional consequences of underexpressionof clusterin in rheumatoid arthritis. J Immunol 2006; 177:6471–6479.

33 Sol IS, Kim YH, Lee KE, Hong JY, Na Kim M, Kim YS, *et al.* Serum clusterin level in children with atopic dermatitis. Allergy Asthma Proc 2016; 37:335–339.

34 Tobisawa S, Honna M, Ishida-Yamamoto A, Saijo Y, Iizuka H. Prognostic factors in 105 Japanese cases of mycosis fungoides and Sezary syndrome: clusterin expression as a novel prognostic factor. J Dermatol Sci 2013; 71:160–166.

35 Yanaba K, Asano Y, Tada Y, Sugaya M, Kadono T, Sato S. A possible contribution of elevated serum clusterin levels to the inhibition of digital ulcers and pulmonary arterial hypertension in systemic sclerosis. Arch Dermatol Res 2012; 304:459–463.