Original Research Article

High-sensitivity C-reactive protein and interleukin-6 as risk predictors in patients with stable angina pectoris

Shekhar Kunal¹, Pradeep Kumar Meena¹*, Pooja Pathak², Himanshu Mahla¹, Kashish Gupta³, Vijay Pathak¹

¹Department of Cardiology, SMS Medical College, Jaipur, Rajasthan, India
²Department of General Internal Medicine, Midland Regional Hospital Port Laoise, County Laois, Republic of Ireland
³Department of Medicine, SMS Medical College, Jaipur, Rajasthan, India

Received: 26 July 2020
Revised: 17 August 2020
Accepted: 19 August 2020

*Correspondence:
Dr. Pradeep Kumar Meena,
E-mail: drpkmeena@yahoo.com

Copyright: © the author(s), publisher and licensee Medip Academy. This is an open-access article distributed under the terms of the Creative Commons Attribution Non-Commercial License, which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

ABSTRACT

Background: Cardiovascular diseases are a leading cause of morbidity and mortality especially in developing countries such as India. Biomarkers such as high-sensitivity C-reactive protein (Hs-CRP) and interleukin-6 (IL-6) can help in risk stratification and better management of patients with stable angina.

Methods: This was a prospective observational study wherein symptomatic patients with stable angina were enrolled. Coronary angiogram was done in those consenting to the procedure. Severity of coronary stenosis was graded as per the modified Gensini score (mGS). Hs-CRP and IL-6 levels were determined pre-procedure and 24 hours post percutaneous coronary intervention (PCI). Based on angiographic profile, patients were subdivided into four groups: group 1: normal coronaries, group 2: single vessel disease, group 3: double vessel disease and group 4: triple vessel disease. Primary outcome was occurrence of major adverse cardiovascular events over one-year period.

Results: A total of 158 patients completed the study with a mean age of 62.8±9.6 years. A significant difference was observed between the four groups in terms of age, Hs-CRP and IL-6 levels. Of the 124 patients undergoing PCI, significant difference was observed in terms of pre and post procedure Hs-CRP (P<0.0001) and IL-6 levels (P<0.0001). Strong positive correlation was seen between Hs-CRP and IL-6 levels with modified Gensini scoring (mGS). Patients with MACE (15/158; 9.4%) had significantly higher levels of Hs-CRP and IL-6. Multivariate logistic regression analysis revealed that Hs-CRP, IL-6, ΔHs-CRP and ΔIL-6 were independent predictors of major adverse cardiovascular events (MACE).

Conclusions: Hs-CRP and IL-6 levels were independent predictors of outcomes and can be used for risk stratification in these patients.

Keywords: Angina pectoris, Biomarkers, Coronary angiogram, Coronary artery disease, Interleukin-6, High sensitivity C-reactive protein

INTRODUCTION

Cardiovascular disease (CVD) is one of the leading cause of death worldwide accounting for nearly one-third of all the deaths.¹ In India, CVD accounts for nearly a quarter (24.8%) of all deaths with a majority of them (52.4%) occurring below 70 years of age.² Atherosclerosis, the underlying pathology responsible for CVD, is a chronic inflammatory process.³ It has been seen that atherosclerosis is often characterized by the presence of a low-grade inflammation which alters the endothelial lining of the coronary arteries.⁴ This is associated with an increase in the level of various inflammatory markers such as acute phase proteins and cytokines. Previous studies
have suggested that inflammation both at local and systemic levels has an important role to play in the destabilization and rupture of atherosclerotic plaques leading to acute coronary events. Recently the focus has shifted from a risk factor based assessment to the use of blood based biomarkers of inflammation in order to improve the risk stratification and determine patient groups who would readily benefit from a particular treatment strategy. Among these, high sensitivity C-reactive protein (Hs-CRP), a prototype marker of the inflammatory process, has been the most studied factor both for the causation as well as the prediction of CVD. In addition, cytokines such as interleukin-6 (IL-6) plays an important role in the chronic inflammatory process and development of atheromatous plaques.

Hs-CRP and IL-6 both are pleiotropic cytokines with a wide impact on cellular and humoral immune response thus serving as marker of inflammation, host defence and tissue injury. IL-6 is a proinflammatory cytokine which has stimulatory effects on T- and B-lymphocytes and leads to the synthesis of acute-phase proteins such as CRP and fibrinogen. In addition, IL-6 leads to increased production of major chemoattractant protein-1 which is a major chemoattractant for monocytes and hence plays a role in lymphocyte activation. All of these suggest that both Hs-CRP and IL-6 can be a marker for inflammatory states in patients with stable angina.

This study aims to determine the serum levels of Hs-CRP and IL-6 in patients with stable angina pectoris and its correlation with severity of the disease. In addition, we also envisaged to determine the short-term prognostic significance of Hs-CRP and IL-6 pre and post coronary angioplasty in patients with stable angina pectoris.

**METHODS**

This was a single center prospective observational study carried over a two-year period in the Department of Cardiology, SMS Medical College, Jaipur. All patients >18 years diagnosed with chronic stable angina (Canadian Cardiovascular Society class 3 or more) which was defined as chest pain brought on by exertion and relieved on rest, with symptoms persisting despite optimal medical therapy. All the enrolled patients had a positive electrocardiogram (ECG) exercise stress test response (>1 mm ST-segment depression). The following patients were excluded: features suggestive of acute coronary syndrome (ACS) at screening or a history of ACS in previous three months; history of previous coronary artery bypass grafting, percutaneous coronary intervention, surgery or trauma in the past three months; valvular heart disease, heart failure or left ventricular ejection fraction (LVEF) <30%; chronic kidney disease, hepatic dysfunction, acute and chronic infections; anemia, peripheral vascular disease or history of autoimmune diseases; inability to provide informed consent or to follow-up after discharge; and co-existing conditions associated with a limited life expectancy of less than six months.

In all these patients, detailed clinical history including symptomatology, presence of CVD risk factors, family history and prior evidence of CVD were recorded. In addition, blood samples for routine hematological and biochemical parameters were collected on admission. Twelve lead electrocardiogram and echocardiographic assessment were carried out with LVEF being estimated using the bi-plane Simpson’s method. Coronary angiogram was performed in all patients consenting for the procedure and revascularization (percutaneous coronary intervention [PCI]) in those as deemed necessary by the treating physician based on clinical risk assessment and severity of the lesion. Standardized definitions of all patient-related variables and clinical diagnoses were used.

**Hs-CRP and IL-6 assessment**

Serum levels of Hs-CRP (diagnostics Biochem Canada Inc.) and IL-6 (dialclone immunoassay, France) were determined using the enzyme linked immunosorbent assay (ELISA) techniques. Commercially available ELISA kits were used for both Hs-CRP and IL-6 levels determination with the sensitivity of the Hs-CRP ELISA kit being 10 ng/ml and that of IL-6 being <2 pg/ml. In all these patients, after an overnight fast, 5 ml of venous blood sample was taken in the morning prior to the performance of an angiogram. Samples were allowed to form clot at room temperature following which they will be transferred to laboratory on ice and centrifuged at 3000 cycles/minute within half an hour. The supernatant from the centrifuge were divided into two aliquots and stored at -80°C until the time of analysis. In addition in all those patients undergoing PCI and consenting to be a part of the study, a repeat 5 ml of venous blood sample was withdrawn 24 hours post PCI.

**Angiographic analysis**

Angiographies were performed according to the standard Judkins technique in all patients post a written informed consent regarding the procedure. Images of the coronary tree were obtained in routine projections in all patients and reviewed separately by two experienced cardiologists who were unaware of the patient’s details including the results of immunoassays. Coronary stenosis was considered only if there was more than 50% reduction in luminal diameter of coronary artery. Based on the presence or absence of coronary stenosis in major coronary arteries, patients subdivided into four groups: group 1: those without CAD, if no coronary artery showed a reduction in lumen diameter ≥ 50%; group 2: those with single vessel disease (SVD), if stenosis was detected in only one coronary artery; group 3: those with double vessel disease (DVD), if stenosis was detected in two coronary arteries and; group 4: those with triple vessel disease (TVD), if stenosis was detected in three coronary arteries. The severity of the coronary artery disease was quantified using the modified Gensini scoring (mGS) system. This is an angiographic scoring system which determines the degree of coronary artery involvement based on the stenosis severity of eight major
coronary branches. These would include left main stem, left anterior descending, diagonal branch, 1st septal perforator, left circumflex artery, marginal or posterolateral branch, right coronary artery and main posterior descending artery. The scoring system is: 0 for no stenosis; 1 for 1-49% stenosis; 2 for 50-74% stenosis; 3 for 75-99% stenosis and 4 for total occlusion. Based on the degree of stenosis, all the vessel scores were summed and a total angiographic score between 0-32 was assigned to each individual.9

**Outcomes**

All the patients were followed up for a period of one year to determine major adverse cardiovascular events (MACE) which was defined as occurrence of either death, myocardial infarction, unstable angina or and any coronary revascularization (surgery and/or PCI). A written informed consent was obtained from all eligible patients prior to inclusion in the study which was approved by the institutional review board.

**Statistical analysis**

Continuous variables with a normal distribution were expressed as mean value and standard deviation while count variables were expressed as frequencies and percent values. The normality of distribution of data was assessed through the Shapiro-Wilks test. Statistical comparison of baseline characteristics was performed using the chi-square test for categorical variables while one-way analysis of variance (ANOVA) was used for the continuous variables. In addition, post-hoc analysis was performed by the Tukey multiple comparison tests. Correlation between Hs-CRP as well as IL-6 levels were performed by the Pearson product-moment correlation coefficients. Univariate and multivariate logistic regression analysis was used to assess the association between Hs-CRP and/or IL-6 levels with the outcome while adjusting for other potential confounders. In addition, receiver operating characteristic (ROC) curves were computed to relate the Hs-CRP and IL-6 levels with outcome. Area under the curve (AUC), or c-statistic was used as a measure of the predictive accuracy of these biomarkers with the diagnostic ability of classified as “excellent” if AUC, or c-statistic values were ≥0.9, “good” if AUC were ≥0.80, “fair” if AUC were ≥0.70 and “poor” if AUC were <0.7010. Data analysis was done using the Statistical Package for the Social Sciences (SPSS) software package version 24 (SPSS, Chicago, Illinois, USA). P values <0.05 shall be considered significant.

**RESULTS**

A total of 172 patients were enrolled in the study of whom 14 patients were excluded as a result 158 patients completed the study. The mean age of the patients was 62.8±9.6 years (range: 40-88 years) with a male predominance (57.6%). Hypertension was the most common co-morbidity seen in 51 (32.2%) patients followed by diabetes in 16 (26.5%) and dyslipidaemia in 32 (20.2%). The mean Hs-CRP levels were 3.44±1.00 mg/l in the study population while the mean IL-6 levels were 87.80±12.25 pg/ml. All the patients underwent coronary angiogram with normal epicardial coronaries reported in 15 (9.6%) subjects. SVD was the most common abnormality in 50 (31.6%) patients followed by triple vessel disease in 47 (29.7%) and double vessel disease in 46 (29.1%) subjects. Left anterior descending artery (LAD) was the most commonly involved coronary artery in 110 (39.9%) patients followed by right coronary artery (RCA) in 79 (28.7%), left circumflex artery (LCX) in 68 (24.3%) and left main coronary artery (LM) in 21 (7.1%) subjects. The mean mGS in the study population was 4.93±4.30 with 124/158 (78.5%) subjects undergoing PCI. Based on the coronary angiographic profile, patients were subdivided into four groups: group 1: normal epicardial coronaries (n= 15), group 2: SVD (n= 50), group 3: DVD (n= 46) and group 4: TVD (n=47) (Table 1).

**Table 1: Demographic parameters of all patients with stable angina (n=158).**

| Parameters                     | Patients |
|--------------------------------|----------|
| Mean age (in years)            | 62.8±9.6 (40-88) |
| Male/females                   | 91 (57.6%)/67 (42.4%) |
| Smoking                        | 65 (41.1%) |
| Hypertension                   | 51 (32.2%) |
| Diabetes mellitus              | 16 (26.5%) |
| Dyslipidemia                   | 32 (20.2%) |
| Hs-CRP (mg/l)                  | 3.44±1.00 (0.57-6.81) |
| IL-6 (pg/ml)                   | 87.80±12.25 (55.54-120.06) |
| Total cholesterol (mg/dl)      | 180.59±34.27 |
| Triglyceride (mg/dl)           | 148.06±70.9 |
| LDL cholesterol (mg/dl)        | 89.81±21.04 |
| HDL cholesterol (mg/dl)        | 54.03±11.76 |
| Coronary angiogram             | 158 (100%) |
| Lesion profile                 |           |
| Normal epicardial coronaries   | 15 (9.6%) |
| Single vessel disease          | 50 (31.6%) |
| Double vessel disease          | 46 (29.1%) |
| Triple vessel disease          | 47 (29.7%) |
| Vessels involved               |           |
| Left main                      | 21 (7.1%) |
| Left anterior descending       | 110 (39.9%) |
| Left coronary circumflex       | 68 (24.3%) |
| Right coronary artery          | 79 (28.7%) |
| Modified Gensini score         | 4.93±4.30 (0-32) |
| Undergoing percutaneous coronary intervention | 124/158 (78.5%) |

*HDL: high density lipoprotein; Hs-CRP: High-sensitivity C-reactive protein; IL-6: interleukin-6; LDL-C: low density lipoprotein; mg/dL: milligram per decilitre; mg/L: milligram per litre; pg/mL: picogram per millilitre
There was a significant difference between the four groups in terms of age, baseline Hs CRP and IL-6 as well as the mGS. Patients with TVD has significantly higher baseline levels of Hs-CRP and IL-6. No difference was observed between the three groups in terms of sex distribution, comorbidities such as hypertension or diabetes and smoking status. In addition, haematological and biochemical parameters such as haemoglobin, total leucocyte counts, blood urea, serum creatinine and electrolytes were similar between the four groups (Table 2). Post-hoc analysis revealed that there was a significant difference between all the groups in terms of Hs-CRP (group 1 versus 2: p<0.0001; group 1 versus 3: p<0.0001; group 1 versus 4: p<0.0001; group 2 versus 3: p<0.0001; group 2 versus 4: p<0.0001 and group 3 versus 4: p=0.008), IL-6 levels (group 1 versus 2: p<0.0001; group 1 versus 3: p=0.001; group 1 versus 4: p<0.0001; group 2 versus 3: p=0.0001; group 2 versus 4: p<0.0001 and group 3 versus 4: p=0.006) and mGS (group 1 versus 2: p=0.03; group 1 versus 3: p<0.0001; group 1 versus 4: p<0.0001; group 2 versus 3: p<0.0001; group 2 versus 4: p<0.0001 and group 3 versus 4: p<0.0001).

**Pre and post-procedure Hs-CRP and IL-6**

Of the 158 patients, PCI was done in 124 (78.5%) of them. There was a significant difference in terms of pre and post procedure Hs-CRP (pre-procedure: 3.60±0.86 versus post-procedure: 8.93±2.83; p<0.0001) and IL-6 levels (pre-procedure: 89.48±11.17 versus post-procedure: 142.89±18.91; p<0.0001). Both the post-procedure Hs-CRP and IL-6 levels were significantly different among the three groups with the highest levels reported in patients with TVD. In addition, the mean difference i.e. ΔHs-CRP (calculated as the difference between post-procedure and pre-procedure Hs-CRP) and ΔIL-6 (calculated as the difference between post-procedure and pre-procedure IL-6) were significantly higher among patients with TVD as compared to the other two groups (Table 2).

**Correlation: IL-6 and Hs-CRP levels**

There was a very strong positive correlation seen between baseline Hs-CRP and IL-6 levels [Figure 1] which was highly significant (r=0.978 [95% CI: 0.970-0.984]; p<0.0001) and a strong correlation between baseline Hs-CRP and modified Gensini score (r=0.754 [95% CI: 0.678-0.814]; p<0.0001) as well as baseline IL-6 and modified Gensini score (r=0.748 [95% CI: 0.670-0.809]; p<0.0001) (Figure 1, 2a and 2b).

**Outcomes**

MACE was observed in 15/158 (9.4%) patients with a significantly higher frequency seen among diabetics and patients with prior history of stroke. Patients with MACE had significantly higher levels of baseline Hs-CRP and IL-6. In addition, among those patients who underwent PCI and had MACE, post-procedure Hs-CRP and IL-6 as well as Δ Hs-CRP and Δ IL-6 levels were significantly higher than those without MACE (Table 3). A ROC curve was plotted to evaluate the predictive ability and cut-off values of baseline Hs-CRP and IL-6 levels as well as that for Δ Hs-CRP and Δ IL-6 levels for prediction of MACE (Figure 3 and 4). ROC analysis showed that the AUC for Hs-CRP was 0.70 (p=0.01) while that of IL-6 was 0.67; (p=0.03). The cut-off levels of Hs-CRP for prediction of MACE was 3.4 mg/l with a sensitivity of 80% and a specificity of 52.3%. Similarly, the cut-off levels of IL-6 were 85.86 pg/ml with a sensitivity of 73.3% and a specificity of 51.7%. The AUC values for Δ Hs-CRP was 0.741 (p=0.003) and that for Δ IL-6 levels was 0.761 (p=0.001). The cut-off levels of Δ Hs-CRP was 6.09 with a sensitivity of 80% and a specificity of 72.5% while that for Δ IL-6 levels was 50.1 pg/ml with a sensitivity of 80% and a specificity of 52.3% (Figure 3).

Univariate logistic regression analysis showed that TVD (OR: 1.49; 95% CI: 0.91-1.95; p=0.05), mGS (OR:1.15; 95% CI:1.05-1.26; p=0.002), serum creatinine (OR: 1.40; 95% CI: 1.15-5.54; p=0.001), baseline IL 6 (OR: 1.06; 95% CI: 1.04-1.11; p=0.015), baseline Hs CRP (OR: 2.42; 95% CI: 1.30-4.50; p=0.005 ), post procedure Hs-CRP (OR: 1.41; 95% CI: 1.13-1.76; p=0.002), post procedure IL-6 (OR: 1.26; 95% CI: 1.1-1.42; p=0.002), Δ Hs-CRP (OR-1.54; 95% CI: 1.17-2.03; p=0.002) and Δ IL-6 (OR: 1.19; 95% CI: 1.1-1.35; p=0.002) were independent predictors of MACE. Multivariate logistic regression analysis revealed that baseline Hs-CRP, baseline IL-6, ΔHs-CRP and ΔIL-6 were independent predictors of MACE (Table 4).
Table 2: Comparison of demographic, clinical and biochemical parameters between the groups.

| Parameters                          | Normal coronaries (n=50) | Single vessel disease (n=46) | Double vessel disease (n=47) | Triple vessel disease (n=47) | P-value |
|------------------------------------|--------------------------|-----------------------------|-----------------------------|-----------------------------|---------|
| Age                                | 56.6±7.4                 | 61.9±9.9                    | 65.4±9.5                    | 63.4±9.2                    | 0.015   |
| Male                               | 7 (46.6%)                | 26 (52%)                    | 30 (65%)                    | 28 (59.5%)                  | 0.46    |
| Female                             | 8 (53.4%)                | 24 (48%)                    | 16 (35%)                    | 19 (40.5%)                  | 0.46    |
| Smoking                            | 4 (26.6%)                | 18 (36%)                    | 23 (50%)                    | 20 (42.5%)                  | 0.37    |
| Hypertension                       | 4 (26.6%)                | 14 (28%)                    | 20 (43.4%)                  | 13 (27.6%)                  | 0.20    |
| Diabetes Mellitus                  | 1 (6.6%)                 | 11 (22%)                    | 9 (19.5%)                   | 11 (23.4%)                  | 0.31    |
| Dyslipidemia                       | 4 (26.7%)                | 6 (12%)                     | 11 (23.9%)                  | 11 (23.4%)                  | 0.16    |
| Stroke                             | 0(0%)                    | 2 (3.1%)                    | 1(2.1%)                     | 3 (6.3%)                    | 0.14    |
| MACE                               | 0                        | 1 (2%)                      | 6 (13%)                     | 8 (17%)                     | 0.009   |
| Baseline IL-CRP (mg/l)             | 1.82±0.72                | 2.92±0.70                   | 3.73±0.63                   | 4.20±0.73                   | <0.0001 |
| Baseline IL -6 (pg/ml)             | 70.49±7.19               | 81.25±8.80                  | 90.97±8.52                  | 97.17±9.93                  | <0.0001 |
| Modified Gensini Score             | 0                        | 2.8±1.1                     | 5.4±2.1                     | 8.3±5.7                     | 0.0001  |
| Total blood sugar (mg/dl)          | 13.8±1.9                 | 13.5±1.6                    | 13.7±1.8                    | 13.6±2.0                    | 0.88    |
| Platelet count (lakh/mm3)          | 9.5±3.1                  | 9.8±1.9                     | 12.8±1.9                    | 12.8±1.9                    | 0.80    |
| Haematocrit (%)                   | 44.4±5.2                 | 44.7±4.1                    | 45.2±5.0                    | 46.4±5.1                    | 0.99    |
| Serum Creatinine (mg/dl)           | 36.8±16.3                | 33.7±14.6                   | 32.1±10.4                   | 40.1±11.1                   | 0.25    |
| Serum sodium (mEq/l)               | 1.2±0.3                  | 1.1±0.3                     | 1.1±0.2                     | 1.2±0.4                     | 0.40    |
| Serum potassium (mEq/l)            | 4.4±0.2                  | 4.3±0.4                     | 4.2±0.4                     | 4.3±0.4                     | 0.98    |
| Post procedure Hs-CRP (mg/l)       | 6.1±1.4                  | 9.7±1.8                     | 11.3±2.3                    | <0.0001                     |
| Post procedure IL -6 (pg/ml)       | 124.4±12.7               | 148.6±10.8                  | 157.9±14.0                  | <0.0001                     |
| ∆Hs-CRP (mg/l)                     | 3.1±1.1                  | 5.9±1.5                     | 7.1±1.8                     | <0.0001                     |
| ∆IL-6 (pg/ml)                      | 42.8±9.7                 | 57.5±8.5                    | 61.1±9.3                    | <0.0001                     |

*MACE: major adverse cardiovascular event; mEq/L: milli-equivalent per litre;

Table 3: Comparison of the parameters between MACE and no MACE sub-groups.

|                      | MACE (n=15) | No MACE (n=143) | P-value |
|----------------------|-------------|-----------------|---------|
| Mean age (in years)  | 63.86 ± 10.61 | 62.76 ± 9.52   | 0.673   |
| Sex (males)          | 10/15(66.7%) | 81/143(56.6%)  | 0.455   |
| Smoking              | 5/15(33.3%)  | 60/143(41.9%)  | 0.518   |
| Hypertension         | 4/15(26.7%)  | 47/143(32.8%)  | 0.625   |
| Diabetes Mellitus    | 6/15(40%)    | 26/143(18.19)  | 0.045   |
| Dyslipidemia         | 1/15(6.6%)   | 31/143(21.7%)  | 0.169   |
| History of stroke    | 2/15(13.3%)  | 4/143(2.8%)    | 0.042   |
| Pre Hs-CRP (mg/l)    | 4.16±1.07    | 3.36±0.97      | 0.001   |
| Pre IL-6 (pg/mL)     | 95.28±13.38  | 87.01±11.90    | 0.012   |
| Post Hs-CRP* (mg/l)  | 11.23±2.88   | 8.61±2.68      | 0.003   |
| Post IL-6* (pg/ml)   | 157.74±18.68 | 140.84±18.09   | 0.001   |
| ∆ hs-CRP* (mg/l)     | 7.07±2.17    | 5.08±2.11      | 0.001   |
| ∆ IL-6* (pg/ml)      | 62.45±11.33  | 52.11±11.79    | 0.002   |

Table 4: Independent predictors of MACE: multivariate logistic regression analysis.

|                      | Odds Ratio | 95% Confidence Interval | P-value |
|----------------------|------------|-------------------------|---------|
| Baseline IL-6 (pg/mL)| 2.81       | 1.56-4.46               | 0.023   |
| Baseline Hs-CRP (mg/L)| 3.84     | 1.23-5.32               | 0.012   |
| Triple vessel disease| 0.90      | 0.21-3.89               | 0.891   |

Continued.
## DISCUSSION

Hs-CRP and IL-6 have been extensively evaluated in patients with acute coronary syndrome (ACS), however data regarding its role in patients with stable angina pectoris seems to be conflicting. The findings in our study showed that both Hs-CRP and IL-6 increased with an increase in severity of CAD. In addition, baseline levels of both Hs-CRP and IL-6 were independent predictors of MACE. This highlights the putative role of these inflammatory biomarkers in patients with stable angina and its impact on outcomes. Although uncertainty exists in few studies regarding Hs-CRP as a marker of severity in patients with stable angina, case-control and cross-sectional studies have shown a strong association. However, a meta-analysis clearly indicated that even after the correction of a number of important factors in heart disease, Hs-CRP remained a risk factor and a powerful predictor of CV events. The role of Hs-CRP as a prognostic marker has been highlighted in the studies by Sinning et al which showed that Hs-CRP has some prognostic benefit however, it adds little to the risk stratification compared with the traditional risk factors. Similarly, a sub study of the prevention of events with angiotensin-converting enzyme inhibition (PEACE) trial comprising 3771 patients showed that a baseline Hs-CRP >1mg/L was associated with significantly higher risk of cardiovascular death, myocardial infarction (MI) and stroke. Mokhtar et al proposed a cut-off level of >5 mg/L for Hs-CRP for prediction of MACE (sensitivity: 93.7% and specificity: 100%). In this study, baseline Hs-CRP >3.4 mg/L had a sensitivity of 80% and a specificity of 52.3% to predict MACE. In patients with stable angina, elevated IL-6 levels occur due to release from various cells.

### Table

| Variable                  | Odds Ratio | 95% Confidence Interval | P-value |
|---------------------------|------------|-------------------------|---------|
| Modified Gensini score    | 1.03       | 0.85-1.25               | 0.721   |
| Serum Creatinine (mg/dL)  | 1.11       | 0.89-2.01               | 0.041   |
| Δ Hs-CRP (mg/L)           | 1.84       | 1.17-2.42               | 0.011   |
| Δ IL-6 (pg/mL)            | 1.55       | 1.13-1.82               | 0.032   |

### Figures

- Figure 2: Scatterplot showing correlation between baseline (a) Hs-CRP and modified Gensini scores, (b) IL-6 levels and modified Gensini scores.
- Figure 3: Receiver operating curve analysis for Hs-CRP and IL-6 as a predictor of MACE.
- Figure 4: Receiver operating curve analysis for ΔHs-CRP and ΔIL-6 as a predictor of MACE.
including the vascular smooth muscle cells as well as the macrophage and the foam cells in atheromatous plaques. In the study by Mokhtar et al, a significant increase in serum level of IL-6 was seen in patients with stable CAD as compared to non-CAD patients. This finding was consistent with those of Sarrafzadegan et al, Tang et al and Caselli et al who too had reported higher concentrations of IL-6 in patients with stable angina. In addition, IL-6 levels also reflect the degree of vascular inflammation and the severity of CAD. Mokhtar et al found significant increase in serum level of IL-6 in MVD and SVD patients as compared with non-CAD patients, a finding seen in our patients too. This finding is in accordance with that of Liu et al who too had reported higher serum levels of IL-6 in patients with multivessel disease as compared with control group. In our study, we observed a significant correlation with IL-6 levels and mGS, a finding reported by Tanidi et al. However, Mohktar et al found no association between IL-6 levels and the Gensini score.

Prognostic significance of Hs-CRP and IL-6 pre and post angioplasty

Acute inflammatory response is an initial consequence of PCI due to the exposure of the thrombogenic surface of the vessel wall to circulating leucocytes and through the recruitment of inflammatory cells from the overstretched adventitia. PCI especially stent implantation stimulates the arterial intimal cellular proliferation and extracellular matrix synthesis which is mediated largely by inflammatory processes. In addition, stent deployment can cause a foreign body reaction further amplifying the inflammatory response. It is still not clear in stable CAD patients whether the magnitude of increase in Hs-CRP post PCI is an independent prognostic marker. In the study by Gach et al, of the 89 stable CAD patients treated by PCI, MACE was observed in 36 patients. Multivariate analysis reported that a prior history of myocardial infarction and a significant increase in Hs-CRP post PCI (P=0.004 and 0.003, respectively) were independent predictors of MACE. In this study, the authors found that a significant increase in Hs-CRP post PCI (ΔHs-CRP) was more predictive of MACE than levels of Hs-CRP pre and post PCI. In our study, both baseline Hs-CRP levels and increase post PCI (ΔHs-CRP) were independent predictors of MACE.

Previous studies have shown a significant association between pre-procedural CRP levels and subsequent cardiac events in patients treated with bare metal stent (BMS). However, the predictive role of Hs-CRP for adverse outcomes in patients implanted with mixed BMS and drug-eluting stent (DES) or only DES implantations has been controversial. In a study among 513 patients undergoing non-urgent PCI, high levels of Hs-CRP was associated with a greater risk of periprocedural MI but not mortality. Xu et al investigated the role of Hs-CRP level both at admission and follow-up in 303 patients with stable angina to determine its predictive value for in-stent restenosis (ISR). The authors concluded that plasma Hs-CRP levels both at admission and on follow-up were independent predictors of ISR in stable CAD patients with DES implantation. Similarly, a meta-analysis comprising six studies reported that high levels of Hs-CRP were associated with an increased risk of ISR. However, its role as a predictive tool for MACE post PCI is less clear. Hojo and colleagues had for the first time demonstrated a correlation between the rise of IL-6 concentration post PCI and the risk of late restenosis. However, a limited sample size (32 subjects) was one of the major study limitations. In a study among 216 patients with stable CAD undergoing elective PCI, baseline IL-6 levels were not predictive of ISR. In our study, we found both baseline IL-6 levels as well as increase in IL-6 levels post PCI (ΔIL-6) to be independent predictors of MACE. We also for the first time proposed a cut-off level of IL-6 as well as ΔIL-6 as a predictor of MACE.

One of the important limitations of our study was that it was a single center study with a limited sample size. Since, we had enrolled only patients with stable angina (CCS class III or more) who were symptomatic despite being on GDMT over a short enrollment period, our sample size was small. Another limitation to our study was a short duration of follow-up. Multiple multicentric studies are needed in order to support the hypothesis regarding the role of Hs-CRP and IL-6 as one of the risk predictors in stable CAD patients.

CONCLUSION

A strong quantitative correlation was observed between increased Hs-CRP and IL-6 levels with future major adverse cardiac events in a population with stable angina. Quantitative determination of these biomarkers were found which might contribute to a better risk stratification over and above the traditional risk factors in these patients. These biomarkers can be developed as a prognosis indicator post PCI however, there is a need of larger multicentric studies to further validate the cut-offs values of Hs-CRP and IL-6 proposed by us.

Funding: No funding sources Conflict of interest: None declared Ethical approval: The study was approved by the Institutional Ethics Committee

REFERENCES

1. Roth GA, Johnson C, Abajobir A, Abd-Allah F, Ahera SF, Abyu G, et al. Global, regional, and national burden of cardiovascular diseases for 10 causes, 1990 to 2015. J Am Coll Cardiol. 2017;70:1-25.
2. Reddy KS, Yusuf S. Emerging epidemic of cardiovascular disease in developing countries. Circulation. 1998;97:596-601.
3. Lusis AJ. Atherosclerosis. Nature. 2000;407:233-41.
4. Madjid M, Willerson JT. Inflammatory markers in coronary heart disease. Br Med Bull. 2011;100:23-38
5. Danesh J, Wheeler JG, Hirschfield GM, Eda S, Eiriksdottir G, Rumley A, et al. C-reactive protein and other circulating markers of inflammation in the prediction of coronary heart disease. N Engl J Med. 2004;350:1387-97.
6. Li H, Sun K, Zhao R, Hu J, Hao Z, Wang F, et al. Inflammatory biomarkers of coronary heart disease. Front Biosci (Schol Ed). 2018;10:185-96.
7. Roberts WL. Workshop on markers of inflammation and cardiovascular disease: application to clinical and public health practice: laboratory tests available to assess inflammation-performance and standardization: a background paper. Circulation. 2004;110(25):572-6.
8. Thompson DK, Huffman KM, Kraus WE, Kraus VB. Critical appraisal of four IL-6 immunoassays. PLoS One. 2012;7:30659.
9. Gensini GG. A more meaningful scoring system for determining the severity of coronary heart disease. Am J Cardiol. 1983;51:606
10. Youngstrom EA. A primer on receiver operating characteristic analysis and diagnostic efficiency statistics for pediatric psychology: we are ready to ROC. J Pediatr Psychol. 2014;39:204-21.
11. Peer A, Falkensammer G, Alber H, Kroiss A, Griesmacher A, Scharl M, et al. Limited utilities of N-terminal pro-B-type natriuretic peptide and other newer risk markers compared with traditional risk factors for prediction of significant angiographic lesions in stable coronary artery disease. Heart. 2009;95:297-303.
12. Hosseinsabet A, Mohebbi A, Almasi A. Association between C-reactive protein and coronary calcium score in coronary artery disease. Cardiovasc J Afr. 2009;20:107-11.
13. Lin T, Liu JC, Chang LY, Shen CW. Association of C-reactive protein and homocystein with subclinical coronary plaque subtype and stenosis using low-dose MDCT coronary angiography. Atherosclerosis. 2010;212:501-6.
14. He P, Xie XY, Ding YP, Chen XL. Correlation between high sensitive C-reactive protein, lipoprotein, blood uric acid and severity of coronary artery disease. Zhonghua Yi Xue Za Zhi. 2010;90:1989-91.
15. Masood A, Jafar SS, Akram Z. Serum high sensitivity C-reactive protein levels and the severity of coronary atherosclerosis assessed by angiographic gensini score. J Pak Med Assoc. 2011;61:325-7.
16. Bamberg F, Truong QA, Koenig W, Schlett CL, Nasir K, Butler J, et al. Differential associations between blood biomarkers of inflammation, oxidation, and lipid metabolism with varying forms of coronary atherosclerotic plaque as quantified by coronary CT angiography. Int J Cardiovasc Imaging. 2012;28:183-92.
17. Barbero U, D’Aschenzo F, Nijhoff F, Moretti C, Biondi-Zoccai G, Mennuni M, et al. Assessing risk in patients with stable coronary disease: when should we intensify care and follow-up? results from a meta-analysis of observational studies of the COURAGE and FAME era. Scientifica (Cairo). 2016:3769152.
18. Sinning JM, Bickel C, Messow CM, Schnabel R, Lubos E, Rupprecht HJ, et al; Atherogene investigators. Impact of C-reactive protein and fibrinogen on cardiovascular prognosis in patients with stable angina pectoris: atheroGene study. Eur Heart J. 2006;27:2962-8.
19. Sabatine MS, Morrow DA, Jablonski KA, Rice MM, Warnica JW, Domanski MJ, et al; PEACE investigators. Prognostic significance of the Centers for Disease Control/American Heart Association high-sensitivity C-reactive protein cut points for cardiovascular and other outcomes in patients with stable coronary artery disease. Circulation. 2007;115:1528-36.
20. Mokhtar ER, Saleh BI, Aboualia AM, Nagheb HM. Relationship between monocyte subsets, IL-6 and hs-CRP with the severity of coronary artery disease in stable angina pectoris patients. Am J Biochem. 2017;7:114-26.
21. Sarrafzadegan N, Sadeghi M, Ghaffarpasand F, Alisaeidi A, Sanei H, Zakeri H, et al. Interleukin-6 and E-selectin in acute coronary syndromes and stable angina pectoris. A comparative study. Herz. 2012;37:926-30.
22. Tang JN, Shen DL, Liu CL, Wang XF, Zhang L, Xuan XX, et al. Plasma levels of C1q/TNF-related protein 1 and interleukin 6 in patients with acute coronary syndrome or stable angina pectoris. Am J Med Sci. 2015;349:130-6.
23. Caselli C, De Graaf MA, Lorenzoni V, Rovai D, Marinelli M, Del Ry S, et al. HDL cholesterol, leptin and interleukin-6 predict high risk coronary anatomy assessed by CT angiography in patients with stable chest pain. Atherosclerosis. 2015;241:55-61.
24. Liu CL, Shen DL, Zhu K, Tang JN, Hai QM, Zhang JY. Levels of interleukin-33 and interleukin-6 in patients with acute coronary syndrome or stable angina. Clin Invest Med. 2013;36:234-41.
25. Tanindri A, Sahnarsslan A, Elbeg S, Cemri M. Relation between MMP-1, MMP-9, TIMP-1, IL-6 and risk factors, clinical presentation, extent and severity of atherosclerotic coronary artery disease. Open Cardiovascular Med J. 2011;5:110-6.
26. Gach O, Legrand V, Biessaux Y, Chapelle JP, Vanbelle S, Pierard LA. Long-term prognostic significance of high-sensitivity C-reactive protein before and after coronary angioplasty in patients with stable angina pectoris. Am J Cardiol. 2007;99:31-5.
27. Walter DH, Fichtlscherer S, Sellwig M, Auch-Schwelk W, Schächinger V, Zeiher AM. Preprocedural C-reactive protein levels and cardiovascular events after coronary stent implantation. J Am Coll Cardiol. 2001;37:839-46.
28. Dibra A, Mehilli J, Braun S, Hadamitzky M, Baum H, Dirschinger J, et al. Association between C-reactive protein levels and subsequent cardiac events among patients with stable angina treated with coronary artery stenting. Am J Med. 2003;114:715-22.

29. Hsieh IC, Chen CC, Hsieh MJ, Yang CH, Chen DY, Chang SH, et al. Prognostic impact of 9-month high-sensitivity C-reactive protein levels on long-term clinical outcomes and in-stent restenosis in patients at 9 months after drug-eluting stent implantation. PLoS One. 2015;10:0138512.

30. Herrmann J, Lennon RJ, Barsness GW, Sandhu GS, Gulati R, Best PJ, et al. High sensitivity C-reactive protein and outcomes following percutaneous coronary intervention in contemporary practice. Circ Cardiovasc Interv. 2012;5:783-90.

31. Xu YL, Li JJ, Xu B, Zhu CG, Yang YJ, Chen JL, et al. Role of plasma C-reactive protein in predicting in-stent restenosis in patients with stable angina after coronary stenting. Chin Med J (Engl). 2011;124:845-50.

32. Zhu X, Chen Y, Xiang L, You T, Jiao Y, Xu W, et al. The long-term prognostic significance of high-sensitive C-reactive protein to in-stent restenosis. Medicine (Baltimore). 2018;97:10679.

33. Chen SL, Liu Y, Lin L, Ye F, Zhang JJ, Tian NL. Interleukin-6, but not C-reactive protein, predicts the occurrence of cardiovascular events after drug-eluting stent for unstable angina. J Interv Cardiol. 2014;27:142-54.

34. Hojo Y, Ikeda U, Katsuki T, Mizuno O, Fukazawa H, Kurosaki K, et al. Interleukin 6 expression in coronary circulation after coronary angioplasty as a risk factor for restenosis. Heart. 2000;84:83-7.

35. Segev A, Kassam S, Buller CE, Lau HK, Sparkes JD, Connelly PW, et al. Pre-procedural plasma levels of C-reactive protein and interleukin-6 do not predict late coronary angiographic restenosis after elective stenting. Eur Heart J. 2004;25:1029-35.

Cite this article as: Kunal S, Meena PK, Pathak P, Mahla H, Gupta K, Pathak V. High-sensitivity C-reactive protein and interleukin-6 as risk predictors in patients with stable angina pectoris. Int J Adv Med 2020;7:1322-30.