RESEARCH ARTICLE

Is SCUBE1 helpful to predict the arterial thrombotic risk in patients with multiple myeloma: a preliminary study

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Abstract

Background: Signal peptide-CUB-EGF-like domain-containing protein 1 (SCUBE1) is expressed in vascular endothelium and human platelets. SCUBE1 levels are increased in acute arterial thrombosis. Multiple myeloma patients are also at increased risk of arterial thrombotic events. This study aimed to measure SCUBE1 levels in newly diagnosed multiple myeloma patients and to define whether SCUBE1 could be a useful marker determining the arterial thrombotic risk.

Methods: SCUBE1 levels of 32 newly diagnosed multiple myeloma patients and 41 healthy control subjects were measured by enzyme-linked immunosorbent assay.

Results: Plasma SCUBE1 levels of multiple myeloma patients and control subjects were 6.22 ± 0.9 and 7.95 ± 1.1 ng/ml, respectively. In the patient group, SCUBE1 levels were significantly lower compared to control subjects ($p < 0.001$).

Conclusions: SCUBE1 level measuring does not help to predict the arterial thrombotic risk in multiple myeloma patients. The significantly lower levels of SCUBE1 in multiple myeloma patients are likely to be due to defective platelets and/or increased TNF-α cytokines and is another proof of platelet dysfunction seen in these patients. HIPPOKRATIA 2019, 23(1): 21-24.

Keywords: Signal peptide-CUB-EGF-like domain-containing protein 1, SCUBE1, multiple myeloma, arterial thrombosis

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Introduction

Signal peptide-CUB-EGF-like domain-containing protein 1 (SCUBE1) is a protein that plays an important role in inflammation and thrombosis and is expressed in the vascular endothelium[1]. SCUBE1 is also highly expressed in human platelets[2]. It is stored in the α-granules of inactive platelets, which are translocated to the surface upon activation[2]. SCUBE1 has been shown to enhance ristocetin-induced platelet agglutination and adhesion[2].

SCUBE1 has been shown to be elevated in patients with acute coronary syndrome and ischemic stroke[3]. Higher SCUBE1 levels have been demonstrated in an experimental model of acute ischemic stroke and acute mesenteric ischemia[4,5]. This leads to plasma SCUBE1 being considered a potential biomarker of platelet activation in acute thrombotic disease.

In patients with multiple myeloma, the incidence of venous thromboembolic events (VTE) such as deep venous thrombosis of the lower extremities, pulmonary embolism, and catheter-related upper extremity thrombosis has been shown to increase[6]. The risk of VTE is at least 10 % in multiple myeloma patients during their disease history[7]. Furthermore, the risk of arterial thrombotic events, such as acute myocardial infarction and ischemic stroke, increases concurrently in these patients[8]. A specific laboratory test is not available at present to accurately identify the risk of arterial thrombosis. In the current study, we aimed to determine the correlation of SCUBE1 level and the risk of thrombosis in patients with newly diagnosed multiple myeloma.

Patients and methods

Study population

This is a prospective cross-sectional study that enrolled 32 newly diagnosed multiple myeloma patients that attended the Hematology outpatient clinic, of Recep Tayyip Erdoğan University, Training and Research Hospital, in Rize, Turkey during 2014. Multiple myeloma was diagnosed according to the international myeloma working group (IMWG) criteria. The patients were classified according to the Durie-Salmon staging system. The control group consisted of 41 healthy individuals. In both groups, the exclusion criteria included a previous diagnosis of hypertension, coronary artery disease, stroke, can-

cer, and the use of aspirin and/or other antiplatelet drugs.

The study adhered to the tenets of the Helsinki Declaration, and approval was obtained by the local Ethics Committee of the Recep Tayyip Erdoğan University, Medical Faculty (decision No: 2014/124). The patients and controls were required to read and sign an informed consent form.

Peripheral blood samples were taken from all participants into tubes containing Sodium citrate after overnight fasting. The samples were centrifuged within one hour at 1,000 g for 15 min, and the separated plasma was stored at -80ºC.

Laboratory analyses

Plasma levels of SCUBE1 were quantified by enzyme-linked immunosorbent assay (ELISA) using commercially available matched antibodies (Eastbiopharm, Hangzhou, China). The intra-assay and inter-assay coefficients of variation (CV) were 10 % and 12 %, respectively.

Statistical analyses

Statistical evaluations were made using the IBM SPSS Statistics for Windows, Version 21.0 (IBM Corp., Armonk, NY, USA). Constant variables are expressed as mean ± standard deviation. Student's *t*-distribution was employed in the comparison of the two groups. The relation between the variables was studied with Pearson's correlation coefficient. A p-value of less than 0.05 was regarded as statistically significant.

Results

Thirty two newly diagnosed multiple myeloma patients (21 males, 11 females) with a mean age of 73.08 ± 7.01 years, were included in the study. Additionally, 41 healthy subjects (15 males, 26 females) with a mean age of 75.63 ± 5.02 years, were enrolled as the control group (Table 1). In the patient group, the male percentage was higher than that of the control group, and the difference was statistically significant (Table 1). According to the Durie-Salmon staging system, 19 of the patients were in stage two, and 13 of them were in stage three. The platelet levels of the patients were lower than those of the control, and the difference was statistically significant (p =0.002) (Table1). The creatinine levels of the patient group were higher than those of the control group, and the difference was statistically significant (p =0.009) (Table 1). The SCUBE1 levels of both groups are shown in Figure 1, and Figure 2, respectively. The SCUBE1 levels in the multiple myeloma patients (6.22 ± 0.9) were found to be significantly lower than those of the control group (7.95 ± 1.1) (p <0.001) (Figure 3).

Discussion

SCUBE1 is a cell surface protein expressed during early embryogenesis and found in vascular endothelium and human platelets[9]. SCUBE1 is also detected in thrombus and atherosclerotic plaque[10].

Patients with multiple myeloma may face various clinical conditions, such as renal failure, anemia, bone lesions, and infections. Furthermore, these patients also have an increased risk of venous and arterial thrombosis[6]. The risk of venous thromboembolism is highest during the first year following diagnosis[7]. This risk increases further with the use of immunomodulatory drugs, corticosteroids, and chemotherapy[11].

Increased tumor load and active myeloma therapy were deemed responsible for the increased risk of venous thrombosis within the first year[7]. In multiple myeloma patients, high levels of factor VIII, von Willebrand factor, acquired activated protein C resistance, increased microparticle-related tissue factor activity, and fibrinogen level lead to the development of venous thrombosis[6,12]. Also, impaired fibrinolysis in these patients increases the risk of both venous and arterial thrombosis[6].

In patients with multiple myeloma, the incidence of arterial thrombotic complications such as acute myocardial infarction and ischemic stroke increases concurrently with that of venous thromboembolism[7,8]. Likewise, monoclonal gammopathy with undetermined significance (MGUS) increases incidence[13]. An increase in the risk of both arterial and venous thrombosis is suggestive of several mutual biological factors such as platelet activation. This perspective is supported by the use of aspirin for the prevention of venous thrombosis in multiple myeloma patients[14].

SCUBE1 has been studied in a number of malignancies, such as renal cell, gastric, and breast cancers, in which higher levels were reported compared with healthy controls[15-17]. In these studies, increased SCUBE1 levels were attributed to the hypercoagulable state of patients with cancer.

This is the first study to investigate plasma SCUBE1 levels in multiple myeloma patients. In these patients,

Table 1: Demographic characteristics of the multiple myeloma patients and controls who were enrolled in this prospective cross-sectional study aimed to define whether SCUBE1 could be a useful marker to determine arterial thrombotic risk.

Variables		Patients (n =32)	Controls (n =41)	p-value
Age		73.08 ± 7.01	75.63 ± 5.02	0.076
Platelet (10^3/µl)		203.78 ± 71.76	242.49 ± 51.12	0.002
Creatinine (mg/dl)		1.76 ± 1.66	0.91 ± 0.21	0.009
Gender	Male	21 (65.6%)	15 (36.6%)	0.014
	Female	11 (34.4%)	26 (63.4%)	

Data are expressed as mean ± standard deviation or as number of subject (percentages in brackets). A p-value <0.05 is considered statistically significant. n: number of subjects.

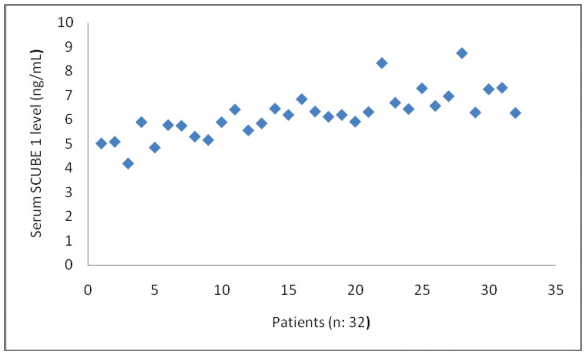

Figure 1: Graph demonstrating the SCUBE1 levels of the 32 newly diagnosed multiple myeloma patients.

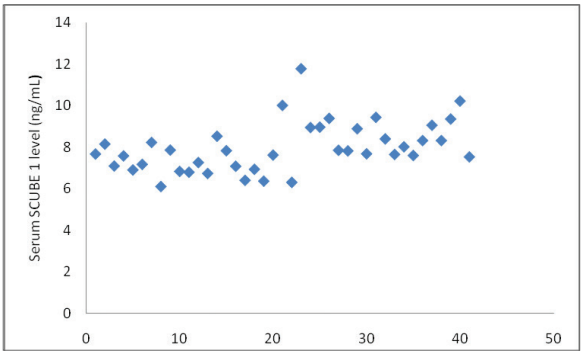

Figure 2: Graph demonstrating the SCUBE1 levels of the 41 healthy control subjects.

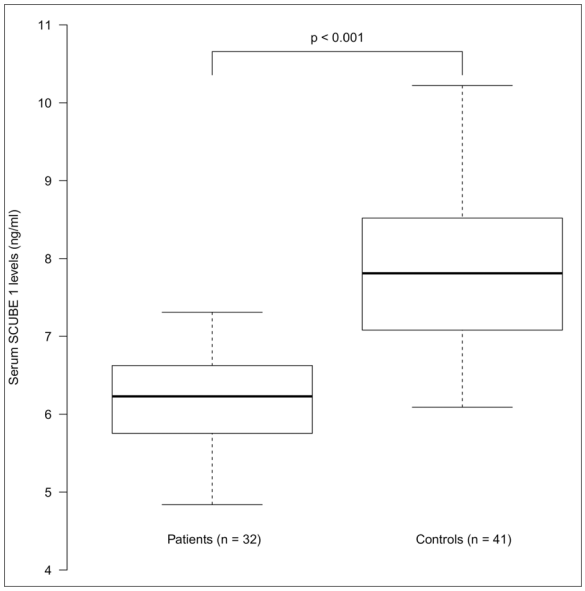

Figure 3: Graph comparing the SCUBE1 levels in the multiple myeloma patients and those of the control group.

SCUBE1 was found to be significantly lower compared to the control group. Dai et al, reported low levels of SCUBE1 in chronic coronary disease, contrary to those in acute coronary syndrome, and acute ischemic stroke[3]. An increase in SCUBE1 level in acute arterial thrombotic events, in which platelet activation and aggregation take place, suggests a role for acute thrombosis. None of the patients or subjects in the control group in our study presented with acute arterial thrombotic events. Since SCUBE1 was lower in patients than controls, the difference could not be attributed to the absence of acute arterial thrombotic events.

SCUBE1 is present in the α-granules of platelets. Patients with multiple myeloma present with platelet disorders such as impaired adhesion and aggregation capacity, reduced platelet factor 3 release, delayed clotting reaction, and abnormalities in platelet metabolism[18]. The coating of platelets with paraproteins was held responsible for these events[18]. Abnormal platelets generated from malignant megakaryocytes may also be another cause. A reduction in the half-life of platelets was also observed in these patients[18]. So, low SCUBE1 levels in multiple myeloma patients may be associated with platelet disorders.

Impaired renal function is known to cause platelet dysfunction[19]. In our study, higher creatinine levels were found in patients compared with the controls. In addition, platelet count was lower in patients than controls. These two findings may be related to low SCUBE1 levels in multiple myeloma patients.

In addition to the α-granules of platelets, SCUBE1 expression in vascular endothelial cells was shown to be reduced following in-vitro tumor necrosis factor-alpha (TNF-α) treatment[1]. As one of the inflammatory cytokines that increase in multiple myeloma is TNF-α, this may account for the lower SCUBE1 levels found in our study.

The limitations of the current study include failure to assess platelet disorders in multiple myeloma patients, the small study population, and the lack of an additional group with MGUS, which could provide more information on SCUBE1 across the myeloma spectrum.

In conclusion, a marker for assessing the risk of future occurrence of venous or arterial thrombotic events in multiple myeloma patients would make the follow-up of these patients easier as it would allow taking suitable prophylactic measures. However, SCUBE1 is not a suitable choice as a marker predicting the risk of arterial thrombosis.

Conflict of interest

The authors declare that there are no conflicts of interest.

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