No Significant Association Between Plasma Endosialin Levels and the Presence or Severity of Coronary Artery Disease

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Abstract

Background: Endosialin, also called tumor endothelial marker-1 or CD248, is a transmembrane glycoprotein that is suggested to play a role in inflammation as well as tumor progression. Endosialin expression was also reported to be upregulated in human atherosclerotic lesions. However, no study has reported blood endosialin levels in patients with coronary artery disease (CAD).

Methods: We investigated the association between plasma endosialin levels and the presence or severity of CAD in 376 patients who underwent elective coronary angiography for suspected CAD. The severity of CAD was represented as the numbers of stenotic coronary vessels and segments.

Results: Of the 376 study patients, CAD was found in 210 patients (one-vessel disease (1-VD), n = 90; two-vessel disease (2-VD), n = 65; and three-vessel disease (3-VD), n = 55). Compared with 166 patients without CAD, 210 patients with CAD had higher C-reactive protein (CRP) levels (median 0.57 vs. 0.43 mg/L, P = 0.007). However, endosialin levels did not significantly differ between patients with and without CAD (0.91 vs. 0.92 ng/mL, P = 0.693). A stepwise increase in CRP levels was found depending on the number of > 50% stenotic vessels: 0.43 in CAD(-), 0.52 in 1-VD, 0.57 in 2-VD, and 0.58 mg/L in 3-VD (P = 0.019). No marked difference was found in endosialin levels among four groups of CAD(-), 1-VD, 2-VD, and 3-VD (0.92, 0.89, 0.98, and 0.87 ng/mL, respectively, P = 0.785). Moreover, no significant correlation was found between endosialin levels and the numbers of > 50% and > 25% stenotic segments or CRP levels. In multivariate analysis, endosialin levels were not a significant factor associated with CAD independent of atherosclerotic risk factors.

Conclusions: Plasma endosialin levels in patients with CAD were found to be not higher than in those without CAD and to be not significantly associated with the presence or severity of CAD.

Keywords: Atherosclerosis; Biomarker; Coronary artery disease; C-reactive protein; Endosialin; Inflammation

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Introduction

Endosialin, also called tumor endothelial marker-1 (TEM-1) or CD248, is a 165-kDa transmembrane glycoprotein that is comprised of a N-terminal extracellular domain, a Sushi domain and three epidermal growth factor (EGF)-like domains and that was initially identified on the blood vessels of tumors and was recognized as an abnormally expressed marker of tumor vascular endothelial cells [1-3]. However, subsequent studies reported that endosialin is not expressed by endothelial cells but by perivascular cells, such as vascular smooth muscle cells, fibroblasts, and CD8 T cells [3-5]. Endosialin expression is also low in normal perivascular cells. During inflammation and tumor progression, endosialin was shown to be upregulated in perivascular cells, adipocytes and some tumor cells [6-8]. In colon cancer, silencing endosialin inhibited the migration and proliferation of tumor-associated fibroblasts [9]. Endosialin-knockout mice showed reduced tumor growth and invasion [10]. This suggests that endosialin plays a role in tumor progression and neovascularization. Regarding the association between endosialin and inflammation, strong expression of endosialin was reported in synovial tissues from patients with rheumatoid arthritis, and endosialin-knockout mice showed less severe arthritis with less leukocyte infiltration and circulating cytokines [11]. These findings thus indicate that endosialin is associated with inflammation, accompanied by a proinflammatory state with increased proinflammatory cytokines [11-13].

Atherosclerotic diseases, such as coronary artery disease

(CAD), are recognized to be a chronic inflammatory disease [14, 15]. Hasanov et al [4] reported that endosialin expression was upregulated in human atherosclerotic lesions and that endosialin-knockout mice showed reduced atherosclerosis. These findings therefore suggest that endosialin may play a role in the pathogenesis of atherosclerosis. Recently, serum endosialin levels in patients with colon cancer were reported to be high [7]. However, no study has reported blood endosialin levels in patients with CAD. Our study was done to elucidate the association between plasma endosialin levels and the presence or severity of CAD in 376 patients undergoing elective coronary angiography.

Materials and Methods

Study patients

In 2008, we started our study to prospectively collect blood samples and clinical and angiographic data from patients undergoing coronary angiography at Tokyo Medical Center. Our study was conducted in compliance with the Helsinki Declaration for research on human beings. Our study was approved by the institutional ethics committee of the hospital (R08-050/R21-037). After written informed consent was obtained, overnight-fasting blood samples were taken on the morning of the day of angiography. A total of 376 consecutive patients who underwent elective coronary angiography for suspected CAD were enrolled in this study to measure plasma endosialin levels. Any patients with acute coronary syndrome (ACS), such as acute myocardial infarction and unstable angina, were excluded from this study. Patients with a history of percutaneous coronary intervention or cardiac surgery, or those with heart failure or aortic diseases were also excluded. Since blood endosialin levels in patients with cancer were reported to be high [8], patients with any cancer were excluded. Hypertension was defined as blood pressures of $\geq 140/90$ mm Hg or on drugs; 223 (59%) patients were taking anti-hypertensive drugs. Hyperlipidemia was defined as a low-density lipoprotein (LDL)-cholesterol level of > 140 mg/dL or on drugs; 148 (39%) patients were taking statin. Diabetes mellitus (DM) (a fasting glucose level of ≥ 126 mg/dL or on treatment) was present in 103 (27%) patients, and 134 (38%) patients were smokers (≥ 10 pack-years).

Measurements of plasma endosialin and C-reactive protein (CRP) levels

Blood samples were collected in ethylenediaminetetraacetic acid (EDTA)-containing tubes and then were centrifuged at 2,000 g for 15 min at 4 °C. The plasma was frozen and stored at -80 °C until analyzed. Plasma endosialin levels were measured by an enzyme-linked immunosorbent assay (ELISA) with a commercially available kit (Human Endosialin ELISA kit, CUSABIO, Wuhan, China). According to the data supplied by the manufacturer, this kit was developed to measure endosialin levels in serum, plasma, tissue homogenates, or cell lysates us-

ing a mouse monoclonal antibody against the extracelluar domain of endosialin and was shown to have no cross-reactivity with analogues, i.e., CD23, CD14 and CD163. The range of values detected by this assay was from 0.31 to 20 ng/mL, and the intra-assay and inter-assay coefficients of variation were < 8% and < 10%, respectively. Plasma high-sensitivity CRP (hsCRP) levels were also measured by a BNII nephelometer (Dade Behring, Tokyo, Japan).

Coronary angiography

Coronary angiograms were recorded on a cineangiogram system (Philips Electronics, Tokyo, Japan). CAD was defined as at least one coronary artery having > 50% luminal diameter stenosis on angiograms. The severity of CAD was represented as the numbers of > 50% stenotic vessels and > 50% and > 25% stenotic segments and the severity score of stenosis. The degree of stenosis in each segment was scored from 0 to 4 points (0, $\leq 25\%$; 1, 26-50%; 2, 51-75%; 3, 76-90%; 4, > 90% stenosis), and then the severity score was defined as the sum of scores of all segments. Coronary artery segments were defined as 29 segments according to the Coronary Artery Surgery Study (CASS) classification. All angiograms were evaluated by a single cardiologist (YM), blinded to the clinical and laboratory data.

Statistical analysis

Any differences between two groups were evaluated by unpaired t-test for parametric variables, by Mann-Whitney U test for nonparametric variables, and by Chi-squared test for categorical variables. Differences among ≥ 3 groups were evaluated by an analysis of variance with Scheffe's test for parametric variables, by Kruskal-Wallis test for nonparametric variables, and by Chi-squared test for categorical variables. Since the distributions of the measured endosialin and hsCRP levels were considered to be highly skewed and to be nonparametric variables by Shapiro-Wilk test, these results were presented as the median value. The correlations between endosialin levels and hemoglobin A1c (HbA1c) or hsCRP levels or the severity of CAD were evaluated by the Spearman's rank correlation test. A multiple logistic regression analysis was performed to determine the independent association between endosialin levels and CAD. All statistical analyses were performed using the SPSS software package (IBM SPSS ver. 25, Tokyo, Japan). A P value of < 0.05 was considered to be statistically significant. Results are presented as the mean ± standard deviation (SD) or the median value and interquartile range.

Results

Of the 376 study patients, CAD was found in 210 patients (56%) (one-vessel disease (1-VD), n = 90; two-vessel disease (2-VD), n = 65; three-vessel disease (3-VD), n = 55). Compared with 166 patients without CAD, 210 patients with CAD

Table 1. Clinical Characteristics of Patients With and Without CAD

	CAD(-) (n = 166)	P value CAD(-) vs. CAD	CAD (n = 210)	1-VD (n = 90)	2-VD (n = 65)	3-VD (n = 55)	P value among four groups
Age (years)	66 ± 11	0.005	69 ± 10	68 ± 10	70 ± 10	71 ± 9	0.008
Gender (male)	99 (60%)	0.001	159 (76%)	68 (76%)	48 (74%)	43 (78%)	0.010
BMI (kg/m^2)	25.3 ± 13.9	0.199	24.0 ± 3.4	24.3 ± 3.7	24.6 ± 2.9	23.1 ± 3.5	0.530
Hypertension	100 (60%)	< 0.001	165 (79%)	68 (76%)	50 (77%)	47 (85%)	0.001
SBP (mm Hg)	129 ± 21	0.012	134 ± 19	133 ± 17	137 ± 21	134 ± 18	0.041
Diabetes mellitus	29 (17%)	< 0.001	74 (35%)	26 (29%)	27 (42%)	21 (38%)	< 0.001
HbA1c (%)	6.0 ± 0.7	< 0.001	6.3 ± 1.0	6.1 ± 0.8	6.4 ± 1.0	6.3 ± 1.0	0.001
Smoking	50 (30%)	0.051	84 (40%)	38 (42%)	27 (42%)	19 (35%)	0.178
Hyperlipidemia	71 (43%)	< 0.001	130 (62%)	56 (62%)	41 (63%)	33 (60%)	0.003
Statin	49 (30%)	0.001	99 (47%)	45 (50%)	30 (46%)	24 (44%)	0.005
LDL-C (mg/dL)	112 ± 27	0.403	115 ± 33	110 ± 34	115 ± 31	121 ± 32	0.151
HDL-C (mg/dL)	59 ± 16	< 0.001	52 ± 13	54 ± 14	51 ± 12	49 ± 14	< 0.001
hsCRP (mg/L)	0.43 (0.22, 0.92)	0.007	0.57 (0.30, 1.19)	0.52 (0.28, 0.95)	0.57 (0.35, 1.11)	0.58 (0.35, 1.48)	0.019

Data represent the mean ± SD or the number (%) of patients, except for hsCRP levels which are presented as the median value and interquartile range. CAD: coronary artery disease; 1-VD: one-vessel disease; 2-VD: two-vessel disease; 3-VD: three-vessel disease; BMI: body mass index; SBP: systolic blood pressure; HbA1c: hemoglobin A1c; LDL-C: low-density lipoprotein cholesterol; HDL-C: high-density lipoprotein cholesterol; CRP: C-reactive protein; hsCRP: high-sensitivity CRP; SD: standard deviation.

were older and had a male predominance; higher prevalence of hypertension, hyperlipidemia and DM; and lower high-density lipoprotein (HDL)-cholesterol levels (Table 1). Plasma hsCRP levels were higher in patients with CAD than in those without CAD (median 0.57 vs. 0.43 mg/L, P = 0.007). A stepwise increase in hsCRP levels was found depending on the number of > 50% stenotic coronary vessels: 0.43 in CAD(-), 0.52 in 1-VD, 0.57 in 2-VD, and 0.58 mg/L in 3-VD (P = 0.019) (Table 1). Moreover, hsCRP levels significantly correlated with the numbers of stenotic segments and the severity score (Table 2).

Plasma endosialin levels did not significantly differ between patients with and without CAD (median 0.91 vs. 0.92 ng/mL, P = 0.693) (Fig. 1). Moreover, no marked difference was found in endosialin levels among the four groups of CAD(-), 1-VD, 2-VD, and 3-VD (0.92, 0.89, 0.98, and 0.87

Table 2. Correlations Between Plasma Endosialin or hsCRP Levels and the Severity of CAD

	rs	P value
Plasma endosialin levels		
The number of > 50% stenotic coronary segments	0.02	0.700
The number of > 25% stenotic coronary segments	0.02	0.752
The severity score of stenosis	0.02	0.782
BMI	-0.03	0.634
Fasting glucose level	-0.04	0.432
HbA1c level	0.12	0.023
LDL-cholesterol level	0.01	0.827
HDL-cholesterol level	-0.04	0.412
hsCRP level	0.08	0.103
Plasma hsCRP levels		
The number of > 50% stenotic coronary segments	0.15	0.003
The number of > 25% stenotic coronary segments	0.16	0.003
The severity score of stenosis	0.15	0.003

rs indicates correlation coefficient by the Spearman's rank correlation test. CRP: C-reactive protein; hsCRP: high-sensitivity CRP; CAD: coronary artery disease; BMI: body mass index; HbA1c: hemoglobin A1c; LDL: low-density lipoprotein; HDL: high-density lipoprotein.
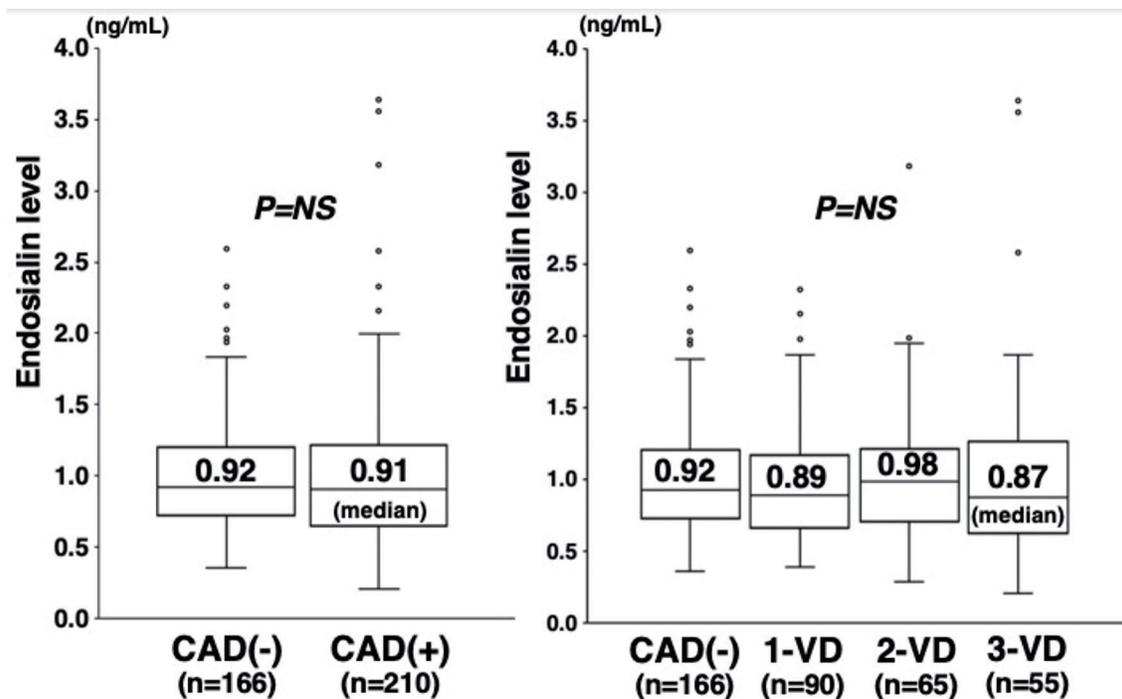
Figure 1. Plasma endosialin levels and the presence of CAD or the number of stenotic coronary vessels. Plasma endosialin levels did not significantly differ between patients with and without CAD (0.91 vs. 0.92 ng/mL) (left). No significant difference was found in endosialin levels among the four groups of CAD(-), 1-VD, 2-VD, and 3-VD (0.92, 0.89, 0.98, and 0.87 ng/mL) (right). The central line represents the median, and the box represents the 25th to 75th percentiles. The whiskers represent the lowest and highest value in the 25th percentile minus 1.5 IQR and 75th percentile plus 1.5 IQR, respectively. NS: not significant; CAD: coronary artery disease; 1-VD: one-vessel disease; 2-VD: two-vessel disease; 3-VD: three-vessel disease; IQR: interquartile range.

ng/mL, respectively, P = 0.785) (Fig. 1). No significant correlation was found between endosialin levels and the numbers of stenotic segments or the severity score (Table 2). Moreover, no correlation was found between endosialin levels and LDL-cholesterol or hsCRP levels. However, endosialin levels significantly but weakly correlated with HbA1c levels (rs = 0.12, P = 0.023) (Fig. 2). In a multivariate analysis, endosialin levels were found to be not a significant factor associated with CAD independent of atherosclerotic risk factors (Table 3).

Discussion

In the present study, plasma endosialin levels in patients with CAD were found to be not significantly higher than in those without CAD and did not significantly correlate with the severity of CAD. In contrast, hsCRP levels in patients with CAD were high and correlated with the severity of CAD. No significant correlation was found between endosialin and hsCRP levels.

Endosialin was reported to be upregulated in synovial tissues from patients with rheumatoid arthritis, and endosialin-knockout mice showed the amelioration of arthritis [11], thus suggesting a role of endosialin in inflammatory diseases. Endosialin upregulation was also reported in fibrotic diseases [16, 17]. Endosialin was upregulated in the fibrotic areas of lung samples from patients with pulmonary fibrosis [16]. Endosialin expression also increased in fibrotic liver of patients with liver cirrhosis, and endosialin-knockout mice showed less liver fibrosis in response to injury [17]. These findings thus suggest that endosialin may be involved in the progression of fibrosis. Fibrosis associated with inflammation is recognized to play a crucial role in atherosclerosis [14, 15, 18]. Vascular fibrosis involves smooth muscle cell proliferation and extracellular matrix accumulation, thereby leading to arterial wall thickening and stenosis [18]. Endosialin was reported to be abundantly expressed in human atherosclerotic lesions, and endosialin-knockout mice had less atherosclerosis with less macrophage recruitment [4]. Thus, endosialin may play a role in the progression of atherosclerosis associated with inflammation and fibrosis. Furthermore, endosialin-knockout mice were reported to be protected against arterial thrombosis [6]. An immunofluorescence analysis also showed increased endosialin expression and infiltrating CD8 T cells in aortic aneurysm specimens, suggesting a role of endosialin in vascular remodeling [3].

Endosialin expression in adipose tissues was shown to increase with obesity [13]. Endosialin-knockout mice showed less inflammation with improvement in insulin sensitivity and glucose tolerance [13]. Furthermore, endosialin-knockout mice had a reduction in fat pad size and serum cholesterol levels [19]. These findings thus implicate endosialin in obesity, glucose tolerance and lipid metabolism, and these effects of endosialin may affect the progression of atherosclerosis. In our study, plasma endosialin levels did not correlate with body mass index (BMI) or LDL-cholesterol levels. However, endosialin levels significantly but weakly correlated with HbA1c levels, suggesting that endosialin may affect glucose tolerance.
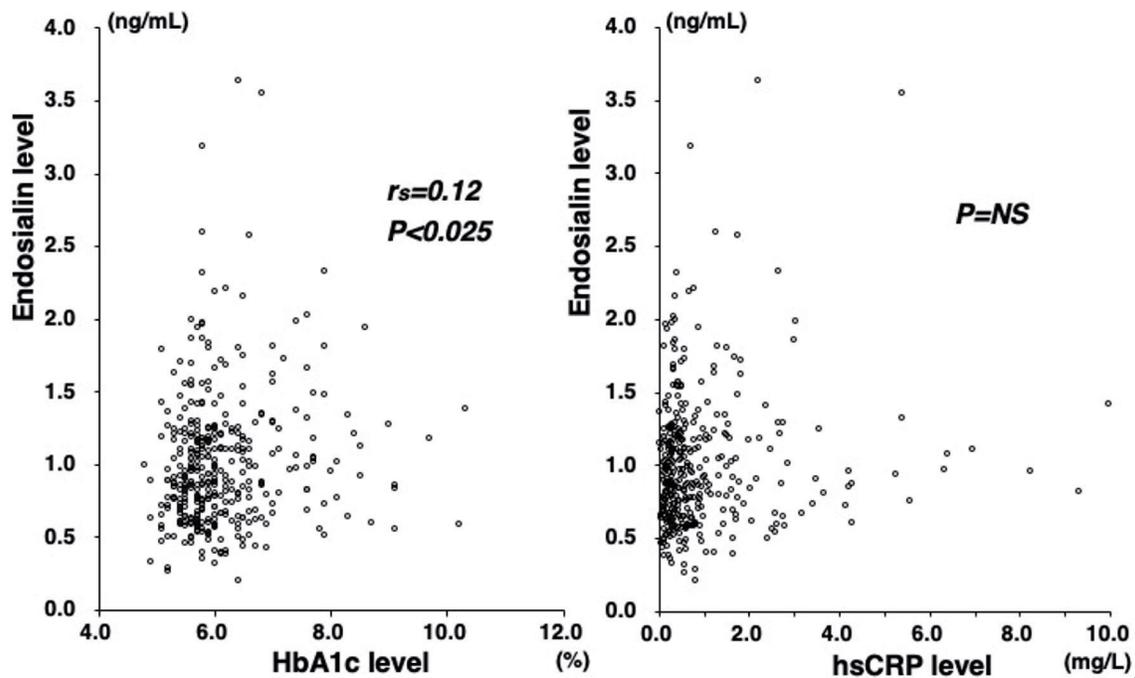
Figure 2. Correlations between plasma endosialin levels and HbA1c or hsCRP levels. Plasma endosialin levels signficantly but weakly correlated with HbA1c levels (rs = 0.12, P < 0.025) (left). However, no significant correlation was found between endosialin and hsCRP levels (right). HbA1c: hemoglobin A1c; hsCRP: high-sensitivity CRP; NS: not significant.

Using the originally developed anti-endosialin monoclonal antibody, O'Shannessy et al. [20] measured serum endosialin levels in 117 patients with colorectal cancer and 100 healthy subjects and reported no difference in endosialin levels between the two groups. In contrast, Pietrzyk et al [7] recently measured serum endosialin levels by ELISA with a commercially available kit in 45 patients with colorectal cancer and 35 healthy subjects and reported endosialin levels to be higher in patients with colorectal cancer than in healthy subjects. Endosialin is recognized to be a soluble protein that can be cleaved and detected in blood [20]. In blood, endosialin was shown to be present as 120 - 150 kDa molecules representing glycosylated extracellular domain fragments [20]. However, no study has shown blood endosialin levels in patients with CAD.

Table 3. Factors Associated With CAD (Multiple Logistic Regression Analysis of the 376 Study Patients)

	Odds ratio (95% CI)	P value
Age (per 10-year increase)	1.35 (1.08 - 1.69)	0.010
Male gender	2.39 (1.43 - 3.99)	0.001
Hypertension	1.75 (1.06 - 2.90)	0.029
Hyperlipidemia	2.17 (1.35 - 3.50)	0.001
Low HDL-cholesterol level	2.09 (1.01 - 4.34)	0.047

The dependent variable was the presence of CAD. The analysis included age (per 10-year increase), male gender, hypertension, hyperlipidemia, statin use, low HDL-cholesterol level (< 40 mg/dL), DM, smoking, and hsCRP and endosialin levels. DM: diabetes mellitus; CAD: coronary artery disease; HDL: high-density lipoprotein; CI: confidence interval.

Since endosialin is suggested to play a role in the progression of atherosclerosis [3, 4, 6], we hypothesized that plasma endosialin levels in patients with CAD would be high. Our present study is the first to report plasma endosialin levels in patients with CAD. Unfortunately, endosialin levels did not significantly differ between patients with and without CAD and did not correlate with the severity of CAD. In a multivariate analysis, endosialin levels were not a significant factor for CAD. As previously reported [21], hsCRP levels were higher in patients with CAD than in those without CAD and correlated with the severity of CAD. Therefore, plasma endosialin levels were not significantly associated with the presence or severity of CAD, and they are unlikely to be a better biomarker for CAD than hsCRP levels. Our results suggest that plasma endosialin levels do not closely reflect the degree of coronary atherosclerosis and inflammation and that endosialin is unlikely to play a major role in the progression of coronary atherosclerosis.

Our study has several limitations. First, angiography was used to assess coronary atherosclerosis. It cannot visualize plaques and only shows the lumen characteristics. Second, our study was cross-sectional in nature and was unable to establish causality, since it only depicted some associations. Third, no patients with ACS, who usually need emergent angiography, were included in our study. Further study in patients with ACS is needed to clarify the potential role of endosialin in this syndrome. Fourth, we did not measure endosialin levels in the coronary sinus. Endosialin levels at this site may be higher than in peripheral blood. Fifth, as we previously reported that plasma hsCRP levels were more closely associated with aortic atherosclerosis than coronary atherosclerosis [21], endosialin levels in patients with CAD may more closely reflect atherosclerosis

in other vascular beds or inflammation in non-vascular tissues than coronary atherosclerosis. Finally, we compared patients with and without CAD and had no healthy controls. Even patients without CAD may have had some atherosclerosis in other vascular beds, as well as in the coronary arteries. Pietrzyk et al [7] measured serum endosialin levels using a kit (Human Endosialin ELISA kit, MyBiosource, USA) in patients with cancer vs. healthy controls (1.31 vs. 0.92 ng/mL). The kit used in their study was different from that in our study (Human Endosialin ELISA kit, CUSABIO, China), but the detection limit was similar (0.31 to 20 ng/mL). In spite of different ELISA kits, their reported levels of healthy controls seem to be similar to those of our patients without CAD (0.92 ng/mL). However, our study was in Japanese patients undergoing angiography, who are generally considered to be a highly select population at high risk for CAD. Our results may not be applicable to the general or other ethnic populations.

Conclusions

Plasma endosialin levels in patients with CAD were found to be not higher than in those without CAD and were not associated with CAD. Our results suggest that plasma endosialin levels cannot be a good biomarker reflecting the presence or severity of CAD and that endosialin is unlikely to play a major role in the progression of coronary atherosclerosis.

Acknowledgments

Financial Disclosure

Conflict of Interest

The authors have no conflict of interest to declare.

Informed Consent

Written informed consent was obtained from all the study patients.

Author Contributions

YK, RO and YM conceived and designed the study. MA, and ES acquired the data. YK and YM analyzed and interpreted the data and drafted the paper. RO, KT and KK critically reviewed and supervised the manuscript. All authors read and approved the final manuscript.

Data Availability

The authors declare that data supporting the findings of this study are available within the article.

Abbreviations

CAD: coronary artery disease; ACS: acute coronary syndrome; DM: diabetes mellitus; CRP: C-reactive protein; hsCRP: high-sensitivity CRP; ELISA: enzyme-linked immunosorbent assay; CASS: Coronary Artery Surgery Study; BMI: body mass index; SBP: systolic blood pressure; LDL-C: low-density lipoprotein cholesterol; HDL-C: high-density lipoprotein cholesterol

References

1. St Croix B, Rago C, Velculescu V, Traverso G, Romans KE, Montgomery E, Lal A, et al. Genes expressed in human tumor endothelium. Science. 2000;289(5482):1197-1202.
2. Rettig WJ, Garin-Chesa P, Healey JH, Su SL, Jaffe EA, Old LJ. Identification of endosialin, a cell surface glycoprotein of vascular endothelial cells in human cancer. Proc Natl Acad Sci U S A. 1992;89(22):10832-10836.
3. Hu X, Wu T, Wang C, Li J, Ying C. CD248+CD8+ T lymphocytes suppress pathological vascular remodeling in human thoracic aortic aneurysms. Exp Biol Med (Maywood). 2021;246(2):121-129.
4. Hasanov Z, Ruckdeschel T, Konig C, Mogler C, Kapel SS, Korn C, Spegg C, et al. Endosialin Promotes Atherosclerosis Through Phenotypic Remodeling of Vascular Smooth Muscle Cells. Arterioscler Thromb Vasc Biol. 2017;37(3):495-505.
5. MacFadyen JR, Haworth O, Roberston D, Hardie D, Webster MT, Morris HR, Panico M, et al. Endosialin (TEM1, CD248) is a marker of stromal fibroblasts and is not selectively expressed on tumour endothelium. FEBS Lett. 2005;579(12):2569-2575.
6. Kapopara PR, Safikhan NS, Huang JL, Meixner SC, Gonzalez K, Loghmani H, Ruf W, et al. CD248 enhances tissue factor procoagulant function, promoting arterial and venous thrombosis in mouse models. J Thromb Haemost. 2021;19(8):1932-1947.
7. Pietrzyk L, Wdowiak P. Endosialin (TEM1) as a Diagnostic, Progression, and Prognostic Serum Marker for Patients With Colorectal Cancer-A Preliminary Study. Cancer Control. 2020;27(1):1073274820903351.
8. Khan KA, McMurray JL, Mohammed F, Bicknell R. C-type lectin domain group 14 proteins in vascular biology, cancer and inflammation. FEBS J. 2019;286(17):3299-

    3332.
9. Christian S, Winkler R, Helfrich I, Boos AM, Besemfelder E, Schadendorf D, Augustin HG. Endosialin (Tem1) is a marker of tumor-associated myofibroblasts and tumor vessel-associated mural cells. Am J Pathol. 2008;172(2):486-494.
10. Tomkowicz B, Rybinski K, Foley B, Ebel W, Kline B, Routhier E, Sass P, et al. Interaction of endosialin/TEM1 with extracellular matrix proteins mediates cell adhesion and migration. Proc Natl Acad Sci U S A. 2007;104(46):17965-17970.
11. Maia M, de Vriese A, Janssens T, Moons M, van Landuyt K, Tavernier J, Lories RJ, et al. CD248 and its cytoplasmic domain: a therapeutic target for arthritis. Arthritis Rheum. 2010;62(12):3595-3606.
12. Teicher BA. CD248: A therapeutic target in cancer and fibrotic diseases. Oncotarget. 2019;10(9):993-1009.
13. Petrus P, Fernandez TL, Kwon MM, Huang JL, Lei V, Safikhan NS, Karunakaran S, et al. Specific loss of adipocyte CD248 improves metabolic health via reduced white adipose tissue hypoxia, fibrosis and inflammation. EBioMedicine. 2019;44:489-501.
14. Ross R. Atherosclerosis - an inflammatory disease. N Engl J Med. 1999;340(2):115-126.
15. Wu MY, Li CJ, Hou MF, Chu PY. New insights into the role of inflammation in the pathogenesis of atherosclerosis. Int J Mol Sci. 2017;18(10):2034.
16. Bartis D, Crowley LE, D'Souza VK, Borthwick L, Fisher AJ, Croft AP, Pongracz JE, et al. Role of CD248 as a potential severity marker in idiopathic pulmonary fibrosis. BMC Pulm Med. 2016;16(1):51.
17. Wilhelm A, Aldridge V, Haldar D, Naylor AJ, Weston CJ, Hedegaard D, Garg A, et al. CD248/endosialin critically regulates hepatic stellate cell proliferation during chronic liver injury via a PDGF-regulated mechanism. Gut. 2016;65(7):1175-1185.
18. Lan TH, Huang XQ, Tan HM. Vascular fibrosis in atherosclerosis. Cardiovasc Pathol. 2013;22(5):401-407.
19. Armitage EG, Barnes A, Patrick K, Bechar J, Harrison MJ, Lavery GG, Rainger GE, et al. Metabolic consequences for mice lacking Endosialin: LC-MS/MS-based metabolic phenotyping of serum from C56Bl/6J Control and CD248 knock-out mice. Metabolomics. 2021;17(2):14.
20. O'Shannessy DJ, Smith MF, Somers EB, Jackson SM, Albone E, Tomkowicz B, Cheng X, et al. Novel antibody probes for the characterization of endosialin/TEM-1. Oncotarget. 2016;7(43):69420-69435.
21. Momiyama Y, Ohmori R, Fayad ZA, Kihara T, Tanaka N, Kato R, Taniguchi H, et al. Associations between plasma C-reactive protein levels and the severities of coronary and aortic atherosclerosis. J Atheroscler Thromb. 2010;17(5):460-467.