

Serum Endocan and Carotid Intima Media Thickness Evaluation in Vitiligo Patients

Khaled M Monib¹, Mohamed S Hussein¹, Amira M Abdelrahman², Mohamed S Darwish³, Rania M Elhady^{4*}

¹ Dermatology, Venereology and Andrology Department,

² Clinical and Chemical Pathology Department,

³ Cardiology Department, Faculty of Medicine, Benha University, Benha, Egypt

⁴ Dermatology, Venereology and Andrology Department, Faculty of Medicine, Zagazig University, Zagazig, Egypt

*Corresponding author: Rania M Elhady, Phone number: 01554464636,

E-Mail: dr.raniaelhady@gmail.com

ABSTRACT

Background: Vitiligo is an acquired pigmentary disorder of the skin characterised by patches and circumscribed depigmented macules caused by epidermal melanocyte loss. A vasculoprotective molecule, endocan is an indicator of endothelial function.

Objective: We aimed to evaluate carotid intima media thickness (CIMT) and serum endocan level in vitiligo patients and to compare their levels with healthy controls.

Methods: This case-control study involved sixty male vitiligo patients aged 18 years or older, in addition to twenty healthy controls of the same age. A comprehensive evaluation of each patient included general and clinical examination, the Vitiligo Extent Tensity Index (VETI) score that was used to quantify the severity of vitiligo, a laboratory analysis to determine the serum endocan level, and Doppler ultrasound imaging of the CIMT.

Results: No statistically significant differences were observed between the patient and control groups with respect to the age, gender, and body mass index. Patients had a significantly higher mean CIMT than the control group.

Conclusion: Serum endocan and CIMT levels were significantly higher in vitiligo patients than in the control group and were significantly correlated with the severity of the disease, according to the study.

Keywords: Serum endocan, Carotid intima media thickness, Vitiligo patients, Epidermal melanocyte.

INTRODUCTION

Vitiligo, which occurs when epidermal melanocytes are selectively lost, is the most prevalent disorder of skin depigmentation, impacting approximately 0.5 % of the global populace. No discernible disparities in incidence rates are observed based on phenotype or race and both sexes are equally impacted [1].

The involvement of genetic, immunological, cytotoxic, neuronal, and inflammatory factors in the complex pathogenesis of vitiligo may account for a broad spectrum of its systemic manifestations. Proinflammatory cytokines such as interleukin-6 (IL-6), tumour necrosis factor (TNF), and IL-1 are recognised for their role in inducing insulin resistance, metabolic complications, and atherosclerosis, all of which are associated with vitiligo [2].

Endocan is a molecule specific to human endothelial cells that promotes angiogenesis and inhibits leukocyte binding to vascular endothelium. Endocan might play a role in the development of a number of inflammatory diseases [3].

Globally, cardiovascular disease is characterised by significant morbidity and mortality. There is some evidence that a high level of endocan may be closely associated with the development and progression of cardiovascular disease. The fact that serum endocan levels were higher in patients with cardiovascular disease initially suggested that endocan levels and cardiovascular disease are related [4].

Enhanced concentrations of endocan in the plasma may indicate endothelial dysfunction and activation. It is also regarded as a possible immuno-inflammatory

marker associated with cardiovascular disease. Endocan plays a crucial role in the prognosis and prediction of cardiovascular disease progression [5].

The intima-media thickness is characterised by a double-line interface between the lumen, intima, and media-adventitia. Its configuration can be observed longitudinally on both walls of the common carotid artery using two-dimensional ultrasound. The effectiveness of carotid intima-media thickness in detecting early non-calcified subclinical atherosclerotic changes was established. The European Society of Hypertension and European Society of Cardiology have reaffirmed carotid intima-media thickness (> 0.9 mm) as an indicator of asymptomatic atherosclerotic changes in their hypertension guidelines [6]. Age and gender are determinants of normal CIMT values, as age increases, CIMT rises significantly and steadily, with men having significantly higher values than women [7].

The purpose of the study was to assess serum endocan level and carotid intima media thickness in vitiligo patients and to compare their levels with healthy controls.

METHODS

This case-control study encompassed sixty individuals diagnosed with vitiligo and twenty healthy volunteers who were matched in age and gender. They were recruited from Benha University Hospitals' Outpatient Clinics of Dermatology, Venereology, and Andrology.

Inclusion criteria: All participants were willing to participate in the study and were at least 18 years old.

Exclusion criteria: Patients who were smokers, pregnant or lactating, had a confirmed history of active cancer, or had other systemic diseases such as cardiovascular disease, diabetes, chronic liver disease, or renal disease, and patients receiving antihyperlipidemic medications. Additionally, vitiligo patients who had been treated systemically for the condition for less than one month prior to the study.

All participants were subjected to the followings:

Full history taking: That included past history of medications, associated systemic diseases or autoimmune diseases, personal history, history of the present condition, history of previous treatment of vitiligo and family history of vitiligo.

General examination: A clinical examination was conducted for all participants to identify any systemic diseases. The BMI was computed utilising the subsequent formula: BMI = weight (kg) / height (m²).

Local examination: Local examination for vitiligo distribution, extent of disease according to VETI score [8], and vitiligo stability as absence of new lesion or progression of existing lesion over last 2 years [9].

Laboratory investigation sampling: Under complete aseptic condition by a standard venipuncture technique, five ml of venousperipheral blood were collected from each participant in plain tubes. At room temperature for 10-20 min, serum was allowed to coagulate and then centrifuged for 20 min at 2,000-3,000 rpm. After supernatant removal, the samples were kept at -20 °C for preservation. Repeated freeze-thaw cycles were avoided.

Measurement of serum endocan level: According to the manufacturers' guidelines, the commercially available specific human ELISA kit was utilized for endocan level assessment.

Assessment of CIMT: B-mode ultrasonography was utilised in conjunction with a linear probe and a Philips Epic 7 ultrasound system operating at a centre frequency of 7.5 to 10 MHz. In the analyses, the mean values of the right and left CIMT readings were utilised. Establishing a CIMT cutoff point of 0.08 cm for atherosclerosis diagnosis.

Ethical considerations: All participants provided informed consents prior to the collection of blood

samples. In accordance with the Helsinki declaration principles, the Local Ethics Committee of the Faculty of Medicine at Benha University approved the entire study design (Approval code: MS 14-4-2022). Personal confidentiality and privacy were upheld throughout the entirety of the research process. Any participant was permitted to withdraw from the study at any time without repercussions. The information gathered was not utilised for any additional intent.

Statistical analysis

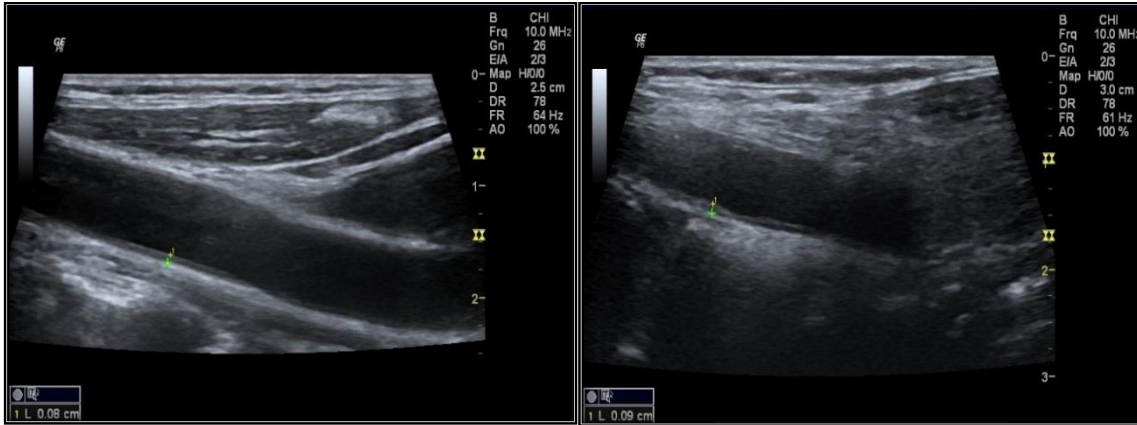
Statistical analysis and data management were conducted utilising SPSS version 28. (IBM, Armonk, New York, United States). The normality of the quantitative data was evaluated utilising the Shapiro-Wilk test, the Kolmogorov–Smirnov test, and direct data visualisation techniques. In accordance with the results of tests for normality, numerical data were condensed into medians and ranges or means and standard deviations. Numerical and percentage summaries were compiled for the categorical data. Comparing quantitative data between study groups required the use of the Mann-Whitney U test for non-normally distributed numerical variables and the independent t-test for normally distributed numerical variables. To compare categorical data, the Chi-square test was applied. Endocan was subjected to ROC analysis in order to predict vitiligo. Calculations included the area under the curve (AUC) with a 95% confidence interval (CI), the optimal cut-off point, and diagnostic indices. Spearman's or Pearson's correlation was employed to establish correlations. Various parameters of Endocan were compared through the utilisation of the Mann-Whitney U test. To forecast vitiligo, a multivariate logistic regression analysis was performed. Probability ratios and 95% CIs were computed. Each statistical test had a dual-sided design. P values ≤ 0.05 were deemed to be statistically significant.

RESULTS

There were no significant differences observed in age (P = 0.191), gender (P = 0.694), or BMI (P = 0.1) between the patient and the control groups (Table 1). 0.08 cm thickness was used as the CIMT cutoff point for atherosclerosis diagnosis. The mean CIMT of patients was significantly higher than that of the control group (Figure 1).

Table (1): General characteristics of patients and control groups

		Patients (n = 60)	Control (n = 20)	Test	P
Age (years)	Mean ±SD	41.9 ±13.2	38.8 ±7.4	t = -1.332	0.191
Gender	Male n (%)	24 (40.0)	9 (45.0)	X ² = 0.155	0.694
	Female n (%)	36 (60.0)	11 (55.0)		
BMI	Mean ±SD	25.8 ±2.6	24.7 ±2.4	t = -1.665	0.1



t: Independent t-test X²: Chi-square test *P < 0.05 is significant
Figure (1): Doppler U/S images of RT and LT carotid arteries of patient no 27. The mean CIMT was 0.085 cm.

DISCUSSION

Numerous systemic and metabolic disorders, including insulin resistance, lipid abnormalities, metabolic syndrome, and atherosclerotic cardiovascular disease, are strongly associated with vitiligo [10].

Important roles are played by oxidative stress and specific proinflammatory markers (homocysteine, TNF-, IL-1, and IL-6) in the pathophysiology of vitiligo and atherosclerosis [11].

Vitiligo has been associated to atherosclerotic cardiovascular disease in a number of studies, and vitiligo patients have a lower cardiovascular risk than controls, according to other research [12, 13].

Vascular endothelial dysfunction is the primary factor that initiates the advancement of atherosclerosis. Endocan has the ability to induce leukocyte migration, increase vascular permeability, and stimulate endothelial cells to produce inflammatory cytokines. These processes are crucial in the pathogenesis of the different phases of atherosclerosis. Endocan, a novel endothelial mediator, controls the proliferation and migration of vascular smooth muscle cells. In the early stages of atherosclerosis, immunohistochemistry identified a significant level of endocan expression. Carotid Doppler ultrasonography is a prevalent and essential modality utilised in the detection of atherosclerosis. Carotid plaque presence or absence and CIMT measurement are useful indicators of atherosclerosis progression and can serve as independent predictors of cardiovascular events [14].

The current study was conducted to evaluate serum endocan level and CIMT by Doppler ultrasonography in sixty patients with vitiligo and compared their levels with twenty healthy volunteers as control group to determine any clinical evidence of subclinical atherosclerosis in absence of other risk factors.

Increased CIMT is considered important surrogate marker of preclinical atherosclerosis [15]. In current study, CIMT of 0.08 cm was used as a cut off point for diagnosis of atherosclerosis. Mean CIMT was

significantly higher in vitiligo patients than in control group ($P < 0.001$). This finding is consistent with the research conducted by **Azzazi et al.** [16] and **Namazi et al.** [13], which demonstrated that individuals with vitiligo had significantly higher mean CIMT and a higher incidence of subclinical atherosclerosis than the control group ($P = 0.001$, $P = 0.002$, respectively).

Increased CIMT in the absence of traditional risk factors, as well as age and sex, is associated with future cardiovascular disease risk, according to a number of studies involving asymptomatic subjects [17]. An additional study documented that a 0.1 mm absolute CIMT increase correlates with a 10 % to 15 % heightened likelihood of future myocardial infarction and a 13 % to 18 % increased likelihood of stroke [18]. Plaque assessment and CIMT screening are likely to detect subclinical atherosclerosis in low-risk individuals [19].

In the present study, CIMT noted significant positive correlation with BMI ($P < 0.001$) implying that participants with high BMI are at high developing subclinical atherosclerosis risk than others. These results are consistent with outcomes reported by other studies [20], which showed that the CIMT was significantly increased with increase in BMI. **Rashid et al.** [21], showed that significant correlation was found between CIMT and BMI in non-randomly selected subjects (>20 years old), and that increase of one BMI unit caused 0.009 increase in CIMT.

Conversely, **Ibinaiye et al.** [22] found a negative correlation between BMI and CIMT among hypertensive patients. Without observing a correlation between BMI and CIMT, **Ge et al.** [23] analysed 526 individuals (mean BMI = 21) who were free of cardiovascular disease.

To the best of our current understanding, no prior research was identified to examine the assessment of serum endocan levels in vitiligo patients. Patients in the current study had a significantly higher median endocan level than the control group.

CONCLUSION

Serum endocan and CIMT levels were significantly correlated with the severity of vitiligo and were found to be highly significant in vitiligo patients relative to the control group. In patients with vitiligo, serum endocan levels were proposed to be independent predictors of cardiovascular risk. Endocan may serve as a potential marker of vitiligo severity, as evidenced by the correlation between elevated serum endocan levels in vitiligo patients and VETI. In addition, the level of serum endocan and CIMT may serve as early indicators of concurrent subclinical atherosclerosis, which elevates the risk of cardiovascular disease in patients with vitiligo.

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