Elevated Inflammatory Cytokines As A Predictor Of Restenosis Following Percutaneous Transluminal Coronary Angioplasty

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A. Statement of study and purpose rationale

In patients who have undergone successful percutaneous transluminal coronary angioplasty (PTCA), restenosis remains the limiting factor in patients long term event-free survival. The restenosis rate following PTCA is as high as 3001, with 950-o occurring within the first six months following the procedure. Clinical variables associated with increased risk of restenosis include male sex, cigarette smoking, diabetes mellitus, hypertension, hypercholesterolemia, end-stage renal disease. Anatomical and procedural variables associated with increased risk of restenosis included proximal stenosis, involvement of the left anterior descending artery, chronically occluded arteries, stenosis greater than 5 to 10 mm in length, and a residual stenosis of greater than thirty percent.

Within the coronary artery post-PTCA, neo-intimal hyperplasia and remodeling are processes thought to lead to restenosis. However, our knowledge of the mechanisms contributing to neo-intimal hyperplasia and remodeling is limited.

There is accumulating evidence that the immune system and inflammation play a significant role in atherosclerosis, as well as unstable angina and acute myocardial infarction. This evidence is available in multiple forms and involves both systemic and local evidence.

Studies have shown an increase white blood cell count to be an independent risk factor for ischemic heart disease. Several markers of inflammation have been shown to be elevated in patients with unstable angina and acute myocardial infarction. In patients with unstable angina, C-reactive protein levels above the 90th percentile are associated with an increased rate of ischemic events in hospital and an increased rate of myocardial infarction or the need for immediate revascularization.

Local evidence of the immune system and inflammation having a significant role in atherosclerosis and acute ischemic events also exists. Autopsy studies have demonstrated T-lymphocytes and macrophages in fatty streaks, the earliest form of coronary artery disease, as well in atheromatous plaques. Macrophages and T-lymphocytes have also been found within the site of plaque rupture in coronary arteries of patients who had died of an acute myocardial infarction. Further, these cells have been found to be in an activated state, as evidenced by detection of interleukin-2 (IL-2) and HLA-DR, suggesting that inflammation plays a role in plaque rupture. Furthermore, monocytes from patients with both stable and unstable angina secrete higher levels of cytokines, tumor necrosis factor (TNF), and interferongamma (IFN-α). Another study demonstrated that thrombin formation in unstable angina patients is caused by monocytes activated by lymphocytes, which are triggered by unknown factors.

The immune system and inflammation have also been implicated in playing a significant role in restenosis following PTCA. Smooth muscle cell migration and smooth muscle cell TPA, both of which are thought to play a role in restenosis, have been shown to augmented by IL-4 and attenuated by IFN-α. Also, a small study demonstrated that elevated of levels of soluble IL-2 receptor, a hallmark of activated T-lymphocytes, after PTCA may correlate with a failed procedure and/or restenosis.

Although some data does exist, the role of the immune system and inflammation in restenosis following successful PTCA has not been fully delineated. If elevated levels of cytokines are found in the serum of patients whose course is complicated by restenosis, these cytokines may serve as markers for restenosis. Furthermore, if these cytokines are found to be causally related to restenosis, inhibition of these cytokines may have significant therapeutic implications. The hypothesis of this study is that patients with unstable angina will have elevated levels of various cytokines, and failure to normalize cytokine

levels will be predictive of restenosis, the need for further revascularization and/or Q-wave myocardial infarction within the first six months after successful PTCA.

B. Description of study design and statistical analysis

Patients enrolled in this study will be consecutive patients admitted to Millstein Pavilion for unstable angina who subsequently undergo PTCA.

The primary outcome of this study will be evidence of clinically significant restenosis as evidenced by unstable angina and angiographically demonstrated restenosis. Restenosis will be defined as a greater than 50 percent narrowing of the diameter of the lumen at the site of previously successful PTCA as determined by the cardiologist. Successful PTCA will be defined as a luminal diameter that is narrowed by no more than 50 percent on the post-PTCA angiogram.

Levels of IL-4, soluble IL-2 receptor, and TNF-OLwill be measured from the serum of each patient collected prior to the procedure and again at one month and three months post-PTCA. Levels will again be measured at six months to determine if patients whose course had not been complicated by restenosis have elevated cytokine levels.

The secondary outcome of this study will be the combined outcome of restenosis, the need for repeat revascularization, or Q-wave myocardial infarction. A baseline electrocardiogram (EKG) will be obtained. An EKG will also be obtained at six months. Q-wave myocardial infarction will be defined as either patients admitted to the hospital with chest pain and EKG and cardiac enzyme evidence of an acute Q-wave myocardial infarction, or patients with new Q-waves on the their six month follow up EKG.

A total of 350 patients will be prospectively enrolled. The sample size is based on the assumption of a restenosis rate of 30%. and that cytokine levels can detect 75%. of the patients with restenosis, with a power of 80%. and type I error rate of 1%. These assumptions require a sample size of 156 patients. 19 patients will be added to account for patients lost to follow-up, for a total of 175 patients. The sample size will be doubled and the patients will be divided into two cohorts.

The first 175 patients will be used to determine the mean values, standard deviation, and range of soluble IL-2 receptor, IL-4, and TNF in patients with unstable angina prior to PTCA, as well as the mean values, standard deviation, and range of these cytokines in patients with and without restenosis following successful PTCA. These values will be used to create an algorithm for the prediction of restenosis in patients with unstable angina following successful PTCA. Once this algorithm is derived, the next 175 patients will be used to confirm the predictive algorithm prospectively. The data will be analyzed to determine the sensitivity and specificity of the predictive algorithm. The McNemar test will be used to compare differences between the groups of patients.

C. Description of study procedures

Blood samples will be withdrawn form the venous sheath already in place for the PTCA before the start of the procedure. Blood samples will be obtained by venipuncture at one month, three month, and six month follow-up appointments.

Approximately 30cc of blood will be drawn on each occasion. Each sample will be divided into locc tubes containing 1cc of 3.13 percent sodium citrate. The plasma will be separated by centrifugation at 3000 rpm for fifteen minutes and frozen at -70 degrees Celsius until assayed. The serum will be assayed for IL4, soluble IL-2 receptor, and TNF-α-using commercially available immunoassay kits.

D. Study Drugs

There will be no study drugs used for the purpose of this study.

E. Study devices

There will be no medical devices used solely for the purpose of this study.

F. Study questionnaires

There will be no study questionnaire used for the purpose of this study.

G. Description of study subjects and method of recruitment

350 consecutive patients 18 years old or older admitted to Millstein Pavilion with unstable angina who undergo successful PTCA will be enrolled. Unstable angina will be defined as at least one episode of chest pain occurring at rest, lasting less than 20 minutes, associated with EKG changes relating the chest pain to myocardial ischemia as determined by the admitting cardiologist, and the lack of evidence of an acute myocardial infarction on EKG and cardiac enzyme analysis.

Patients with a history of acute myocardial infarction within the previous four weeks documented by EKG and/or cardiac enzyme elevation will be excluded. Patients with prior coronary artery bypass grafts will be excluded. Patients with unstable angina secondary to restenosis of a vessel which had previously undergone PTCA or coronary stent placement will be excluded. Patients with evidence of systemic inflammation and/or cancer will be excluded.

Eligible patients will be approached by their cardiologist regarding enrollment. The study will be explained, and informed consent will be obtained.

Enrollment will not be limited by sex or race.

H. Confidentiality

All data in this study will be strictly confidential. Patients will be given unique identification numbers and all data will be stored in a secure location accessible only to the investigators.

I. Location of study

This study will be administered in the cardiac catheterization laboratory at Millstein Pavilion.

J. Risks and benefits

There will be no additional risk to the patient beyond that associated with PTCA and venipuncture. No immediate benefit will be gained by participation in the study. However, this study may enhance the understanding of restenosis, as well as identify serum markers associated with restenosis. This may ultimately have diagnostic, prognostic, and therapeutic implications.

K. Alternative therapies

This study does not include the use of experimental therapies.

L. Compensation and cost

The patients will incur no additional cost by participating in the study. There will be no compensation offered.

M. Minors and research subjects

This study will not enroll minors.

N. Radiation or radioactive substances

Patients will not be exposed to any additional X-rays beyond the normal amount of exposure during coronary angiography and PTCA.