Predictive Value of Circulating VE-Catherin in Asymptomatic Coronary Artery Disease Patients with Controlled Lymphoproliferative Diseases

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Abstract

Aim: To evaluate the prognostic value of circulating VE-catherin for cumulative survival in asymptomatic coronary artery disease patients with full or partial remission of lymphoproliferative diseases.

Methods: One hundred eighty two out subjects with full or partial remission of lymphoproliferative diseases (43 patients with multiple myeloma, 101 subjects with chronic lymphoid leukemia, and 10 patients with Hodgkin's and non-Hodgkin's lymphoma. Observation period was up to 3 years. Blood samples for biomarkers measurements were collected. ELISA method for measurements of circulating level of VE-catherin was used. Concentrations of VE-catherin for cumulative survival cases due to advanced CHF were tested.

Results: One hundred ninety six cumulative clinical events occurred in 42 patients (23.1%) within the follow-up, with their distribution being as follows: 29 deaths, 65 cardiac arrhythmias, 17 cardiac ischemic events, 9 strokes, 37 chronic heart

failures and 39 hospital admissions for cardiovascular reasons. Medians of circulating levels of VE-catherin in free-events subject cohort and subjects cohort with cardiovascular events were 0.45 ng/ml (95% confidence interval [CI] = 0.34-0.66 ng/ml) and 1.44 ng/ml (95% CI = 0.88-1.63 ng/ml) ($P < 0.001$). In multivariate logistic regression circulating VE-catherin independently predicted cumulative cardiovascular events (odds ratio [OR] = 1.09; 95% CI = 1.02–1.12; $P = 0.001$) within 3 years of observation period. However, NT-pro-BNP and E/Em remained statistically significant predictors for cumulative cardiovascular events (OR = 1.06; 95% CI 1.03–1.14; $P < 0.001$ and OR = 1.05; 95% CI = 1.01 – 1.09; $P < 0.001$), whereas T2DM, hypertension, obesity, LVEF, and multi-vessel lesion did not.

Conclusion: among patients with documented lymphoproliferative diseases and known asymptomatic coronary artery disease increased circulating VE-catherin associates with increased 3-year cumulative cardiovascular events.

Keywords: VE-catherin; coronary artery disease; lymphoproliferative disease; survival; prognosis

Introduction

Endothelial damage and perivascular leukocyte infiltrates are vital in the development of lymphoma and multiple myeloma [7]. Recent studies have found that endothelial dysfunction might be result in lymphoproliferative diseases progression and adverse effects of drug implementation, such as bendamustin, kladribin [5]. On the other hand, there is a direct correlation between microvessel density in lymphoproliferative diseases and parameters of disease progression [8]. Endothelial cells participate in inflammatory events leading to atherogenesis by regulating endothelial cell permeability via the expression Vascular endothelial-cadherin (VE-Cadherin) on their surface [12]. VE-cadherin is cell adhesion molecules localized at the endothelial junction, which plays critical roles in angiogenesis, neovascularization, neoplasm development, stem cells mobbing, and endothelial integrity [4]. Indeed, VE-cadherin chiefly organizes the opening and closing of the endothelial barrier [2]. It has found that VE-cadherin as a transmembrane protein probably modulates intensity of angiogenesis in lymphoproliferative diseases and may be useful in prognosis of response to treatment [11]. However, the predictive role of VE-cadherin as a marker of cardiovascular events in patients in full or partial remission of lymphoproliferative diseases is not still clear. **The aim** of the study to evaluate the prognostic value of circulating VE-catherin for cumulative survival in asymptomatic coronary artery disease patients with full or partial remission of lymphoproliferative diseases.

Design and Methods

One hundred eighty two out subjects with full or partial remission of lymphoproliferative diseases (43 patients with multiple myeloma, 101 subjects with chronic lymphoid leukemia, and 10 patients with Hodgkin's and non-Hodgkin's lymphoma) who underwent cardiac computer tomography (CT) angiography and documented asymptomatic coronary artery disease (CAD) were enrolled in the study. All subjects gave their written informed consent to participation in the study. Diagnosis and staging of lymphoproliferative diseases were defined by current clinical practice guidelines [10, 13]. To be achieving remission chemotherapy with FC, R-FC, R-CHOP, CHOP, mini-CHOP, COP, ABVD, VTD, CTD, TD, bendamustine, cladribine was used accordingly contemporary clinical guidelines. All subjects were at full or partial remission stage at baseline.

Echocardiography in B-mode was performed accordingly to Recommendation of American Society of Echocardiography on the scanner "MyLab 50" (Italy) using a transducer with a frequency of 2.5-3.5 MHz. End-diastolic and end-systolic LV volumes were obtained using a two-dimensional reference sector according to Simpson's method, and LV ejection fraction (LVEF) was calculated by accordingly conventional methods [3].

Coronary vessel-wall, and plaque geometrical, and compositional parameters were measured on contrast-enhanced spiral computer tomography (CT) angiography [1]. Contrast-enhanced spiral CT was performed on the "OPTIMA CT 660" scanner (GE Medical Systems, and Milwaukee, USA) with 2 rows of detectors (32 × 2 CT system) during the end-expiratory breath-hold. After noncontrast localization image acquisition, injection of nonionic contrast "Omnipak" (Amersham Health, Ireland) was used to determine the optimal coronary arterial image. Standardized calcium scores were obtained with beam energy of 120 kV, full rotation time of 350 milliseconds, and tube current of 300 mA. The images were reconstructed in 0.6-mm axial slices. Scans were electrocardiogram-gated and were triggered at either 40% or 75% phase contingent on heart rate.

All blood samples were collected after fasting in cooling vacutainer and after that it was immediately centrifugated (4°C for 6.000 × 15 min). After centrifugation serum was blind coded and stored at -70° until used. VE-cadherin levels were measured by ELISA technique. Human VE-cadherin Quantikine ELISA Kit (R&G, United Kingdom) was used for examination. All determinations were done by duplicating. The mean intra-assay coefficients of variation were <10% of all cases.

Fasting plasma glucose (FPG) was quantified by the glucose oxidase procedure; HbA1c was measured by ion-exchange high-performance liquid chromatography (HPLC; Bio-Rad, Hercules, CA, USA).

Concentrations of total cholesterol (TC) and high density lipoprotein (HDL) cholesterol were determined by a Dimension Clinical Chemistry System (Dade Behring Inc, Newark, NJ). Low density lipoprotein (LDL) cholesterol was calculated by using the formula of Friedewald W.T., Levy R.I., Fredrickson D.S. (1972). All measurements and blood sample for VE-cadherin, glucose, creatinin, glycated hemoglobin (HbA1c), TC, LDL-C, HDL-C were collected at the same visit.

Clinical Events: Screening and Diagnostics

Clinical interviews were carried out every month for three years after baseline. The following are the clinical events verified: newly diagnosed strokes or TIAs; death for any reasons and sudden cardiac death; coronary ischemic events (myocardial infarction, unstable angina) that needed hospital admission for cardiovascular reasons, new-onset chronic heart failure. Newly diagnosed strokes were confirmed with CT. All clinical events were presented as cumulative.

Statistical Analysis

All statistical analyses were performed in SPSS for Windows v. 17.0 (SPSS Inc., Chicago, IL, USA). All values were given as mean and 95% CI or median and percentiles. An independent group t-test was used for comparisons for all interval parameters meeting the criteria of normality and homogeneity of variance. For interval parameters not meeting these criteria, the non-parametric Mann-Whitney test was used to make comparisons between the groups. Comparisons of categorical variables between the groups were performed using the Chi2 test, and the Fisher exact test. VE-cadherin concentration was normally distributed and it was not positively skewed. However, the data were not transformed. The potential factors (age; sex, smoking, systolic and diastolic BP, VE-cadherin, fasting plasma glucose, hypercholesterolemia, TC, LDL cholesterol, creatinin level, HbA1c) that may be associated with cardiovascular events was identified first by the univariate analysis (ANOVA), then multivariate logistic regression analyses were used to identify the predict factors. A calculated difference of $P < 0.05$ was considered significant.

Results

One hundred ninety six cumulative clinical events occurred in 42 patients (23.1%) within the follow-up, with their distribution being as follows: 29 deaths, 65 cardiac arrhythmias, 17 cardiac ischemic events, 9 strokes (5 lacunar infarctions and 2 cardioembolic strokes), 37 chronic heart failures and 39 hospital admissions for cardiovascular reasons.

General characteristics of study patients are presented in table 1. Significant differences between the both cohorts of patients for demographics (age, sex), conventional risk factor (smoking, arterial hypertension, dyslipidemia, T2DM, BMI, obesity and overweight), biochemical (creatinine, total cholesterol, LDL cholesterol, and HDL cholesterol, fasting blood glucose) and some hemodynamic parameters (mean systolic and diastolic BP, heart rate, LV EF, E/Am, and E/Em) were not found. On the other hand, there was significant difference between both cohorts in incidences of heart failure ($P < 0.001$). Therefore, circulating level of NT-pro-BNP was much higher in free-events subject cohort when compared with subjects cohort with cardiovascular events occurred.

All hypertensive patients were treated according to current clinical guidelines with diet, lifestyle modification and drug therapy that included ACE inhibitors / ARBs, aspirin or other antiagregants, statins. Metformin was prescribed in 3 (2.1%) and 5 (11.9) T2DM patients in both cohorts, in other cases diet and life-style modification were recommended only. Because incidences of heart failure was seen significantly often in subjects cohort with cardiovascular events in comparison with free-events subject cohort, however, ACEI / ARAs, mineralcorticoid receptors antagonists, and diuretics have been used increasingly.

Medians of circulating levels of VE-catherin in free-events subject cohort and subjects cohort with cardiovascular events were 0.45 ng/ml (95% confidence interval [CI] = 0.34-0.66 ng/ml) and 1.44 ng/ml (95% CI = 0.88-1.63 ng/ml) ($P < 0.001$). The data suggested that VE-catherin plasma levels were directly related to NT-pro-BNP ($r = 0.54$, $P = 0.001$), T2DM ($r = 0.42$, $P = 0.006$), gender ($r = 0.40$, $P < 0.001$ for male), E/Am ($r = 0.39$, $P = 0.001$), E/Em ($r = 0.38$, $P = 0.001$), multi-vessel lesion of coronary arteries ($r = 0.32$, $P = 0.001$), TC ($r = 0.32$, $P = 0.001$), age ($r = 0.28$, $P = 0.001$), smoking ($r = 0.26$, $P = 0.001$) and inversely to LVEF ($r = -0.36$, $P = 0.001$) and eGFR ($r = -0.34$, $P = 0.001$). No significant association between the levels of circulating VE-catherin with fasting plasma glucose, HbA1c, mean systolic and diastolic BP, and medications for both cohorts of the patients was found.

Univariate and multivariate logistic regressions were used to assess whether biomarker was able to better discriminate between two patient cohorts. In the univariate logistic regression analysis, the main factors independently related with cumulative cardiovascular events were VE-catherin, NT-pro-BNP, E/Em, LVEF, T2DM, hypertension, obesity and multi-vessel lesion of coronary artery. In multivariate logistic regression circulating VE-catherin independently predicted cumulative cardiovascular events (odds ratio [OR] = 1.09; 95% CI = 1.02–1.12; $P = 0.001$) within 3 years of observation period. However, NT-pro-BNP and E/Em remained statistically significant predictors for cumulative cardiovascular events (OR = 1.06; 95% CI 1.03–1.14; $P < 0.001$ and OR = 1.05; 95% CI = 1.01 – 1.09; $P < 0.001$), whereas T2DM, hypertension, obesity, LVEF, and multi-vessel lesion did not.

Table 1 General characteristic of patients participating in the study

Variables	Free-events subjects (n=140)	Subjects with cardiovascular events (n=42)	P-value
Age, years	59±11.47	61±9.73	0.56
Males, n (%)	73 (52.1)	29 (69.0)	0.21
Arterial hypertension, n (%)	15 (10.7)	5 (11.9)	0.28
Dyslipidemia, n (%)	28 (20.0)	11 (26.2)	0.36
T2DM, n (%)	17 (12.1)	5 (11.9)	0.56
BMI, kg/m ²	26.5 (95% CI=25.95-27.05)	28.2 (95% CI=27.02-28.38)	0.42
Obesity, n (%)	32 (23.6)	10 (23.8)	0.82
Overweight, n (%)	20 (14.3)	5 (11.9)	0.52
Adherence to smoking, n (%)	3 (2.1)	8 (1.9)	0.44
Heart failure, n (%)	9 (6.4)	18 (42.9)	<0.001
GFR, mL/min/1.73 m ²	103.9±19.4	105.5±14.4	0.48
HbA1c, %	5.22±0.7	5.42±0.83	0.58
Fasting blood glucose, mmol/L	4.63±0.57	4.71±0.67	0.69
Creatinine, µmol/L	68.88±13.78	73.02±12.96	0.72
Total cholesterol, mmol/L	4.94±0.75	5.0±0.75	0.56
LDL-C, mmol/L	2.79±0.72	3.0±0.74	0.42
HDL-C, mmol/L	1.52±0.33	1.46±0.38	0.44
NT-pro-BNP, pg /mL	9.54±18.94	35.99±45.85	0.062
Systolic BP, mm Hg	120.9±24.59	123.75±12.79	0.64
Heart rate, beats per 1 min.	79.4±11.67	74.66±4.64	0.62
LVEF, %	56.12±3.18	53.8±4.34	0.54
E/Am, U	1.11±0.17	0.98±0.21	0.12
E/Em, U	7.9±1.98	9.2±2.12	0.16
ACEI or ARAs, n (%)	15 (10.7)	18 (42.9)	<0.001
acetylsalicylic acid, n (%)	124 (88.5)	40 (95.2)	0.16
Statins, n (%)	83 (59.3)	11 (28.6)	<0.01
metformin, n (%)	3 (2.1)	5 (11.9)	0.001
diuretics, n (%)	3 (2.1)	23 (54.8)	0.001
Mineralcorticoid receptors antagonists, n (%)	9 (6.4)	15 (35.7)	0.001

Note: * - statistically differences between parameters in the two groups ($P < 0.05$); CI – confidence interval; CAD – coronary artery disease, T2DM – type two diabetes mellitus, GFR - Glomerular filtration rate, HDL-C - high-density lipoprotein cholesterol, LDL-C - Low-density lipoprotein cholesterol, BP – blood pressure, BMI - Body mass index, BNP – brain natriuretic peptide, LVEF - Left ventricular ejection fraction, U – unit, Em - early diastolic myocardial velocity, Am - late diastolic myocardial velocity, E – peak velocity of early diastolic left ventricular filling, ACEI – angiotensin-converting enzyme inhibitor, ARAs – angiotensin-2 receptors antagonists.

Discussion

Despite the presence of guidelines related to multiple myeloma, leukemia, Hodgkin's and non-Hodgkin's lymphoma, there are still gaps between best evidence as described in guidelines and quality of care in daily practice [9]. Little is known about factors that affect this discrepancy. It is postulated that a part of the patients with documented lymphoproliferative diseases might be identified at high cardiovascular risk. The conventional treatments used in most cases and coexisting conventional risks factors contribute worsening endothelial function and destroying endothelial layer. In this regard, biological markers that devoted severity of endothelial dysfunction look promising. We found that raised level of VE-catherin associated with a greater risk of cardiovascular events in asymptomatic coronary artery disease patients with lymphoproliferative diseases. Moreover, predictive value of VE-catherin was superior when compared with LVEF and NT-pro-BNP. While natriuretic peptides are strong prognostic indicators in heart failure and coronary syndrome, and possibly superior to traditional prognostic factors, the levels of NT-pro-BNP must be interpreted according to the clinical picture [6]. Looking for novel biomarkers are required to be improving reclassification of the risk in cumulative cardiovascular events over a clinical prediction model. Further research is necessary to understand the prognostic value of VE-catherin in patients with documented lymphoproliferative diseases.

In conclusion, among patients with documented lymphoproliferative diseases and known asymptomatic coronary artery disease increased circulating VE-catherin associates with increased 3-year cumulative cardiovascular events.

Conflict of interests: not declared

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Limitations of the study. This study has some limitations. We believed that a greater cohort would be desirable to improve the power of the study. We also relied on clinical data to rule out infection and other inflammatory diseases before sampling, but we couldn't exclude that some patients had unrecognized the conditions responsible for the elevated VE catherin levels observed.

References

- [1] D.A. Bluemke, S. Achenbach, M. Budoff, T.C. Gerber, B. Gersh, L.D. Hillis, W.G. Hundley, W.J. Manning, B.F. Printz, M. Stuber, P.K. Woodard, Noninvasive coronary artery imaging: magnetic resonance angiography and multidetector computed tomography angiography: a scientific statement from the American Heart Association Committee on Cardiovascular Imaging and Intervention of the Council on Cardiovascular Radiology and Intervention, and the Councils on Clinical Cardiology and Cardiovascular Disease in the Young, *Circulation*, **118** (2008), 586 - 606.
- [2] D.E. Conway, M.T. Breckenridge, E. Hinde, E. Gratton, C.S. Chen, M.A. Schwartz, Fluid shear stress on endothelial cells modulates mechanical tension across VE-cadherin and PECAM-1, *Curr. Biol.*, **23** (2013), 1024 - 30.
- [3] J.M. Gardin, D.B. Adams, P.S. Douglas, H. Feigenbaum, D.H. Forst, A.G. Fraser, P.A. Grayburn, A.S. Katz, A.M. Keller, R.E. Kerber, B.K. Khandheria, A.L. Klein, R.M. Lang, L.A. Pierard, M.A. Quinones, I. Schnittger, American Society of Echocardiography. Recommendations for a standardized report for adult transthoracic echocardiography: a report from the American Society of Echocardiography's Nomenclature and Standards Committee and Task Force for a Standardized Echocardiography Report, *J. Am. Soc. Echocardiogr*, **15** (2002), 275–290.
- [4] J. Gavard, Endothelial permeability and VE-cadherin: A wacky comradeship, *Cell Adh. Migr.*, **6** (2013), 55 - 461.
- [5] T. Ishida, Triplets therapy for the patients of multiple myeloma, *Nihon Rinsho*, **9** (2013), 1691 - 1696.
- [6] D. Korenstein, J.P. Wisniversky, P. Wyer, R. Adler, D. Ponienman, T. McGinn, The utility of B-type natriuretic peptide in the diagnosis of heart failure in the emergency department: a systematic review, *BMC Emerg. Med.*, **7** (2007), 6.

- [7] J. Laubach, T. Hideshima, P. Richardson, K. Anderson, Clinical translation in multiple myeloma: from bench to bedside, *Semin. Oncol.*, **5** (2013), 549 - 553.
- [8] O. Sezer, K. Niemoller, C. Jakob, I. Zavrski, U. Heider, J. Eucker, Relationship between bone marrow angiogenesis and plasma cell infiltration and serum beta2-microglobulin levels in patients with multiple myeloma, *Ann. Hematol.*, **80** (2001), 598 - 601.
- [9] J.J. Stienen, P.B. Ottevanger, L. Wennekes, S.A. van de Schans, H.M. Dekker, N.M. Blijlevens, van der R.W. Maazen, J.H. van Krieken, R.P. Hermens, Delivering high-quality care to patients with a non-Hodgkin's lymphoma: barriers perceived by patients and physicians, *Neth. J. Med.*, **1** (2014), 41 - 48.
- [10] E. Terpos, G. Morgan, M.A. Dimopoulos, M.T. Drake, S. Lentzsch, N. Raje, O. Sezer, R. García-Sanz, K. Shimizu, I. Turesson, T. Reiman, A. Jurczyszyn, G. Merlini, A. Spencer, X. Leleu, M. Cavo, N. Munshi, S.V. Rajkumar, B.G. Durie, G.D. Roodman, International Myeloma Working Group recommendations for the treatment of multiple myeloma-related bone disease, *J. Clin. Oncol.*, **18** (2013), 2347 - 2357.
- [11] K. Vandyke, A.W. Chow, S.A. Williams, L.B. To, A.C. Zannettino, Circulating N-cadherin levels are a negative prognostic indicator in patients with multiple myeloma, *Br. J. Haematol.*, **4** (2013), 499 - 507.
- [12] T. Wrobel, G. Mazur, D. Wolowiec, B. Jazwiec, E. Sowinska, K. Kuliczowski, sVE-cadherin and sCD146 serum levels in patients with multiple myeloma, *Clin. Lab. Haematol.*, **1** (2006), 36 - 39.
- [13] X.G. Zhou, Towards standardization of lymphoma diagnosis, *Zhonghua Bing Li Xue Za Zhi*, **4** (2013), 220 - 221.

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