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RESEARCH

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Vascular endothelial growth factor in hypersensitivity pneumonitis and connective tissue disease-associated interstitial lung disease

Yasmine Hamdy El-Hinnawy[1], Nehal El-Ghobashy[2], Radwa Marawan Abdel Halim[3] and Gihan Saad Abo Elwafa[1*]

Abstract

Background A growing evidence on the role of vascular endothelial growth factor (VEGF) in the pathogenesis of interstitial lung diseases accumulated over the past decade; with the development of nintedanib for the treatment of fibrotic interstitial lung diseases, our aim was to quantify serum levels of VEGF in patients' hypersensitivity pneumonitis (HP) and connective tissue disease-associated interstitial lung diseases (CTD-ILD) with an assessment of its relationship with functional status parameters and echocardiographic findings.

Methods Spirometry, 6-min walking test, echocardiography, and serum VEGF levels were assessed in HP and CTD-ILD patients.

Results The study included 31 HP patients, 30 CTD-ILD patients, and 29 control subjects. VEGF level was significantly higher in HP patients than in patients with CTD-ILD and control subjects. VEGF level showed positive correlation with 6-min walk distance and forced vital capacity percent predicted and inverse correlation with percent desaturation in 6-min walk test, dyspnea score, and echocardiographic findings in both groups.

Conclusion Serum VEGF is higher in HP patients than in patients with CTD-ILD and control.

Keywords Vascular endothelial growth factor, Hypersensitivity pneumonitis, Connective tissue disease-associated interstitial lung diseases

Introduction

Biomarkers are of great interest in interstitial lung diseases (ILD), as they can help in the diagnosis, the assessment of severity, and predicting the prognosis of the disease. They are simple, easier, and more rapid than invasive procedures for diagnosis. Vascular endothelial growth factor (VEGF) is a tyrosine kinase glycoprotein important in the regulation of endothelial function, capillary permeability, and angiogenesis, playing an important role in maintaining normal lung, angiogenesis of endothelial cells, and restoration of pulmonary circulation. Given that inflammation, abnormal angiogenesis, and altered fibrosis are involved in the pathogenesis of ILD, VEGF is believed to have a role [1]. Hypersensitivity pneumonitis (HP) showed an upregulation of alveolar epithelial apoptosis markers suggesting an important role for alveolar epithelial cell integrity in this disease [2].

*Correspondence:
Gihan Saad Abo Elwafa
gigi2012chest@gmail.com
[1] Department of Chest Diseases, Faculty of Medicine, Cairo University, Manial, Cairo, Egypt
[2] Department of Rheumatology and Rehabilitation, Faculty of Medicine, Cairo University, Manial, Cairo, Egypt
[3] Department of Clinical and Chemical Pathology, Faculty of Medicine, Cairo University, Manial, Cairo, Egypt

Previous studies raised a question about the diagnostic utility of VEGF in HP as they found an elevated serum level in HP patients when compared with the control group [3, 4].

VEGF is believed to contribute in the progression of connective tissue disease-associated interstitial lung diseases (CTD-ILD), as it plays a dual role in encouraging and inhibiting pulmonary fibrosis. Also, VEGF subtype expression imbalance can contribute to the development of the disease, in which proliferation of endothelial cells and fibroblasts can be induced by VEGF [5]. Our aim was to compare serum VEGF level in HP and CTD-ILD patients in relation to control and to assess the relationship between serum level of VEGF, functional severity, and presence of pulmonary hypertension.

Methods

This prospective case control study was conducted from February 2023 and December 2023. The study was conducted in accordance with the Helsinki Declaration and was approved by the research ethics committee of our institute (No: N-144–2023). A written informed consent was obtained from all patients.

Patients with HP and CTD-ILD aging 18 years or more were included. HP was diagnosed according to ATS/JRS/ALAT clinical practice guidelines for the diagnosis of hypersensitivity pneumonitis in adults [6]. Connective tissue diseases were diagnosed according to relevant guidelines [7–9].

Data collection

Medical history included comorbidities, smoking history, and modified Medical Research Council (MMRC) dyspnea score. High-resolution computed tomography (HRCT) to determine the type and extent of ILD, 6-min walking test (6MWT), spirometry to assess the functional capacity, and transthoracic echocardiography to assess the probability of pulmonary hypertension and right heart affection were done. Quantification of serum VEGF level was done using enzyme-linked immunosorbent assay (human VEGF Elisa kit, Cloud-Clone Corp, Houston, USA).

Statistical methods

The program used for statistical analysis was IBM SPSS (Statistical Package for the Social Science; IBM Corp, Armonk, NY, USA) release 22 for Microsoft Windows. Data were statistically described as mean ± standard deviation (SD) or frequencies (number of cases) and percentages when indicated. The following tests were used: Kolmogorov–Smirnov test, Mann–Whitney *U* test, and Spearman rank correlation equation. Two-sided *P* values were considered statistically significant if less than 0.05.

Results

The study included 90 subjects: 31 patients with HP, 30 patients with CTD-ILD, and 29 age- and sex-matched healthy control subjects.

HP patients

This group of patients comprised 28 females (90.3%) and 3 males (9.7%). The mean age was 48.03 ± 11.07 years. Only 12 patients (38.7%) had comorbidities, in the form of systemic hypertension, diabetes mellitus, and ischemic heart disease.

Regarding radiological findings, 14 patients (45.5%) had evidence of fibrosis, and 17 patients (54.8%) were non-fibrotic. Regarding functional assessment, the mean of MMRC dyspnea score was 2.2 ± 1, the mean of percent desaturation during 6MWT was 12 ± 9%, and the mean 6-min walking distance (6MWD) was 220 ± 131 m. Spirometry results showed that the mean value of forced vital capacity percent predicted (FVC%) was 44 ± 16%.

Regarding echocardiographic findings (available for only 27 patients), 27 patients (100%) had normal right ventricular systolic function, 6 patients (22.2%) had right side dilatation, and 9 patients (33.3%) had evidence of pulmonary hypertension.

The mean value of serum VEGF was 664.58 ± 375.95 pg/ml, and the mean value of serum VEGF in the control group was 205.69 ± 44.89 pg/ml (*P* value < 0.0001).

Table 1 shows the correlation between serum VEGF level, functional assessment parameters, and echocardiographic findings in the HP group.

Table 1 Correlation between serum VEGF level, functional assessment parameters, and echocardiographic findings in the HP group

VEGF		
% desaturation during 6MWT	Correlation coefficient	−0.108
	P value	0.590
6MWD	Correlation coefficient	0.059
	P value	0.771
MMRC	Correlation coefficient	−0.235
	P value	0.203
FVC %	Correlation coefficient	0.194
	P value	0.304
Presence of pulmonary hypertension	Correlation coefficient	−0.101
	P value	0.617
Right-sided dilatation	Correlation coefficient	−0.114
	P value	0.570

Table 2 Relationship between serum VEGF level and radiological findings in the HP group

VEGF			
Radiological findings	**Mean ± SD**	***N***	***P* value**
Fibrotic	666.71 ± 351.871	14	0.968
Non-fibrotic	662.82 ± 405.470	17	

Table 3 Correlation between percent desaturation during 6MWT and FVC% and MMRC score in the HP group

% Desaturation		
FVC %	Correlation coefficient	−0.477
	P value	**0.014**
MMRC	Correlation coefficient	0.578
	P value	**0.002**

Table 4 Relationship between percent desaturation during 6MWT and radiological findings in the HP group

% Desaturation			
Radiological findings	**Mean ± SD**	***N***	***P* value**
Fibrotic	13.91 ± 10.015	11	0.3
Non-fibrotic	10.19 ± 9.050	16	

Table 2 shows the relationship between serum VEGF level and radiological findings in the HP group.

Table 3 shows the correlation between percent desaturation during 6MWT and FVC% and MMRC score in the HP group.

Table 4 shows the relationship between percent desaturation during 6MWT and radiological findings in the HP group.

CTD-ILD patients

This group of patients comprised 24 females (80%) and 6 males (20%). The mean age was 46.9 ± 11.96 years. Only 7 patients (23.3%) had comorbidities in the form of systemic hypertension and diabetes mellitus. In our study group, connective tissue diseases were systemic sclerosis, rheumatoid arthritis, dermatomyositis/polymyositis, and mixed connective tissue disease.

Regarding radiological findings, 9 patients (30%) had radiological evidence of fibrosis, and 21 patients (70%) were non-fibrotic. Regarding the functional assessment, the mean of MMRC dyspnea score was 1.7 ± 0.9, the mean of percent desaturation during 6MWT was 4 ± 4.5%, and the mean 6MWD was 269 ± 122 m. Spirometry results showed that mean value of FVC% was 63 ± 24%.

Table 5 Correlation between serum VEGF level, functional assessment parameters, and echocardiographic findings in the CTD-ILD group

VEGF		
% desaturation during 6MWT	Correlation coefficient	−0.037
	P value	0.855
6MWD	Correlation coefficient	0.095
	P value	0.629
MMRC	Correlation coefficient	−0.179
	P value	0.343
FVC %	Correlation coefficient	0.400
	P value	**0.029**
Tricuspid annular plane systolic excursion	Correlation coefficient	−0.249
	P value	0.194
Presence of pulmonary hypertension	Correlation coefficient	−0.183
	P value	0.343
Right-sided dilatation	Correlation coefficient	−0.020
	P value	0.917

Table 6 Relationship between serum VEGF level and radiological findings in the CTD-ILD group

VEGF			
Radiological findings	**Mean ± SD**	***N***	***P* value**
Fibrotic	211.00 ± 11.456	9	0.874
Non-fibrotic	208.62 ± 69.250	21	

Regarding echocardiographic findings (available for 29 patients only), 28 patients (96.6%) had normal right ventricular systolic function, 3 patients (10.3%) had right side dilatation, and 11 patients (37.9%) had evidence of pulmonary hypertension.

The mean value of serum VEGF was 209.33 ± 57.83 pg/ml (*P* value 0.789 between CTD-ILD and control groups).

Table 5 shows the correlation between serum VEGF level, functional assessment parameters, and echocardiographic findings in the CTD-ILD group.

Table 6 shows the relationship between serum VEGF level and radiological findings in the CTD-ILD group.

Table 7 shows the correlation between percent desaturation during 6MWT and FVC% and MMRC score in the CTD-ILD group.

Table 8 shows the relationship between percent desaturation during 6MWT and radiological findings in the CTD-ILD group.

Discussion

Very few studies examined the role of VEGF in HP. In the small numbers studied, serum VEGF levels were elevated compared with controls [2]. In our study, the mean value

Table 7 Correlation between percent desaturation during 6MWT and FVC% and MMRC score in the CTD-ILD group

% Desaturation		
FVC %	Correlation coefficient	−0.409
	P value	**0.034**
MMRC	Correlation coefficient	0.472
	P value	**0.013**

Table 8 Relationship between percent desaturation during 6MWT and radiological findings in the CTD-ILD group

% desaturation			
Radiological findings	**Mean ± SD**	***N***	***P* value**
Fibrotic	6.13 ± 4.121	8	0.060
Non-fibrotic	3.42 ± 4.513	19	

Table 9 Mean value of serum VEGF in study groups

HP	**Control**	***P* value**
664.58 ± 375.95	205.69 ± 44.89	< 0.0001
CTD-ILD	Control	*P* value
209.33 ± 57.83	205.69 ± 44.89	0.789

of serum VEGF in the HP group was significantly higher than that of the CTD-ILD group (however, the 2 groups were not matched regarding functional status) and control group (*P* value < 0.0001) (Table 9); this agrees with previous studies by Navarro et al. [3] and Yamashita et al. [4].

Serum VEGF level showed a positive correlation with 6MWD, FVC%, and inverse correlation with percent desaturation during 6MWT, MMRC score, presence of pulmonary hypertension, and right-sided cardiac affection, but this was statistically insignificant. Yamashita et al. [4] stated that the serum levels of VEGF were not correlated with the pulmonary function tests in HP patients.

As far as we know, no previous studies assessed the relationship between serum VEGF level and results of 6MWT, MMRC score, radiological findings, or echocardiographic findings in patients with HP. Zhong and Luo [10] found that serum VEGF was significantly and positively correlated with HRCT scores in IPF patients, suggesting an association with disease severity. In addition, elevated serum VEGF levels were closely associated with impairment of lung function. Ando et al. [11] reported no correlation between serum VEGF level and the results of pulmonary function tests in IPF patients. Another study by Ventetuolo et al. [12] showed that VEGF levels were not correlated with FVC, 6MWD, or New York Heart Association functional class in patients with IPF and that there was no significant correlation between VEGF levels and hemodynamics.

In our CTD-ILD group, there was no statistically significant difference between the mean value of serum VEGF and that of control group (*P* value 0.789). Previous studies by Hashimoto et al. [13] and Kikuchi et al. [14] found that serum VEGF levels were significantly higher in patients with rheumatic diseases compared with healthy controls, and VEGF levels were correlated with the presence of ILD. Saranya et al. [15] reported that median serum VEGF in systemic sclerosis patients was significantly higher than in controls; also, De Santis et al. [16] found that serum VEGF was higher in systemic sclerosis patients versus healthy controls with lower VEGF levels in the serum of patients with evidence of ILD.

In this study, serum VEGF levels showed positive correlation with 6MWD, and a significant positive correlation with FVC %, but showed inverse correlation with percent desaturation during 6MWT, MMRC score, presence of pulmonary hypertension, tricuspid annular plane systolic excursion, and right-sided cardiac dilatation, and this was statistically insignificant. Saranya et al. [15] found that serum VEGF levels were inversely correlated with FVC and that there was a significant positive correlation with the MMRC dyspnea score.

Our results showed that there was no statistically significant relationship between serum VEGF levels and radiological findings, while De Santis et al. [16] reported that among systemic sclerosis cases, serum VEGF was directly correlated with ground glass and reticular pattern extent on HRCT, and Lv et al. [5] showed that VEGF levels were positively correlated with CTD-ILD severity by HRCT.

Limited exercise tolerance is a major symptom of ILD, resulting in reduced ability to perform daily activities and poor quality of life [17]. Exercise-induced desaturation is an index of the severity of interstitial lung disease [18]. The 6MWT is a simple test of exercise capacity that is commonly used to assess the functional status and follow-up treatment responses in ILD patients [19].

In our study, percent desaturation during 6MWT showed a significant inverse correlation with FVC% and a significant positive correlation with MMRC dyspnea score in both groups but showed a statistically insignificant relationship with radiological findings on HRCT in both groups. A relationship between desaturation during 6MWT and parameters of pulmonary function was confirmed by Rosa et al. [20] and Aktan et al. [21], while Seema et al. [22] found no statistically significant correlation between percent desaturation during 6MWT and

spirometry results in CTD-ILD patients. Villalba et al. [23] found that in patients with scleroderma-associated ILD, a statistical association was found between percent desaturation and dyspnea score, fibrosis on chest radiograph, FVC < 80% of the predicted value, and presence of ground glass or reticular opacities on HRCT.

Conclusion

From our results, we could conclude that serum VEGF levels were significantly higher in the HP group than the control group, but in CTD-ILD patients, the level was almost the same as control group. However, serum VEGF level could not be correlated to functional status or disease severity in both groups.

Abbreviations

6MWD	Six-minute walking distance
6MWT	Six-minute walking test
CTD-ILD	Connective tissue disease-associated interstitial lung diseases
FVC%	Forced vital capacity percent predicted
HP	Hypersensitivity pneumonitis
HRCT	High-resolution computed tomography
ILD	Interstitial lung diseases
MMRC	Modified Medical Research Council
SD	Standard deviation
VEGF	Vascular endothelial growth factor

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Authors' contributions

All authors shared equally in the conception, design of the work; acquisition, analysis, and interpretation of data; and drafting and revising of the work. All authors have approved the submitted version and agreed both to be personally accountable for the author's own contributions and to ensure that questions related to the accuracy or integrity of any part of the work, even ones in which the author was not personally involved, are appropriately investigated, resolved, and the resolution documented in the literature.

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Availability of data and materials

All data generated or analyzed during this study are included in this published article.

Declarations

Ethical approval and consent to participate

This study was approved by research ethics committee, Faculty of Medicine, Cairo University (No: N-144–2023). Written informed consent was taken from every patient.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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