Vascular versus myocardial dysfunction in acute coronary syndrome: Are the adhesion molecules as powerful as NT-proBNP for long-term risk stratification?

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Abstract

Objectives—To determine if elevations of adhesion molecules in acute coronary syndrome (ACS) are useful for risk stratification.

Design and methods—A cell adhesion array (Randox Ltd.) and NT-proBNP were measured in 216 ACS patients.

Results—Kaplan–Meier and Cox models indicate early elevations of NT-proBNP but not the adhesion molecules are predictive of future death/myocardial infarction.

Discussion—Elevations of adhesion molecules early after pain onset in ACS are not useful for long-term risk stratification.

Keywords

Cell adhesion; NT-proBNP; MI; Death; ACS

Introduction

The biological functions of the cell/vascular adhesion molecules have been well described in atherosclerosis [1]. Both the selectins (L-, P-, E-selectins) and integrins (VCAM-1, ICAM-1) play a role in atherogenesis by recruitment of leukocytes via key migratory steps

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such as tethering, rolling (e.g., selectins) and adhesion (e.g., integrins) [1]. It has been proposed that the adhesion molecules may be elevated in the plaque destabilization phase and may be associated with endothelial activation or damage in ACS [2,3]. A recent report indicated that high concentrations of adhesion molecules, one week after the ACS event, were associated with an increased long-term risk of death, MI, or recurrent ACS [2]. However, a multi-marker approach by Apple et al. [4] evaluating a variety of biomarkers including some related to plaque destabilization (MMP-9, MPO), plaque rupture (CD40L, PIGF), and myocardial dysfunction (NT-proBNP), demonstrated that only NT-proBNP was independently predictive of short-term (120 days) mortality and/or cardiac events (e.g., myocardial infarction (MI), percutaneous coronary intervention, coronary artery bypass graft). In this study, we sought to determine if there was an independent long-term risk (up to 8 years post event) of death and MI associated with elevated NT-proBNP concentrations and/or elevations of the adhesion molecules (ICAM-1, VCAM-1, E-selectin, P-selectin, L-selectin) early after presentation in patients with ACS.

Methods

The study population has been previously described in detail [5–7]. In brief, following research ethics board approval, subjects ($n=216$) from our ACS study population (448 patients originally recruited in 1996) were selected if there were at least 2 EDTA specimens available in storage (-70°C) per subject. The decision was made to analyze at least 2 samples so rises that might occur after the initial sample collection that could be informative were not missed. The specimen pair chosen consisted of the earliest available specimen (1st specimen: median 2 h from pain onset; interquartile range (IQR): 2–4 h) and the next specimen closest to 9 h post onset of symptoms (later specimen: median 9; IQR: 9–9 h). In the event that the first specimen was obtained after 6 h post onset, then the next specimen drawn at least 3 h later was chosen for the specimen pair. The EDTA samples were thawed for the first time and measured for cTnT (4th generation) and NT-proBNP (Elecsys®, Roche). After a second thaw, the cell adhesion array (evidence investigator™, Randox Ltd) was measured to obtain concentrations for VCAM-1, ICAM-1, E-selectin, P-selectin, L-selectin. The interassay ($n=20$) imprecision (CV) ranged from 10.8–21.6% for the cell adhesion array, and 3.6–6.3% for cTnT and NT-proBNP. There is data to support the stability of these biomarkers during long-term storage [7–9]. During the analytical phase of the study, 4 specimens for cTnT and 15 specimens for the cell adhesion array from the 432 specimens measured either had high background or instrument error codes and these specimens were not used in the statistical analysis.

The concentrations of cTnT, NT-proBNP and the adhesion molecules were linked to the existing cTnI concentrations (AccuTnI, 2nd generation, Access®, Beckman Coulter) and health outcomes databases (Registered Persons Data Base for mortality outcomes and the Canadian Institute for Health Information Discharge Abstract Database for hospital discharges associated with MI [10]). Based on the death date and earliest subsequent readmission for MI, indicators were created to reflect whether or not an event (death or MI readmission) occurred within 8 years post presentation (patients who died without previous MI readmission, follow-up were censored at the date of death). The biomarker concentrations of NT-proBNP and the adhesion molecules in the 1st specimen were used to

determine the risk in a manner similar to the design of Apple et al. [4]. For our study cohort, tercile analysis for the combined endpoint (death/MI) was used for all analyses to maximize the number of events. Kaplan–Meier curves were constructed to display cumulative probability of AMI-free survival to 8 years post presentation. Differences between strata were assessed using the log-rank test statistic. Cox proportional hazard models assessed the association of elevated NT-proBNP levels and time to an event: model 1 adjusts for age (continuous variable), sex and cTnT concentration in the second specimen (cTnT 0.01 as reference), whereas model 2 adjusts for age (continuous variable), sex, and cTnI in the second specimen (cTnI 0.01 as reference). Significance of the hazard ratios was based on the Wald chi-square statistic, with significance set at $p<0.05$. Between-group comparisons of central tendency were based on the Wilcoxon signed rank test. Analyses were performed using SAS version 9.1.3.

Results

We have previously reported an increase in NT-proBNP concentrations early after the onset of pain in our ACS population [7]. In comparing the concentrations of adhesion molecules we observed higher concentrations in the 1st specimen (median 2 h post pain onset) vs. the later specimen (median 9 h post onset) for L-selectin (early 1st specimen median concentration: 1354 $\mu\text{g/L}$ vs. later specimen: 1265 $\mu\text{g/L}$; $p<0.001$), E-selectin (17.7 $\mu\text{g/L}$ vs. 17.0 $\mu\text{g/L}$; $p=0.001$), and ICAM-1 (323 $\mu\text{g/L}$ vs. 316 $\mu\text{g/L}$), but not for P-selectin ($p=0.176$) or VCAM-1 ($p=0.291$). Kaplan–Meier analyses based on biomarker terciles showed increased probability of death/MI for increasing NT-proBNP concentrations ($p<0.001$). However, there was no difference between the tercile groupings for the adhesion molecules for outcomes up to 8 years following the initial event ($p>0.05$) (Fig. 1).

Cox proportional hazard models were then generated for NT-proBNP. The cTnT and cTnI concentrations at the second time point (9 h post onset) were used in models 1 and 2, respectively. The 2nd time point was chosen (“later” specimens), because the concentrations of cTn are greater at this time point compared to the earlier time point (2 h post onset), and thus might better reflect infarct size a known determinant of prognosis. After adjusting for age, sex and the cTn levels, the top NT-proBNP tercile group (i.e., >373 ng/L) was at greater risk for death/MI as compared to the lowest NT-proBNP group (Table 1).

Discussion

Adhesion molecules have roles in leukocyte recruitment and capture and may be associated with endothelial activation or damage in ACS [1,2]. Studies have examined the long-term risks of the adhesion molecules in the setting of coronary artery disease [8,9] and after an ACS event [2], but little is known on the long-term effects of elevated concentrations of the adhesion molecules early after the onset of pain in an ACS population. Our results indicate that measurements of the adhesion molecules early after onset of symptoms do not identify patients at greater long-term probability for death and/or re-current MI. This finding might be viewed as unexpected as the biological roles of the adhesion molecules, especially during the vascular inflammation/dysfunction phase of ACS, would select these biomarkers as good candidates for risk stratification [2,3]. It may be that values measured acutely are influenced

by the acute event itself and that later samples, for example at 1 week post event, as evaluated by others might provide better information concerning the underlying pathophysiology. Moreover, there are many therapeutic agents used for patients with cardiovascular disease (e.g., aspirin) that may effect the concentration of the cell adhesion molecules, which in turn may limit the usefulness of measuring these markers during the acute event [11]. Our population, sampled over a decade ago, received much less therapeutic intervention than a present day cohort would (e.g., only 27% of the population received ASA during their presentation). Thus, the opportunity to detect these effects was enhanced for our population and so the absence of an association in our data between recurrent MI and/or death and cell adhesion molecules then makes it highly unlikely that these biomarkers would be predictive in a far more aggressively treated present day ACS population. Our findings confirm previous work showing that myocardial dysfunction, as seen by elevated NT-proBNP concentrations, identifies groups of individuals with a greater probability of death over the long term. Moreover, the risks for long-term death/MI remained significant for those subjects with the highest NT-proBNP concentration (e.g., >373 ng/L) even after adjusting for either cTnT or cTnI.

Our study confirms and builds on existing data that myocardial dysfunction in ACS is a strong predictor for poor outcomes [4]. Despite limitations in our study (e.g., small sample size), we believe that these data further refine which biomarkers, in addition to cTn, should be used for risk stratification during the very early stages of ACS.

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Fig. 1. Kaplan-Meier survival curves for long-term probability of death/MI in an ACS cohort based on tercile analysis for NT-proBNP (a); L-selectin (b); P-selectin (c); E-selectin (d); ICAM-1 (e); and VCAM-1 (f).

Table 1

Cox proportional hazard models for long-term risk of death/MI for NT-proBNP elevations early after onset of symptoms suggestive of cardiac ischemia

Model	Time since presentation	NT-proBNP (ng/L) tertiles 2 and 3	Hazard Ratio relative to NT-proBNP <65 ng/L	Lower 95% CL	Upper 95% CL	Chi-square <i>p</i> -value
Model 1	6 months	65–373	3.10	1.04	9.26	0.043
		>373	3.71	1.33	10.36	0.012
	2 years	65–373	2.57	1.03	6.39	0.042
		>373	3.89	1.66	9.14	0.002
Model 2	8 years	65–373	1.78	0.99	3.19	0.053
		>373	3.00	1.69	5.32	<0.001
	6 months	65–373	2.19	0.77	6.22	0.141
		>373	3.23	1.20	8.68	0.020
	2 years	65–373	2.01	0.84	4.83	0.119
		>373	3.63	1.58	8.32	0.002
	8 years	65–373	1.63	0.93	2.88	0.091
		>373	3.11	1.79	5.42	<0.001

Model 1: adjusted for age, sex, and cTnT levels (0.01 as reference).

Model 2: adjusted for age, sex and cTnI categories (0.01 as reference).