OPEN ACCESS
Scholars International Journal of Biochemistry
Abbreviated Key Title: Sch Int J Biochem
ISSN 2616-8650 (Print) |ISSN 2617-3476 (Online)
Scholars Middle East Publishers, Dubai, United Arab Emirates
Journal homepage: http://saudijournals.com/sijb/

Original Research Article

Plasma Plasminogen Activator Inhibitor-1 (PAI-1) Level - An Novel Prognostic Biomarker in ST Elevation Acute Myocardial Infarction among Young South Indian Patients

Latha P[1], ChandraKala K[2*]
[1,2]Department of Biochemistry, Tirunelveli Government Medical College, Tirunelveli, TamilNadu, India

***Corresponding author:** ChandraKala K
| **Received:** 15.02.2019 | **Accepted:** 24.02.2019 | **Published:** 28.02.2019
DOI:10.21276/sijb.2019.2.2.7

Abstract

Background: Incidence of younger age Acute Myocardial Infarction (AMI) is increasing worldwide. This study was aimed to ascertain plasma PAI-1 as an independent biomarker in STEMI, Quantification and Establishing the indicative evidence range of Plasma PAI-1 and ascertaining it's correlation with CK-MB in young south Indian ethnics. ***Methodology:*** This cross sectional study was conducted at MGMGH, Tiruchirapalli. Study subjects includes 40 Patients with typical chest pain, shows ST Elevation in ECG, rise in CK-MB and without any other risk factors of AMI. 40 age and sex matched control subjects were studied at the same time. Plasma PAI-1 (ELISA KIT-KOCH 3071) was assayed within six months of sample collection. Analysis of Serum Urea, Creatinine, Glucose, Lipid Profile, CK-MB and others risk factors of AMI was done on admission. Statistics was analyzed using SPSS -19.0. ***Results:*** There was a positive significance association observed in plasma PAI-1 ($P \leq 0.001$), Serum Urea: $P \leq 0.001$, Serum Glucose: $P \leq 0.04$, Serum AST: $P \leq 0.001$, Serum CK-MB: $P \leq 0.001$ and Serum HDL: $P \leq 0.008$ between patients and control subjects. The Mean and SD of plasma PAI-1 for patients and Controls are 3450.76 ± 1406.68 and 1966.03 ± 1406.68. Furthermore an inverse association observed between plasma PAI-1 and HDL level. ***Conclusion:*** This study statistically confirmed the independent association between STEMI and plasma PAI-1 and established its analytical range as 3000-5000 pg/ml, wherein, it is 1000- 2000 pg/ml for controls also observed the inverse association of Plasma PAI-1 with serum HDL levels.

Keywords: PAI-1, Acute Myocardial Infarction, young AMI, Biomarkers in AMI, ST elevation AMI, Plasminogen in STEMI.

INTRODUCTION

Coronary Heart Disease (CHD) is the most common cause of death in world. One in four men and one in six women died from CHD. The incidence of CHD varies from ethinics to ethinicswith the highest age-adjusted rates of 600–1000 deaths/100 000 in countries of the former Soviet Union and the lowest at around 60 per 100 000 in Japan (World Health Organization 2002). CHD will be the commonest single cause of death in developed countries over the next 20 years and will increase in frequency to become the commonest cause of disease-related disability in both developed and developing countries by the year 2020 [17]. ST Elevation Myocardial Infarction (STEMI) is the most common cause of death in developing countries like India. About 9% of new events occur in patients under 45 years of age; it is estimated that a genetic element is involved in some 20%-60% of these cases. Certainly, it is known that fibrinolytic activity which is mainly controlled by Plasma Plasminogen activator inhibitor -1 (PAI-1) is reduced in patients under 45 years of age with Acute Myocardial Infarction (AMI) [1]

Most myocardial infarctions are caused by a disruption in the vascular endothelium associated with an unstable atherosclerotic plaque that stimulates the development of an intracoronary thrombus, which results in coronary artery occlusion and reduced blood flow to cardiac tissue [2].

Plasminogen activator inhibitor type-1 (PAI-1) plays main role in inhibition of the activity of fibrinolytic system [3], it achieves this via inhibition of tissue Plasminogen activator (tPA [4, 5] inhibition of the inhibitor of the urokinase type activator (uPA). An increase in plasma concentration due to genetic and acquired conditions like young AMI, Stroke and Metabolic syndrome are therefore associated with

thrombotic events [6]. Over expression of PAI-1 may also promote development of weak plaques with thin fibrous caps due to inhibition of both u-PA receptor- and integrin-mediated cell adhesion and migration. In addition increased plasma PAI-1 levels have been reported in survivors of myocardial infarction (MI) compared with the general population. Therefore, PAI-1 might play an important role in the pathogenesis of CAD. High levels of PAI-I in Indians are reported in association with hypertriglyceridemia and hyperinsulinemia. This combination promotes thrombosis by impairing fibrinolysis [3]. Indian relevance of PAI-1 with other AMI marker was studied and showed with positive association with PAI-1 and AMI [1, 7, 8]. Plasma PAI-1 as an early marker of STEMI/AMI was reported a decade ago but no study has been reported from India, in particular, south India.

Aim and objectives

Aim of this cross sectional study are to ascertain plasma PAI-1 as an independent biochemical marker in STEMI, Quantification and Establishment of the indicative analytical range of Plasma PAI-1 in young south Indian ethnics with Acute Myocardial Infarction and to find out association of plasma PAI-1 with other risk factors of AMI.

MATERIALS AND METHODS

Study design

This cross-sectional study was conducted at Mahatma Gandhi Memorial hospital, Trichy, Study subjects includes 40 Patients (sample size calculated by conventional statistical formula) with acute myocardial infarction who had typical chest pain, shows electrocardiographic changes (ST Elevation) and a transient rise in cardiac enzymes to more than twice the upper, Age less than 45 years. Control subjects were 40 healthy men and women who came with some patients and healthy volunteers' age less than 40 years, during May 2016 to March 2017. Ethical clearance was obtained from K.A.P.V.Government Medical College, Trichy, Tamil Nadu.Exclusion criteria for this study was Case and Control Subjects With Renal Disease, Severe (Neuro) Psychiatric Problems, Known Diabetic Patients, Known Hypertensive Patients, Known History of Thromboembolic Disorders, Smokers, Alcoholic, History of Previous Coronary Artery Disease and Obese Individuals.

Sample collection

Under sterile condition, 4ml of peripheral venous blood was withdrawn using sterile disposable syringes from all the study subjects. 2 ml was transferred to another eppendorf tube, centrifuged at 2500 rpm for 20 minutes and plasma was separated and 500µl was stored in sterile eppendorf at -20^{0}C for PAI-1 (ELISA KIT-KOCH 3071) and the same was analyzed within six months of sample collection. Remaining blood was transferred to another eppendorf tube for the analysis of Serum Glucose, Serum Urea, Serum Creatinine, Serum Total Cholesterol, Serum Triacylglycerol, Serum High Density Lipoprotein, Serum Electrolytes and Serum Creatine Kinase-MB (CK-MB). Measurement of plasmaPAI -1 Antigen by ELISA (Catalog No.KHC3071 & KHC3072. Pub.No.MAN0014692 Rev 1.0).

RESULTS AND STATISTICS

The statistical analyses were done using SPSS-19.0. The biochemical parameters between AMI patients and controls were tested by using student's t test. Logistic regression analysis, Odds ratio with two tailed p values and 95% Confidence intervals (CI) were calculated. Level of significance for p-value was set at point <0.05 (If p <0.001 shows a strong significance). Pearson correlation was used to compare PAI -1 with other parameters of AMI. Levene's Test used to check Equality of Variances between patients and control.

Table-1: Minimum age, Maximum age, Mean age and SD of the patients and control groups

S.No	GROUP	Minimum Age	Maximum Age	Mean Age ± SD	P -value
1	Patients	18	40	36.48±5.26	≤ 0.61
2	Control	19	40	32.33± 6.09	

Table 2: Levene's Test for Equality of Variances for different risk factors of AMI with plasmaPAI-1

Variable	Group	Mean ± SD	Std. Error Mean	P value	95% Confidence Interval of the Difference	
					Lower	Upper
Systolic BP	Patients	106.97±9.1	1.59	≤ 0.906	4.778	5.38
	Control	106.67±11.3	1.97		4.7	5.38
Diastolic BP	Patients	74.24±8.3	1.44	≤ 0.871	4.0	3.39
	Control	74.55± 6.6	1.15		4.0	3.40
BMI	Patients	23.38± 1.9	0.34	≤ 0.205	0.32	1.48
	Control	22.80± 1.67	0.29		0.3	1.48
Urea	Patients	36.91± 6.19	1.07	≤ 0.001	2.7	8.662
	Control	31.18± 5.73	0.99		2.79	8.663
Sugar	Patients	126.42± 8.04	6.62	≤ 0.003	8.51	36.762
	Control	103.79±14.22	2.47		8.35	36.918
Creatinine	Patients	0.94± 0.16	0.02	≤ 0.443	0.10	0.0481
	Control	0.97± 0.14	0.025		0.10	0.0481
APTT	Patients	25.33±6.17	1.065	≤ 0.513	1.53	3.04
	Control	24.58±2.45	0.42		1.5	3.0
Sodium	Patients	138.79± 3.40	0.59	≤ 0.547	3.16	9.3
	Control	135.69±17.68	3.07		3.2	9.46
Pottasium	Patients	3.967±0.43	0.07	≤ 0.240	0.12	0.23
	Control	3.912±0.28	0.04		0.12	0.23
T.Cholesterol	Patients	168.39±29.00	5.04	≤ 0.532	21.4	5.46
	Control	176.39±25.67	4.46		21.4	5.4
TGL	Patients	169.03±59.07	10.28	≤ 0.532	23.9	45.9
	Control	158.03±81.46	14.18		24.0	46.0
VLDL	Patients	33.80±11.81	2.05	≤ 0.532	4.7	9.1
	Control	31.60±16.29	2.8363		4.8	9.2
LDL	Patients	95.89± 24.36	4.24	≤ 0.320	21.45	7.11
	Control	103.0± 33.07	5.75		21.48	7.14
HDL	Patients	38.70±3.86	0.67	≤ 0.007	4.9	1.134
	Control	41.73±3.84	0.66		4.9	1.134
CK-MB	Patients	138.58±108.0	18.80	≤ 0.01	214.2	204.8
	Control	133.24±192.7	63.18		217.7	208.4
PAI-1	Patients	3450.7±1406.6	244.87	≤ 0.001	907.93	2061.52
	Control	1966.03±878.7	152.97		7324907.93	2063.67
Prothrombin Time	Patients	16.94± 13.19	2.29	≤ 0.090	0.578	8.669
	Control	12.89± 1.65	0.287		0.663	8.754

Table-3: Multivariate Analysis of Plasma PAI-1 with various risk factors of AMI among south Indian Ethnics

GROUP	Variables	P -value	95.0% Confidence Interval for B	
			Lower Bound	Upper Bound
Patients	SUGAR	≤ 0.698	12.499	18.385
	UREA	≤ 0.057	2.540	162.829
	AST	≤ 0.927	22.343	24.456
	ALT	≤ 0.983	16.349	16.005
	HDL	≤ 0.475	89.647	187.391
	CKMB	≤ 0.16	1.094	9.906
Control	SUGAR	≤ 0.836	27.634	22.533
	UREA	≤ 0.526	98.882	51.784
	AST	≤ 0.121	58.995	7.292
	ALT	≤ 0.044	1.441	91.825
	HDL	≤ 0.358	48.729	130.064
	CKMB	≤0.589	0.834	0.483

Table 4: Shows significant association of risk factors between patients and Control in relation to plasma PAI-1 Mann-Whitney U and Wilcoxon W- rank tests (P≤0.05 is significant).

	SBP	Sex	DBP	BMI	Urea	Sugar	Creat-nine	PT	APTT	Na^+	K^+	T.Chole sterol	TGL	VLDL	LDL	HDL	CK-MB	PAI-1
Mann-Whitney U	528	379	541	436	279.5	387.5	450	477	534.5	532	455	456	443	443	454	338	91	205
Wilcoxon W	1089	940	1102	997	840.5	948.5	1011	1038	1095	1093	1016	1017	1004	1004	1015	899	652	766
Z	0.22	2.74	0.04	1.39	3.4	2.01	1.2	0.87	0.12	0.161	1.15	1.13	1.3	1.3	1.16	2.66	5.8	4.35
P value ≤	0.82	0.006	0.96	0.16	0.001	0.04	0.2	0.38	0.89	0.872	0.24	0.25	0.19	0.19	0.2	0.007	0.001	0.001

DISCUSSION

Plasma PAI-1 is an early marker of STEMI/AMI was reported a decade ago but no study has been reported from India, in particular, south India. The variation in the association of PAI- 1 on AMI has been observed in ethnic, age and gender groups. Therefore, there is a need to assess the relationship between carriage PAl-1 production, and the development of STEMI in Indian patients aged less than 45 years. Consequently, the quantification of PAI 1 in plasma is essential for early diagnosis and prevention of fatal complications of AMI in young south Indians. Hence this study is more relevant in the context of region wise data creation to support nationalized early diagnostic and therapeutic measures of AMI.

In some studies from India, the percentage of patients below the age of 40 years suffering from acute myocardial infarction (AMI) is reported as high as 25-40% [1, 7]. The risk-factor evaluation must start earlier. Along with the traditional risk factors, the 4G allele is an independent risk factor for the appearance of STEMI in the patients under 45 years of age. The detection of this allele along with other risk factors may, therefore, be useful in primary prevention [8].

The Mean and SD of plasma PAI-1 for patients and Controls are 3450.76±1406.68 and 1966.03±1406.68 (Table 4.2) and there was a positive significance variation of plasma PAI-1 ($P \leq 0.001$) observed between patients and controls. Further, a positive significant changes between patients and controls in Serum Urea: $P \leq 0.001$, Serum Glucose: $P \leq 0.04$, Serum HDL: $P \leq 0.007$, Serum CK-MB: $P \leq 0.001$, Plasma PAI-1: $P \leq 0.001$ were also observed (Table 2). The review of literatures showed that this is the first study involving the south Indian population to evaluate the association of Plasma PAI-1in STEMI in patient's ≤40 years of age. Inconsistency in PAI-1 plasma concentrations has been reported in different ethnic groups around the world [9] and it was 5000pg/ml for Indian euthenics. Festa *et al.* [10] reported the ethnic differences in the distribution of 4G/5G polymorphism to be a determining factor in the plasma concentration of PAI-1. In some case plasma PAI-1 appears to be governed by environmental factors such as smoking [11] along with certain components of metabolic syndrome such as obesity and the insulin concentration and dyslipidemia [12] or the interaction between smoking and this syndrome [13,14] which also contribute. However, interaction with other, traditional risk factors is almost certainly involved in the development of a STEMI and it is important to identify them if primary prevention from early in life is to be improved. In the present work, plasma PAl-1 concentrations measured in post-STEMI were highest in those with AMI subjects as reported by Serrano Rios *et al.* in patients with metabolic syndrome. Such increases in PAl-1 have been associated with AMI, Panahloo *et al.* report that they can remain high for six months [15]. These findings have the same opinion with the proposal of Sobel *et al.* [16] that the over expression of PAI-1 leads to a reduced smooth muscle fiber content in atherosclerotic plaques, inducing a reduction in the amount of collagen and extracellular matrix proteins, a reduction in resistance to atheroma, the development of a vulnerable plaque, and its eventual breakage and consequent AMI.

Further, an increased concentration of PAI-1 favors a state of hypofibrinolysis via the inhibition of tPA and therefore a reduction in the transformation of Plasminogen into plasmin, a key enzyme in the regulation of the fibrinolytic system. It might, therefore, be hypothesized that the 4G allele is associated with high concentrations of PAI-1 and accordingly with two mechanisms that favor the onset of an AMI. The formation of vulnerable plaques and a reduction in fibrinolysis. This could be of particular interest in explaining the pathophysiological mechanisms behind STEMI in young patients. The distribution of age of patients and control subjects detailed in table 4.1 and values are Mean Age and SD of Patients: 36.48 ±5.26 ,Mean Age of Controls: 32.33 ± 6.09, Minimum and maximum age for patients:18 and 43 years, Minimum and Maximum age for controls:19 and 42 years.

Levene's Test for Equality of Variances in table 2 shows details of positive association of various risk factors for AMI with plasma PAI-1 of between patients and control subjects with its Mean, SD and P values. Mean and standard deviation of Serum Glucose for patients and Controls were` 126.42±38.04 and 103.79 ±14.22 ($P \leq 0.04$), Mean and standard deviation of Serum Urea for patients and Controls are 36.91±6.19 and 31.18±5.73 ($P \leq 0.001$), Mean and standard deviation of Serum HDL for patients and Controls 38.70± and41.73 ±3.843 ($P \leq 0.007$),Mean and standard deviation of serum CK-MB for patients and Controls 138.58±108.014 and 133.24± 192.761 (≤ 0.01), Mean and standard deviation of plasma PAI-1 for patients and Controls 3450.76±1406.68 and1966.03±1406.68 ($P \leq 0.001$).After adjusting with all other risk factors of AMI which shows positive significance in Levene's Test for Equality of Variances in table 2. Consequently Serum urea shows positive significance in multivariate assay (Table 3).

CONCLUSION

This study confirmed an association of Plasma PAI-1and STEMI Indian euthenics. PAI-1 might play an important role in the pathogenesis of CAD. This study ascertained plasma PAI-1 as an independent biochemical marker for the Indian young AMI patient's age less than 45 years. The study established an analytical range of plasma PAI-1 in Indian young patients AMI was 3000-5000 pg/ml (Normal is 1000-2000 pg/mg). It also showed an inverse association of PAI-1 antigen levels with the HDL-cholesterol levels. Hence, it is concluded that the increased PAI-1 associated with increased risk of AMI. The study confirmed that there won't be any association between and PAI-1 antigen levels in control subjects, whereas, there is a positive association between plasma PAI-1 antigen level in AMI patients. Hence it is concluded that the causality of increased plasma PAI-1 itself is an important cause of AMI. Along with the traditional risk factors, the Furthermore, this study anticipated correlation plasma PAI-1 with other risk factors of MI and found a positive relationship with Urea, HDL, and glucose. There was no significant connection between PAI-1 in Sex, Serum AST, ALT, Creatinine, PT, APTT Serum sodium and Potassium levels. The other important conclusion is that there was no connection between CK-MB and PAI-1.

REFERENCES

1. Hastie, N., Wrights, A. (2007). Genes in common diseases. Cambridge University Press, New York. Sep 10.
2. Mannucci, P. M., Bernardinelli, L., Foco, L., Galli, M., Ribichini, F., Tubaro, M., & Peyvandi, F. (2005). Tissue plasminogen activator antigen is strongly associated with myocardial infarction in young women. *Journal of Thrombosis and Haemostasis*, *3*(2), 280-286.
3. Hamsten, A., Walldius, G., Szamosi, A., Blombäck, M., Faire, U., Dahlén, G., ... & Wiman, B. (1987). Plasminogen activator inhibitor in plasma: risk factor for recurrent myocardial infarction. *The Lancet*, *330*(8549), 3-9.
4. Alexander, T., Victor, S. M., Mullasari, A. S., Veerasekar, G., Subramaniam, K., Nallamothu, B. K., & TN-STEMI Programme Investigators. (2013). Protocol for a prospective, controlled study of assertive and timely reperfusion for patients with ST-segment elevation myocardial infarction in Tamil Nadu: the TN-STEMI programme. *BMJ open*, *3*(12), e003850.
5. Kraus WE. Genetic approaches for the investigation of genes associated with coronary heart disease. American heart journal. 2000 Oct 1;140(4):S27-35.
6. Nordt, T. K., Lohrmann, J., & Bode, C. (2001). Regulation of PAI-1 expression by genetic polymorphisms: impact on atherogenesis. *Thrombosis research*, *103*, S1-S5.
7. Dennis, L. (2005) Kasper, mdeugene braunwald, mdanthony s. Fauci m, editor. Harrison's principles of internal medicine. 16th ed. Mcgraw-hill.
8. Reddy, K. S., & Yusuf, S. (1998). Emerging epidemic of cardiovascular disease in developing countries. *Circulation*, *97*(6), 596-601.
9. Ye, S., Green, F. R., Scarabin, P. Y., Nicaud, V., Bara, L., Dawson, S. J., ... & Arveiler, D. (1995). The 4G/5G genetic polymorphism in the promoter of the plasminogen activator inhibitor-1 (PAI-1) gene is associated with differences in plasma PAI-1 activity but not with risk of myocardial infarction in the ECTIM study. *Thrombosis and haemostasis*, *73*(03), 837-841.
10. Festa, A., D'Agostino Jr, R., Rich, S. S., Jenny, N. S., Tracy, R. P., & Haffner, S. M. (2003). Promoter (4G/5G) plasminogen activator inhibitor-1 genotype and plasminogen activator inhibitor-1 levels in blacks, Hispanics, and non-Hispanic whites: the Insulin Resistance Atherosclerosis Study. *Circulation*, *107*(19), 2422-2427.

11. Eliasson, M., Asplund, K., Evrin, P. E., & Lundblad, D. (1995). Relationship of cigarette smoking and snuff dipping to plasma fibrinogen, fibrinolytic variables and serum insulin. The Northern Sweden MONICA Study. *Atherosclerosis*, *113*(1), 41-53.
12. Henry, M., Tregouet, D. A., Alessi, M. C., Aillaud, M. F., Visvikis, S., Siest, G., ... & Juhan-Vague, I. (1998). Metabolic determinants are much more important than genetic polymorphisms in determining the PAI-1 activity and antigen plasma concentrations: a family study with part of the Stanislas Cohort. *Arteriosclerosis, thrombosis, and vascular biology*, *18*(1), 84-91.
13. Burzotta, F., Di Castelnuovo, A., Amore, C., D'Orazio, A., Di Bitondo, R., Donati, M. B., & Iacoviello, L. (1998). 4G/5G promoter PAI-1 gene polymorphism is associated with plasmatic PAI-1 activity in Italians: a model of gene-environment interaction. *Thrombosis and haemostasis*, *79*(02), 354-358.
14. Naran, N. H., Chetty, N., & Crowther, N. J. (2008). The influence of metabolic syndrome components on plasma PAI-1 concentrations is modified by the PAI-1 4G/5G genotype and ethnicity. *Atherosclerosis*, *196*(1), 155-163.
15. Panahloo, A., Mohamed-Ali, V., Lane, A., Green, F., Humphries, S. E., & Yudkin, J. S. (1995). Determinants of plasminogen activator inhibitor 1 activity in treated NIDDM and its relation to a polymorphism in the plasminogen activator inhibitor 1 gene. *Diabetes*, *44*(1), 37-42.
16. Sobel, B. E., Woodcock-Mitchell, J., Schneider, D. J., Holt, R. E., Marutsuka, K., & Gold, H. (1998). Increased plasminogen activator inhibitor type 1 in coronary artery atherectomy specimens from type 2 diabetic compared with nondiabetic patients: a potential factor predisposing to thrombosis and its persistence. *Circulation*, *97*(22), 2213-2221.
17. Murray, C. J., & Lopez, A. D. (1997). Alternative projections of mortality and disability by cause 1990–2020: Global Burden of Disease Study. *The Lancet*, *349*(9064), 1498-1504.