Anti-β2 Glycoprotein-I Antibody in Acute Myocardial Infarction

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Abstract: Problem statement: Ischemic cardiac manifestations have been reported in a various percentage of patients with anti-phospholipid antibodies. Data concerning the relation between anti-Phospholipid (aPL) antibodies and myocardial infarction in subjects without evidence of overt autoimmune disease are conflicting. Anti-beta2 glycoprotein-I (anti-beta2-GPI) antibody is detected in various diseases like rheumatoid arthritis, systemic lupus erythematosus and anti-phospholipid antibody syndrome. The study of anti-beta2-GPI antibody in Acute Myocardial Infarction (AMI) might shed light on etiologic mechanisms in the pathogenesis of acute coronary syndromes. The purpose of the present study was to determine association of plasma aPL antibodies, namely, anti-beta2-GPI antibodies, with AMI. This study was designed to investigate whether prevalence of anti-beta2-GPI antibodies, in patients who had acute myocardial infarction and to analyze their relationship with traditional cardiovascular risk factors.

Approach: We investigated the prevalence of anti-beta2-GPI IgG in a well characterized group of patients with AMI as a case group. Sera from 74 patients with AMI and from 76 healthy subjects, matched for age and sex as a control group. Using ELISA to evaluate the presence of IgG isotype of anti-beta2-GPI autoantibodies in their sera.

Results: The prevalence of anti-beta2-GPI IgG in the control group (10.50%) resulted significantly lower than in patients with AMI (37.80%) (p<0.005). There was significant difference between positive anti-β2-GPI test in patients with STEMI and those with NSTEMI (66.7% Vs 36.4%), (p = 0.020).

Conclusion: Our findings suggest that anti-beta2-GPI IgG antibodies seemed to behave as independent risk factors for myocardial infarction, which may represent a link between autoimmunity and atherosclerosis in patients with acute myocardial infarction. Further studies with bigger patients are needed to explore association of anti-β2-GPI IgG with STEMI and NSTEMI.

Key words: Anti-beta2 glycoprotein-I, Acute Myocardial Infarction (AMI), anti-Phospholipid (aPL) antibodies, GPI antibodies, Anti-Cardiolipin Antibodies (ACA), lupus anticoagulant, blood samples, control group, traditional cardiovascular

INTRODUCTION

Cardiovascular Diseases (CVD) remain one of the leading causes of deaths despite several advancements in the medical interventions. Among these, the ischemic heart diseases, Acute Myocardial Infarction (AMI) in particular, is one of the most alarming values (Upaganlawar et al., 2011).

Myocardial Infarction (MI) is the combined result of environmental factors and personal predispositions.

Factors such as low serum adiponectin (Shojaie et al., 2009a) and low annexin V levels (Shojaie et al., 2009b) and infectious diseases such as Mycoplasma pneumonia (Pourahmad et al., 2009) are a part of involving factors in MI.

Immunological factors may be involved in the etiopathogenesis of atherosclerosis (Jahromi et al., 2010a) and inflammation (Jahromi et al., 2010b). Antiphospholipid antibodies (aPL) in addition to repeated miscarriages and pregnancy complications
(Jahromi et al., 2010c) are associated with cardiovascular diseases (Jahromi et al., 2010a), however their pathogenic mechanisms are still matter of investigation.

In addition to the Classical Lupus Anticoagulant (LAC) and Anti-Cardiolipin Antibodies (ACA), other anti-Phospholipid Antibodies (aPL) were shown to target anionic phospholipids and other plasma proteins, including phosphatidylethanolamine, protein C, protein S, β2-Glycoprotein I (beta2-gpl) and annexin V (Tincani et al., 2010).

The frequency of anti-beta2-gpI antibodies, as well as their role in patients with acute myocardial infarction, has been a controversial issue and the exact role and clinical significance of anti-β2GPI antibodies in MI pathogenesis is doubtful.

Our study provides a complete profile of anticardiolipin and anti-beta2-gpI antibodies in patients with acute coronary heart disease, analyzes their frequency in patients with acute myocardial infarction and raises the possibility that anti-beta2-gpI antibodies act as independent risk factors for acute myocardial infarction in south of IRAN by the traditional Enzyme-Linked Immunosorbent Assay (ELISA) method.

MATERIALS AND METHODS

Subjects: This case-control research recruited 74 consecutive patients with Acute Myocardial Infarction (AMI) including 31 men and 14 women with the mean age of 62.7±13.1 years old who were taken to the emergency room of Peymanieh Hospital (Jahrom, Iran) with the chief complaint of chest pain from May 2010 to July 2010.

The diagnosis of myocardial infarction was established by cardiologists according to previously reported algorithms, such as clinical history, serial electrocardiographic alterations and laboratory tests confirming myocardial necrosis (Antman et al., 2008) and yet the cardiologists continued to ignore the results of antibody titers. The exclusion criteria for patients were as follows: (a) infective endocarditis; (b) neoplasias (current or past); (c) infection by the human immunodeficiency virus or treponema pallidum; (d) presence of known hereditary causes of thrombosis, such as homocystinuria or mutation of factor V (Leiden) and (e) previous diagnosis of antiphospholipid syndrome or another disease of the connective tissue.

We also selected 76 healthy individuals as our control group and matched them for age, sex and other CAD risk factors such as Hypertension (HTN), Diabetes Mellitus (DM) and Hyper Lipidemia (HLP). The exclusion criteria were as follows: (a) osteonecrosis; (b) infections, neoplasias, hereditary disorders, antiphospholipid syndrome, or diseases of the connective tissue.

The study protocol was accepted by research ethics committee of Jahrom University of Medical Sciences and informed consents were obtained from all participants before enrollment. The patient or his legal representative provided written informed consent.

Historical, demographic and clinical data were obtained through a review of medical records and interviews with patients and their families. The risk factors for myocardial infarction were as follows: (1) age, sex; (2) history of hypertension (diagnosis confirmed when the systolic or diastolic pressures were > 160 or 95 mmHg, respectively, or when the patient was using antihypertensive medication) 12; (3) smoking, according to the criteria of the British Council for Medical Research; (4) history of heart disease (atrial fibrillation or coronary heart disease, defined as previous myocardial infarction, angina, or revascularization procedure); (5) history of diabetes mellitus, according to the medical history or the use of insulin or an oral anti-diabetes drug; (6) hypercholesterolemia, based on total cholesterol > 200 mg dL⁻¹, LDL-cholesterol > 130 mg dL⁻¹, or total cholesterol/HDL-cholesterol ratio > 5 (Donahue et al., 1988).

Blood samples (5 cc) were obtained by venipuncture from the patients immediately after admission before starting any IV medications by skilled personnel. Blood samples were centrifuged and frozen within, at most, 2 hours after collection and stored at -70°C until laboratory testing with ELISA.

Serum anti-β2-GPI IgG level was measured by quantitative ELISA (Decavele et al., 2011), using the Aeskulia kit (Ref, 3206) following the manufacturer instructions. Results <19 U mL⁻¹ were interpreted as negative, while specimens with values ≥19 U mL⁻¹ were considered positive.

Statistical analyses were performed by SPSS (version 15; SPSS, Inc., Chicago, IL).

Data were expressed as mean ± 1SD. Continuous variables with little-to-mild skewness were summarized as mean ± SD and compared using Student’s t-test.

RESULTS

The demographic and clinical characteristics of the study groups, as well as laboratory variables are shown in Table 1. In the patient group 11 cases (14.9%) had Non-St Elevation MI (NSTEMI) and 63 (85.1%) had ST Elevation MI (STEMI).
Table 1: Demographic and clinical characteristics of study and control groups

| Variable                  | Case group | Control group | p-value |
|---------------------------|------------|---------------|---------|
| Age (years)               | 61.2±11.60 | 61.9±12.07    | 0.691   |
| Male, n (%)               | 57 (77.0)  | 60 (76.3)     | 0.883   |
| Current smoker, n (%)     | 39 (52.7)  | 17 (22.4)     | 0.027*  |
| HTN, n(%)                 | 36 (46.6)  | 13 (17.1)     | 0.024*  |
| Type 1 DM, n (%)          | 1 (1.4)    | 9 (11.8)      | 0.158   |
| Type 2 DM, n (%)          | 25 (33.8)  | 14 (18.4)     | 0.159   |
| Total cholesterol (mg dL$^{-1}$) | 195.2±40.3 | 163.8±43.2    | 0.038*  |
| LDL-C (mg dL$^{-1}$)      | 112.8±35.6 | 106±28.7      | 0.360   |
| HDL-C (mg dL$^{-1}$)      | 46.2±11.4  | 42.3±10       | 0.110   |
| LDL/HDL ratio             | 4.18       | 4.37          | 0.841   |
| Triglyceride (mg dL$^{-1}$)| 143.7±87.09| 104.4±41.15   | 0.010*  |
| +anti-$\beta$-GPI test, n | 28 (37.8)  | 8 (10.5)      | 0.003*  |

HTN: Hypertension, LDL-C: Low Density Lipoprotein-Cholesterol, HDL-C: High Density Lipoprotein-Cholesterol; Values are presented as mean ± SD or %, +anti-$\beta$-GPI test=more than 19 U/ml anti-$\beta$-GPI IgG

There was no significant difference between the two groups regarding the following variables: age, sex, HTN, DM, LDL, HDL, total cholesterol and TG. Prevalence of positive anti-$\beta$-GPI test (more than 19 U/ml anti-$\beta$-GPI IgG levels) in patients with AMI on admission were significantly higher than those in the control group 37.8% Vs 10.5% (p = 0.003) (Table 1).

We examined the association between positive anti-$\beta$-GPI tests and selected cardiovascular risk factors. There was a significant association between positive anti-$\beta$-GPI tests with Type hypertension. But there was not found any significant association between positive anti-$\beta$-GPI test with Type 1 DM, Type 2 DM, age, sex, LDL, HDL, TG, total cholesterol and adjusted smoking.

There was significant difference between positive anti-$\beta$-GPI test in patients with STEMI and those with NSTEMI (66.7% Vs 36.4%), (p = 0.020).

**DISCUSSION**

In this case-control study, in line with previous studies (Greco et al., 2009; 2010). We found that high prevalence positive anti-$\beta$-GPI test were associated with AMI among Iranian patients independent of traditional cardiovascular risk factors.

We found high prevalence of anti-$\beta$2GPI antibodies in patients with acute MI Vs. healthy control group consistent with current concepts on the immune pathogenesis of atherosclerosis. aPLs have been associated with CAD and acute MI (Greco et al., 2010).

There was no correlation between aPLs and standard cardiovascular risk factors such as smoking, hypertension and diabetes, as seen in a previous study (Jahromi et al., 2010a).

Previous studies have shown that the elevation of anti-$\beta$-2-GPI antibodies in human hypertension and its potential role in development of pregnancy-induced hypertension (Yamada et al., 2010).

An association was found between Anti-beta2GPI with CAD severity and adverse outcomes (Greco et al., 2009; 2010).

Studies have suggested that IgM isotypes may be more important in arterial events, whereas IgG isotypes may be more important in venous disease. Others have also found IgA anti-$\beta$2GPI to be important in nonautoimmune patients with MI. It was reported that anti-$\beta$2GPI antibodies were a significant risk factor for MI independent of other risk factors (Greco et al., 2010). In this study there was a significant association between anti-$\beta$2GPI IgG with HTN in the case and the control groups. The same results were found in other studies (Yamada et al., 2010). But there was not found any significant association between anti-$\beta$2GPI IgG with Type 1 DM, Type 2 DM, age, sex, LDL, HDL, TG, total cholesterol and adjusted smoking and also with type of MI, LV systolic Function (EF) and mortality in our cases.

These results suggest that anti-$\beta$2GPI IgG is independent of these variables.

As there was significant difference between positive anti-$\beta$-GPI test in patients with STEMI and those with NSTEMI, it can be concluded that anti-$\beta$2GPI IgG can cause more STEMI than NSTEMI. But, unfortunately we did not any data indicating association of anti-$\beta$-2GPI IgG with these parameters to compare the results.

**CONCLUSION**

These results show an association between Acute MI and high prevalence of anti-$\beta$2GPI IgG. We believe that the data from our study and from other studies (Urbanus et al., 2009; Greco et al., 2009; 2010) support the potential important role of anti-$\beta$2GPI IgG in acute coronary syndrome. Further studies with bigger patients are needed to explore association of anti-$\beta$-GPI IgG with STEMI and NSTEMI.

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