Serum deoxyribonuclease I activity can be used as a novel marker of transient myocardial ischaemia: results in vasospastic angina pectoris induced by provocation test

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Aims Serum deoxyribonuclease I (DNase I) activity has recently been highlighted as a potential diagnostic marker for detection of acute myocardial infarction. To evaluate whether serum DNase I activity is useful for detection of myocardial ischaemia, we investigated alteration of its levels after onset of vasospastic angina pectoris (VSAP), resulting in transient myocardial ischaemia, induced by the intracoronary ergonovine provocation test.

Methods and results Twenty-nine consecutive patients with suspected VSAP were subjected to the test. Patients were categorized as VSAP-positive (n = 13) or -negative (n = 16) based on development of angina. Serum samples were examined for DNase I activity before, immediately after, and 3, 6, and 24 h after the provocation tests. The serum DNase I activity increased significantly from the baseline 3 h after the provocation test in 11 patients of the VSAP-positive group whose levels of troponin T were within the normal range. Median of the percentage differences from the baseline in serum DNase I activity 3 h after the test was 32.1% (25th and 75th percentile: 28.6 and 42.0%, respectively; P = 0.000012). In the VSAP-negative group, levels of DNase I activity remained unchanged at any point of time after the provocation test.

Conclusion Transient myocardial ischaemia resulting from VSAP induces a significant elevation of serum DNase I activity. Therefore, serum DNase I activity may be applicable as a useful marker for detecting transient myocardial ischaemia.

KEYWORDS Ischaemia; Vasospastic angina pectoris; Deoxyribonuclease I

Introduction

A lack of rapid and reliable tests has often hampered the diagnosis of acute and transient myocardial ischaemia in patients with acute coronary syndromes (ACS). To improve the process of triage of patients with ACS, useful diagnostic markers that can confirm or exclude myocardial ischaemia are needed. Until recently, however, there have been no biomarkers capable of detecting the presence of reversible myocardial ischaemia.¹,² Deoxyribonuclease I (DNase I, EC 3.1.21.1) is an endonuclease that preferentially attacks double-stranded DNA in a Ca²⁺-dependent manner to produce oligonucleotides with 5'-phospho and 3'-hydroxy termini.³ Recently, we have demonstrated that the serum level of DNase I activity is abruptly elevated within ~3 h after the onset of symptoms in patients with acute myocardial infarction (AMI), thereafter exhibiting a marked time-dependent decline within 12 h, and a return to the basal level within 24 h.⁴ Moreover, when percutaneous coronary intervention (PCI) was performed to patients with stable angina pectoris, irrespective of any alteration in the levels of creatine kinase isoenzyme MB (CK-MB) and cardiac troponin T (c-TnT), serum DNase I increased significantly from the basal level by 3 h after completion of the PCI procedure.⁵ On the basis of these findings, we postulated that myocardial ischaemia rather than injury...
induces this elevation of serum DNase I activity. Therefore, it is anticipated that serum DNase I may be a sensitive marker for detection of transient myocardial ischaemia. However, further evaluation of the relationship between serum DNase I elevation and myocardial ischaemia is required before serum DNase I can be applied clinically for diagnosis of myocardial ischaemia.

It is well known that coronary vasospasm is involved in the pathophysiology of variant angina pectoris, AMI, and sudden cardiac death. It is especially important to determine whether episodic or chronic coronary vasospasm is involved in the pathophysiology of variant angina pectoris, AMI, and sudden cardiac death. Coronary vasospasm has been considered to be the most important cause of variant angina pectoris because it induces less myocardial damage and mechanical stress than balloon angioplasty. Provocation testing is performed to clarify the aetiology of angina pectoris, especially in patients without distinct coronary artery disease; it is worth noting that patients in whom angina does and does not develop as a result of the provocation test provide two distinct background-matched case–control groups to study myocardial ischaemia.

In this clinical study, we investigated whether the levels of serum DNase I activity in such patients could be altered in response to transient myocardial ischaemia induced during the intracoronary ergonovine provocation test. We then evaluated the utility of DNase I as a marker for the detection of transient myocardial ischaemia.

Methods

Subjects

Fifty-six consecutive Japanese patients with suspected VSAP, who had atypical chest pain at rest and were admitted to our institutions between March 2005 and December 2006, were subjected to diagnostic coronary angiography (CAG). Among them, 21 patients were excluded from the study because they were found to have ≥50% organic stenosis by coronary angiography. We, therefore, planned to perform the intracoronary ergonovine provocation test on the remaining 35 patients who had no significant organic stenosis (<50% lumen diameter). However, six of these patients declined to take part in the study, and ultimately 29 patients were, therefore, enrolled. Twelve patients who underwent only CAG and were recruited as a CAG group, were the same group as those recruited in our previous study; 66 patients admitted to our institutions underwent diagnostic CAG for typical exertional chest pain. Forty-eight patients were excluded because they had conditions such as >25% stenosis in a major coronary artery, valvular disease, myocarditis, and signs or symptoms of acute ischaemic conditions in the 2 weeks before catheterization. Furthermore, six patients declined to take part in the study. Ultimately, 12 patients were enrolled as the control group.

The study protocol conformed to the Declaration of Helsinki and was approved by the Human Ethics Committee of our institutions; each subject included in the study gave written informed consent before study participation.

Catheterization procedure, angiographic analysis, and sample collection

At least 24 h prior to the provocation test, nitrates, calcium channel blockers, and other anti-anginal drugs, except sublingual nitroglycerin, were withdrawn from all patients. All the CAG procedures were performed by a radial approach using the standard Judkins technique after administration of 3000 U of heparin in the morning. In the provocation test, 0.01 mg of ergonovine maleate was injected into either the left or the right coronary artery of the patients through a catheter within 4 min. If coronary spasm was not provoked, the patients were administered an additional 0.04 mg of ergonovine maleate. A standard 12-lead electrocardiogram (ECG) was recorded every 30 s to assess ST-segment shift. Heart rate and blood pressure of each patient were monitored continuously during the procedure. Patients informed the examiner if chest pain occurred during the provocation test. When angina developed with chest pain and ST-segment shift, angiography was immediately performed, and subsequently 0.25 mg of nitroglycerin was injected into the responsible coronary artery to relieve the symptoms.

Coronary vasospasm was defined as transient total (100%) or near-total (99%) occlusion of the provoked coronary artery, which was reversible with nitroglycerin. Patients were categorized as test positive if they developed angina with ST-segment shift and chest pain, and exhibited a thrombolysis in myocardial infarction (TIMI) 0 or TIMI 1 flow on the coronary angiogram. Among the 29 patients subjected to the intracoronary ergonovine provocation test, 13 were test positive (VSAP-positive group) and 16 were test negative (VSAP-negative group).

Blood samples were taken from the radial sheath before and soon after the end of the provocation test. Follow-up blood samples were obtained from an antecubital vein 3, 6, and 24 h after completion of the provocation test. Serum samples were separated from each blood sample by centrifugation and stored at −80 °C until use.

Measurement of deoxyribonuclease I activity and cardiac markers in serum of patients

Levels of DNase I activity in serum samples were measured by the single radial enzyme diffusion (SRED) method, as described previously. One unit of enzyme assayed corresponds to 0.6 ng of purified human DNase I. We had previously determined the mean intra-individual percentage difference in activity levels to be 7.0 ± 2.7% in healthy volunteers. The upper limit of the normal range of intra-individual percentage differences was estimated to be 12.4% (mean ± ZSD) as a tentative cutoff value. When the percentage differences in serum DNase I activity levels from the baseline in each patient exceeded the cutoff level, elevation of activity levels was considered positive. To clarify whether the activity measured by the SRED method was entirely derived from DNase I, we employed an inhibition assay using anti-human DNase I antibody, as described previously. Serum CK-MB concentration was determined with an automated chemiluminescence system (Ciba Corning Diagnostics Corp., Medfield, MA, USA) and serum c-TnT with an electrochemiluminescence immunoassay system (Elecys 1010 System, Roche Diagnostics Corp., Mannheim, Germany), in accordance with the manufacturer’s instructions. Cutoff levels of CK-MB and c-TnT indicative of positive activity were taken as 5.20 and 0.01 μg/L, respectively. Furthermore, serum myoglobin was determined with an automated immunochromilumimetric assay system (Chemilumi ACS, Centaur, Bayer Medical Co. Ltd., Tokyo, Japan).

Statistical analyses

Percentage differences from the baseline at each time interval after completion of the procedure were used to clearly reflect the within-patient variability of serum DNase I activity and myoglobin levels, and expressed tentatively as the median and 25th and 75th percentile range. The values were calculated as follows:

\[
\text{Percentage difference} = \left( \frac{A - B}{B} \right) \times 100
\]

where A is the assay level after the provocation test and B is the baseline assay level before the provocation test.

The Tukey-Kramer multiple comparison test was used to assess differences in values measured before and at various time points after the provocation test. The analysis did not account for the
correlations among the time points. Categorical variables in the clinical background were compared by χ² test or unpaired non-parametric test. Correlations between variables were evaluated using Pearson’s correlation coefficient. Data analysis was performed with StatView software, version 5.0, and R software, version 2.5.1 (http://www.r-project.org/). Differences were examined using a two-sided test at a significance level of 0.05. On the basis of our previous study, for the 12 patients used as a CAG group, the power to detect a 46% difference in serum DNase I activity levels for a two-sided test with a level of significance of P = 0.05 was 83.3%.

Results

Clinical characteristics of the study group

The clinical characteristics of all the patients included in the VSAP-positive and -negative groups, in addition to the CAG group, are summarized in Table 1. There were no significant differences in mean age, sex, and prevalence of coronary risk factors such as hypertension, high cholesterol levels, smoking status, or obesity between the three groups. In the VSAP-positive group, all the patients (100%) experienced chest pain during the provocation test. Among them, two (15%) and 11 (85%) patients exhibited TIMI 0 and TIMI 1 flow, respectively, on the coronary angiogram. All the former and latter patients showed a transient ST-segment elevation and depression, respectively. No patients in the VSAP-negative and CAG groups had narrowing of the involved coronary artery, ECG changes, or chest pain during the procedure. None of the patients in this study had major complications such as ventricular rhythm disturbance, refractory spasm, or myocardial infarction.

Alterations of serum deoxyribonuclease I activity levels during the provocation test

In almost all the patients in the VSAP-positive group who developed angina as a result of the provocation test, a significantly marked elevation in serum DNase I activity was observed 3 h after the test; the activity level then tended to return to baseline by 24 h (Figure 1A). Time-dependent alterations in the levels of serum DNase I activity exhibited close similarity to those observed in AMI patients after onset, and patients with stable angina pectoris after the PCI procedure. All the enzyme activity detected in the serum samples by the SRED method was completely abolished by anti-human DNase I antibody, confirming that the activity was entirely derived from authentic DNase I. Obvious alterations in the activity level were not found in all the patients (VSAP-negative group) who did not develop angina during the provocation test (Figure 1B) and those in the CAG group. None of the patients in the VSAP-positive and -negative groups showed any significant elevation of c-TnT and CK-MB induced by the test within the same time window as that of serum DNase I. Although alterations in the levels of serum myoglobin occurred even in the VSAP-negative groups in the same manner as the VASP-positive groups, in terms of percentage differences [19.1% (−6.84, 34.7) and 21.6% (−1.86, 26.3), respectively] from the baseline at 3 h after the test, there was no significant difference between them, suggesting that alterations of serum myoglobin induced by the test might not have been due to myocardial ischaemia.

Table 2 shows the percentage differences from the baseline of serum DNase I activity levels after the provocation test in the VSAP-positive and -negative groups, together with the results in the CAG group. In the VSAP-positive group (n = 13), the percentage difference at 3 h after the test compared with the baseline was 32.1% (28.6, 42.0), being significantly higher than the baseline levels (P = 0.000012). The percentage difference declined and returned to baseline at 6 and 24 h after the test [3.67% (−7.67, 28.9) and −8.23% (−16.1, 1.06), respectively]. When the upper limit (12.4%) of the normal range of intra-individual percentage difference in the healthy volunteers was used as the cutoff level of DNase I elevation, 11 (85%) out of 13 patients in the VSAP-positive group were determined to be DNase-I-elevation-
positive. However, no significant elevation of the DNase I activity was observed in either the VSAP-negative group or the CAG group; all the patients in these groups were DNase-I-elevation-negative.

In the VSAP-positive group, there was no correlation between the highest post-procedural serum DNase I levels and TIMI flow grade, distribution of the occluded coronary artery, or ST-segment change during the provocation test. Although the extent of the increase in serum DNase I activity observed after the provocation test was relatively small compared with that in patients with the onset of AMI and after PCI, the levels of serum DNase I activity induced at 3 h after the test in the VSAP-positive group were significantly higher than those in the VSAP-negative group ($P = 0.0029$; Figure 2).

**Discussion**

**Major findings**

In this clinical study, we have demonstrated that coronary spasm induced by the intracoronary ergonovine provocation test can induce temporary elevation of DNase I activity in serum of the patients. The levels of activity increased significantly from the baseline by 3 h after the provocation test in the VSAP-positive group, whereas none of the VSAP-negative group and patients who underwent CAG only exhibited any alterations of serum DNase I activity at any time after the provocation test and angiography. Recently, we have reported that serum DNase I activity could be used as a sensitive marker for detection of transient myocardial ischaemia induced by PCI. However, Giannitsis and Katus pointed out that PCI has several shortcomings and may not be an ideal model for evaluating the effect of myocardial ischaemia, because there is concern that peri-interventional myocardial infarction could develop due to side branch occlusion, distal embolization, or major coronary dissection, and also because inflation of the balloon at high pressure would cause endothelial injury or plaque disruption. Therefore, we performed this study to address these issues. In this clinical study, coronary spasm provoked by the test was relieved within 1 min after intracoronary injection of nitroglycerin. Furthermore, none of the patients in the VSAP-positive group showed significant elevation of c-TnT and CK-MB. Obviously, in comparison with the mechanical stress resulting from balloon inflation or provisional coronary stenting in the PCI, the provocation test is less likely to cause endothelial injury. Accordingly, it is plausible

| Table 2 Percentage differences from the baseline of serum deoxyribonuclease I activity levels, soon, and at 3, 6, and 24 h after the provocation test and coronary angiography$^a$ |
|---|---|---|---|---|
| Group | No. of patients | Percentage difference from baseline after the procedure$^b$ | Deoxyribonuclease-I-elevation-positive, n (%) |
| | | Soon | 3 h | 6 h | 24 h |
| Provocation test | | | | | |
| Vasospastic angina pectoris-positive | 13 | 4.18 | 32.1 | 3.67 | −8.23 | 11 (85%) |
| (−5.82, 16.2) | (28.6, 42.0) | (−7.67, 28.9) | (−16.1, 1.06) |
| Vasospastic angina pectoris-negative | 16 | −3.74 | −8.38 | −0.71 | −12.1 | 0 (0%) |
| (−7.24, 3.37) | (−17.7, 4.32) | (−8.25, 4.32) | (−24.1, 1.98) |
| Coronary angiography | 12 | 2.94 | −3.38 | n.d.$^d$ | −5.25 | 0 (0%) |
| (−4.45, 19.3) | (−17.7, 7.96) | | (−14.5, 5.45) |

$^a$Data taken from Arakawa et al. $^5$
$^b$Values are expressed as the median, and 25th and 75th percentile values in parentheses.
$^c$Significantly different compared with baseline.
$^d$Not determined.

Figure 1 Changes in serum deoxyribonuclease I level as a function of time after the intracoronary ergonovine provocation test in 13 vasospastic angina pectoris-positive patients (A) and 16 vasospastic angina pectoris-negative patients (B). Data are expressed as the percentage difference in activity levels from the baseline at each time point after completion of the procedure. Broken line represents upper limit of the normal range of intra-individual variation in serum DNase I activity.

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that coronary artery spasm induced by the test did not cause myocardial damage, severe endothelial injury, or plaque disruption such as that possibly occurring after PCI. All the patients with TIMI 1 flow on coronary arteriograms showed ST-depression on the ECG. ST-elevation denotes transmural myocardial ischaemia, whereas ST-depression indicates nontransmural or subendocardial ischaemia, and the degree of endothelial injury and ischaemia-reperfusion injury may be less in patients showing TIMI 1 flow on arteriograms than in those with TIMI 0. However, even if patients did show TIMI 1 flow on coronary angiograms during the provocation test, the serum DNase I activity was significantly elevated from the baseline 3 h after the test. These findings allow us to conclude that vasospasm induced by the ergonovine provocation test could provide a good in vivo model of transient myocardial ischaemia, especially considering that VSAP-negative patients in whom no angina develops could serve as a background-matched control group for myocardial ischaemia, the VSAP provocation test may be superior to PCI as a human model of transient myocardial ischaemia.

**Evaluation of serum deoxyribonuclease I activity as a marker for myocardial ischaemia**

Ideally, in patients suspected to have ACS, it is essential to detect myocardial ischaemia before the occurrence of irreversible myocardial cell damage. Thus, identification of a biochemical marker that is more sensitive and specific for myocardial ischaemia and can be measured rapidly in serum would be clinically valuable. In our study group, most patients in whom coronary vasospasm developed as a result of the intracoronary ergonovine provocation test showed a transient and definitive elevation of serum DNase I activity, indicating that this may be due mainly to myocardial ischaemia as described above, whereas in all the VSAP-negative patients without occurrence of myocardial ischaemia, no rise in serum DNase I activity levels was observed after the test. These findings confirm and expand upon our previous report that in a human model of ischaemia induced by balloon angioplasty and in AMI patients, serum DNase I activity increases early and the level returns to the baseline within 24 h. An ideal biomarker for myocardial ischaemia should be detectable early after ischaemia onset and its level should fall within a period of about 24 h so that recurrent ischaemia can be detected and exacerbation of stable coronary disease by exertional ischaemia can be minimized. From this standpoint, although biochemical markers of vasospastic coronary disease, such as TG-rich lipoprotein and highly sensitive C-reactive protein, have been characterized, these markers cannot be applied to the detection of myocardial ischaemia. The observed time dependency of alterations in serum DNase I activity after the onset of myocardial ischaemia indicates that serum DNase I possesses these attributes. Thus, considering our results as a whole, serum DNase I appears to be suitable as a biochemical marker for detection of transient and reversible myocardial ischaemia. Ischaemic modified albumin (IMA), measured by the albumin-cobalt binding assay, has recently been shown to be a sensitive and early biochemical marker of ischaemia. In 85 and 88% of the patients with ischaemia induced by VSAP in this study and by PCI, respectively, significant elevation of serum DNase I activity occurred, and were judged to be ischaemia-positive. On the basis of cross-study comparison, serum DNase I may be not inferior to IMA (83–95%) with regard to sensitivity for detection of myocardial ischaemia. Although ACS, a well-known life-threatening disorder, is often confirmed in accordance with ACC/AHA guidelines, frequent absence of typical chest pain, ST-segment shift, regional wall motion abnormalities, and cardiac marker elevation makes definitive diagnosis of ACS difficult. Accordingly, assessment of serum DNase I could be useful for diagnosis of ACS inducing myocardial ischaemia, and may prove to be a tool for risk stratification.

**Study limitations**

First, the prevalence of VSAP is higher in Japanese patients with coronary heart disease than in their Caucasian counterparts. Therefore, there is a need to examine whether this elevation of DNase I levels in patients with provoked vasospasm is comparable in other ethnic groups. Furthermore, our sample size was small. It has been reported that IMA is elevated from the baseline even at 30 min after PCI. In this study, we examined alterations in the levels of serum DNase I activity at 3 h after the provocation test. It will be necessary to examine the response of serum DNase I to transient myocardial ischaemia at a stage earlier than 3 h after the test, for evaluation of the enzyme as a biochemical marker for early detection of myocardial ischaemia. As a cardiac marker, serum heart-type fatty-acid-binding protein is sensitive, although its levels in serum may be increased after the onset of trauma, renal dysfunction, and heart failure. In contrast, we have previously reported that serum DNase I activity is not increased after onset of such conditions. Although this and our previous studies demonstrated that transient myocardial ischaemia might be involved in elevation of serum DNase I activity levels, it has not been necessarily possible to definitely prove that serum DNase I is an early and specific biomarker for myocardial ischaemia. Therefore, further studies using larger numbers of patients will be required to evaluate whether serum DNase I activity is a clinically useful diagnostic marker for detection of transient...
myocardial ischaemia. Secondly, it remains to be clarified how myocardial ischaemia induced by AMI, PCI, and VSAP could elevate the levels of serum DNase I activity in patients. In this study, some increase in heart rate and blood pressure was observed immediately after the onset of the ST-segment changes with symptoms in all the VSAP-positive patients, but not in the VSAP-negative patients (data not shown). Furthermore, we could not examine correlations of pulmonary artery wedge pressure, left ventricular end-diastolic pressure, or endothelin level with serum DNase I activity level. Therefore, this study was unable to rule out the possibility that some potential factors such as haemodynamic changes in the left ventricle or stress during ischaemia might have affected levels of serum DNase I activity. Further clarification of the mechanism responsible for DNase I elevation and the physiological significance of DNase I in myocardial ischaemia will undoubtedly have important biological and clinical implications.

Conclusion

Transient myocardial ischaemia occurring in VSAP induced by the intracoronary ergonovine provocation test results in a significant elevation of DNase I activity in the serum of patients. Thus, elevation of the serum DNase I activity may be a useful marker for detecting transient myocardial ischaemia.

Conflict of interest: none declared.

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References

1. Wu AHB. The ischemia-modified albumin biomarker for myocardial ische-


2. Morrow DA, de Kemos JA, Sabatine MS, Antman EM. The search for a bi-


polymorphic deoxyribonuclease I: purification, characterization and multi-


4. Kawai Y, Yoshida M, Arakawa K, Kumamoto T, Morikawa H, Masamura K,

Tada H, Ito S, Hoshizaki H, Oshima S, Taniguchi K, Terasawa H, Miya-

mori I, Yasuda T. The diagnostic use of serum deoxyribonuclease I activity

as a novel early-phase marker in acute myocardial infarction. Circula-


5. Arakawa K, Kawai Y, Kumamoto T, Morikawa N, Masahiro Y, Tada H, Kawa-

deoxyribonuclease I activity can be used as a sensitive marker for
detection of transient myocardial ischaemia induced by percutaneous coro-


695–702.

7. Maseri A, Severi S, Nes MD, L’Abbate A, Chierchia S, Marzilli M,

Ballestra AM, Parodi O, Biagini A, Distante A. Variant angina: one aspect of a

continuous spectrum of vasospastic myocardial ischemia. Am J Cardiol 1978;


8. Yasue H, Kugiyama K. Coronary spasm: clinical features and pathogen-


10. Bertrand ME, LaBlanche JM, Timly PA, Thieuleux FA, Delforge MR,

Carre AG, Asseman P, Berzin B, Libersa C, Laurent JM. Frequency of pro-

coked coronary arterial spasm in 1089 consecutive patients undergoing


of coronary artery spasm by a direct local action of ergonovine. Circula-

tion 1987; 75: 577–582.


in human tissues and body fluids by a single radial enzyme-diffusion


quantification of DNase I activity in one-microliter serum samples. Clin


novel screening methods for selecting monoclonal antibodies which

specifically inhibit DNase I enzyme activity. Immunol Invest 1998; 27:

145–152.

16. Hallermayer K, Klenner D, Vogel R. Use of recombinant human cardiac
troponin T for standardization of third generation troponin T method.


17. Müller-Bardorff M, Hallermayer K, Schröder A, Ebert C, Borgya A,

Gerhardt W, Rempgis A, Zehelein J, Katus HA. Improved troponin T

ELISA specific for cardiac troponin T isoform: assay development and


18. Giannitsis E, Katus HA. Mirror, mirror on the wall: the quest for the ear-


19. Tsimikas S, Lau HK, Han KR, Shortai B, Miller ER, Segev A, Curtiss LK,

Mitztum JL, Strauss BH. Percutaneous coronary intervention results in

acute increases in oxidized phospholipids and lipoprotein(a): short-term

and long-term immunologic responses to oxidized low-density lipopro-


20. Scheinman MM, Abbott JA. Clinical significance of transmural versus non-

transmural electrocardiographic changes in patients with acute myocar-

21. Yamauchi H, Homma Y, Haneda S. Biochemical markers of vasospastic


22. Hung M-J, Cheng W-J, Yang N-I, Cheng C-W, Li L-F. Relation of high-

density lipoprotein levels with coronary vasospastic angina pec-

torise in patients without hemodynamically significant coronary artery


Reduced albumin–cobalt binding with transient myocardial ischaemia

after elective percutaneous transluminal coronary angioplasty: a prelimi-
nary comparison to creatine kinase-MB, myoglobin, and troponin I. Am

Heart J 2001; 141: 985–991.

24. Sinha MK, Gaze DC, Tippins JR, Collinson PO, Kaski J-C. Ischemia modified

albumin is a sensitive marker of myocardial ischemia after percutaneous coro-

25. Braunwald E, Antman EM, Beasley JW, Caiff RM, Chetinisi MD,

Hochman JS, Jones RH, Kereiakes D, Kupersmith J, Levin TN,


guidelines for the management of patients with unstable angina and

non-ST segment elevation myocardial infarction. J Am Coll Cardiol 2000;

36: 959–969.


artery vasomotor: differences between Japanese and Caucasian patients.


27. Okamoto F, Sohmyi K, Okihara Y, Kawamura K, Asayama K, Kinuma H,

Nishimura S, Ishii H, Sunahara N, Tanaka T. Human heart-type cyto-

plasmatic fatty-acid-binding protein (H-FABP) for the diagnosis of acute

myocardial infarction: clinical evaluation of H-FABP in comparison with

myoglobin and creatine kinase isoenzyme MB. Clin Chem Lab Med 2000;

38: 231–238.

28. Pelsers MHAL, Hermens WT, Glazt JFC. Fatty-acid-binding proteins as


Influence of renal function on serum and urinary heart fatty-acid-binding


30. Goto T, Takase H, Toriyama T, Sugihura T, Sato K, Ueda R, Dohi Y. Circulat-

ing concentrations of cardiac proteins indicate the severity of congestive