Time course of early changes in plasma markers of collagen turnover following percutaneous transluminal coronary angioplasty

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INTRODUCTION: Marked changes occur in the collagen framework of the heart following acute ischemia, which is associated with adverse ventricular remodelling. Plasma markers of collagen turnover are useful in the assessment of remodelling and have predictive value, but their exact temporal dynamics following ischemia are unclear.

OBJECTIVE: To characterize the early temporal dynamics of plasma markers of collagen turnover in a human model of coronary artery occlusion.

METHODS: Fourteen patients undergoing elective percutaneous coronary intervention (PCI) to a single coronary artery were recruited in addition to a control group of eight patients undergoing elective diagnostic coronary arteriography. Sequential assessment of plasma levels of procollagen type I carboxyterminal propeptide and C-telopeptide for type I collagen (CITP) as markers of synthesis and degradation, respectively, was performed over a 16 h period.

RESULTS: The ischemic burden in the PCI group was high, with 13 of the 14 patients demonstrating transient ST segment shift or positive troponin. Mean plasma levels of CITP on admission were 3.1 ng/mL and 3.0 ng/mL in the PCI and control groups, respectively (P value nonsignificant). There was a sequential increase in plasma CITP following PCI, peaking at 4.7 ng/mL at 16 h ($P < 0.01$), with no change in the control group. There were no significant changes in plasma levels of procollagen type I carboxyterminal propeptide in either group.

CONCLUSIONS: Plasma levels of CITP demonstrated early temporal dynamics of collagen degradation following transient coronary artery occlusion supporting the use of plasma markers of collagen turnover as an early tool in the assessment of the remodelling process following myocardial ischemia.

Key Words: Angioplasty; Collagen; Remodelling

Évolution dans le temps des premiers changements affectant les marqueurs plasmatiques du renouvellement collagénique après une angioplastie coronarienne transluminale percutanée

INTRODUCTION : Des changements marqués affectent la structure du collagène du cœur après l'ischémie aiguë et sont associés à un remodelage ventriculaire nocif. Le dosage des marqueurs plasmatiques du renouvellement du collagène permettent d'évaluer le remodelage et possèdent une valeur prédictive, mais leur progression précise après l'ischémie reste à élucider.

OBJECTIF : Caractériser les premiers stades de cette progression des marqueurs plasmatiques du renouvellement du collagène dans un modèle humain d'occlusion coronarienne.

MÉTHODE : Les auteurs ont recruté 14 patients soumis à une intervention coronarienne transluminale percutanée (ICTP) élective ne touchant qu'une coronaire, en plus d'un groupe témoin de huit patients devant subir une artériographie coronarienne diagnostique élective. Des mesures séquentielles des taux plasmatiques du propeptide carboxyterminal du procollagène de type 1 et du C-télopeptide pour le collagène de type 1 (CITP), à titre de marqueurs respectifs de la synthèse et de la dégradation, ont été effectuées sur une période de 16 heures.

RÉSULTATS : Le fardeau ischémique dans le groupe soumis à l'ICTP a été élevé puisque 13 patients sur 14 ont présenté un changement transitoire du segment ST ou se sont révélés troponine-positifs. Les taux plasmatiques moyens de CITP à l'admission étaient de 3,1 ng/mL et de 3,0 ng/mL dans les groupes sous ICP et témoins, respectivement (valeur p non significative). On a noté une augmentation séquentielle des CITP plasmatiques après l'ICP, qui atteignait un pic de 4,7 ng/mL au bout de 16 heures ($p < 0,01$), sans modification dans le groupe témoin. On n'a noté aucun changement significatif des taux plasmatiques de propeptide carboxyterminal du procollagène de type 1 dans aucun des groupes.

CONCLUSIONS : Les taux plasmatiques de CITP ont manifesté une dynamique temporelle précoce de dégradation du collagène après une occlusion coronarienne transitoire, ce qui appuie l'utilisation des marqueurs plasmatiques du renouvellement collagène comme outil d'évaluation précoce du remodelage après ischémie myocardique.

Changes to the collagen framework of the heart occur following myocardial infarction, with as much as 25% net collagen loss within the first 24 h (1). These changes are associated with the development of ventricular remodelling (2,3), which is associated with an adverse prognosis (4-6).

Type I collagen is the most abundant subtype within the heart (7). Its metabolism can be assessed biochemically by analysis of plasma levels of procollagen type I carboxyterminal propeptide (PICP) (8,9) as a marker of synthesis, and C-telopeptide for type I collagen (CITP) as a marker of breakdown (10).

We have previously shown time-dependent changes in plasma levels of PICP and CITP following ST elevation myocardial infarction (11) and non-ST elevation acute coronary syndromes (12), and demonstrated

that these plasma markers of collagen turnover may be useful in the non-invasive assessment of left ventricular remodelling (13).

In these studies, the changes in plasma levels of CITP and PICP were apparent from admission, despite all patients being recruited within 6 h of the onset of symptoms, suggesting that collagen remodelling occurs within a few hours of the ischemic stimulus. As such, the remodelling process may be amenable to early biochemical assessment, but the exact early temporal dynamics of these plasma markers are unclear. To investigate this further, we examined the early time course of plasma levels of CITP and PICP in a human model of controlled acute coronary artery occlusion by recruitment of a cohort of patients undergoing percutaneous coronary intervention (PCI), and performed sequential blood sampling to assess temporal dynamics of plasma CITP and PICP.

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Figure 1) Pattern of change of plasma C-telopeptide for type I collagen (CITP) in percutaneous coronary intervention (PCI) and control groups demonstrated as mean absolute values at each time point (A) and mean percentage change from baseline (B). * $P < 0.05$; ** $P < 0.01$

METHODS

Patient population

Fourteen patients undergoing elective PCI to a single lesion in a single coronary artery in the regional cardiac catheterization laboratory were recruited for the study. Exclusion criteria included factors influencing collagen metabolism or elimination of propeptides, such as surgery within the past six months, hypertension, renal or hepatic dysfunction, in addition to factors making interpretation of the electrocardiogram difficult such as the presence of a permanent pacemaker system, left bundle branch block or concurrent digoxin therapy. All patients were naive to angiotensin-converting enzyme inhibitors or angiotensin II receptor blockers. To exclude changes in plasma markers of collagen turnover due to catheter manipulation in the aorta and coronary ostia or to the effects of intracoronary injection of contrast media, a group of eight patients undergoing day-case elective coronary angiography were recruited as a control group.

Blood sampling and biochemical analyses

To avoid the discomfort of repeated venepuncture, blood was drawn from an indwelling venous cannula. In the angioplasty group, six sequential samples were taken for the measurement of plasma PICP and CITP at serial time points – preprocedure, 1 min following first balloon inflation, at the end of the procedure, and 4 h, 6 h and 16 h following the procedure. In the control group, blood was drawn on only three occasions for the measurement of these propeptides – preprocedure, at the end of the angiogram and 6 h following the procedure. This was due to the fact that elective diagnostic coronary angiography was performed as a day case and, therefore, a 16 h sample would have been impractical. In addition, it was observed in previous studies (11-13) that changes in plasma markers of collagen turnover are apparent within 6 h of the onset of ischemic symptoms.

Plasma PICP and CITP were measured by commercially available radioimmunoassays (Orion Diagnostica, Finland) using previously validated techniques (10,14). In the laboratory at Stobhill Hospital (Glasgow, United Kingdom), intra-assay and interassay

variability are less than 4% and less than 7%, respectively. Troponin was measured by standard microparticle enzyme immunoassay (Bayer Corporation, USA).

Assessment of periprocedural ischemia

The presence of periprocedural ischemia was identified in the cohort undergoing PCI by continuous electrocardiographic monitoring in the catheterization laboratory and by the measurement of plasma troponin I at 16 h. A positive troponin (greater than 0.07 $\mu\text{g/L}$, greater than the 99th percentile of the normal population) or transient ST segment elevation or depression greater than 1 mm in two leads during balloon inflation was taken as evidence of induced ischemia.

Statistical analysis

Statistical analysis was performed using Minitab statistical software (Minitab Inc, USA). All continuous variables are expressed as mean $\pm$ SD. Results are also expressed as percentage change from baseline. Changes over time were investigated by repeated measures ANOVA. Biochemical variables were not normally distributed so they underwent log transformation before using a two-tailed Student's *t* test for comparison of means, although all data are presented in the nonlogarithmic format. $P < 0.05$ was considered to be significant. The present study was designed with 90% power to detect a change in CITP of 25% at 6 h compared with baseline and a 90% power to detect a 50% change in PICP at 6 h in the angioplasty group.

RESULTS

Baseline characteristics

The mean age of the PCI group was 60 years versus 65 years in the control group undergoing diagnostic angiography, with a similar percentage of male patients in both groups (64% versus 62.5%, respectively). In the PCI group, the culprit artery was the left anterior descending artery in six patients, the right coronary artery in five and the circumflex artery in the remaining three. Intracoronary stents were deployed in the majority of cases (12 of 14). The mean time of balloon inflation was 54 s with an average of 1.8 inflations per procedure. The acute ischemic burden in the PCI group was high. Ten patients had elevated troponin at 16 h indicating a degree of myocardial necrosis, although none had elevated creatine kinase. Nine patients had ST elevation or depression on the electrocardiogram (ECG) during balloon inflation. Only one patient had no evidence of ST segment shift and a negative troponin, and was classed as not showing evidence of periprocedural ischemia. No patient in the control group developed ECG changes.

CITP

Figure 1 demonstrates the change in CITP over time, plotted with absolute measurements of mean CITP (Figure 1A) and as percentage change from baseline (Figure 1B). The mean preprocedure CITP in the PCI group was 3.1 ± 0.27 ng/mL with a sequential rise in group mean CITP over time. The initial rise was rapid and at 6 h following balloon inflation, the mean CITP was above the normal range at 4.0 ± 0.33 ng/mL ($P < 0.05$). A continued rise was seen over time, reaching a peak of 4.7 ± 0.51 ng/mL ($P < 0.01$). The mean CITP in the control group was within the normal range at all time points (Figure 1A). With regard to percentage change from baseline, all patients but one in the PCI cohort demonstrated a sequential rise in plasma CITP, starting immediately following balloon inflation, continuing initially in an exponential fashion, with the rate of rise decreasing over time, heading toward a plateau at 16 h. Statistical significance was achieved at 6 h following inflation ($P < 0.05$), and the largest proportional rise was between 6 h and 16 h with the peak CITP at 16 h being an average of 48% higher than preprocedure values (Figure 1B) ($P < 0.01$). There was no significant change over time in the control group, with levels within 2 SDs of a normal population mean at all time points. There was also no significant change in plasma CITP in the patient with negative troponin and no ECG changes.

PICP

Changes over time of plasma PICP within the PCI group were initially heterogeneous, with some patients displaying an early rise following angioplasty and others showing an early fall in plasma PICP. The group mean initially rose above preprocedure values during the procedure, then fell below preprocedural values by 6 h, with a trough at 16 h (Figures 2A and 2B), although this did not reach statistical significance (143 ng/mL versus 113 ng/mL, $P=0.09$). There was no significant change over time in the control group.

DISCUSSION

Serological markers of collagen turnover have been successfully used as an assessment of collagen metabolism, due to changes in the equilibrium between synthesis and degradation in a variety of clinical arenas including hypertension (15,16), heart failure (17) and atrial fibrillation (18). However, these are stable, chronic conditions with alterations occurring slowly over a period of time in contrast to the rapid changes seen in acute ischemic events (11,12). Although the study numbers are small, the study was designed to provide insight into the early temporal dynamics of these markers and additional information to our previously published work in the setting of acute myocardial infarction (11,13) and non-ST elevation acute coronary syndromes (12).

The present study demonstrates the very early time-dependent changes in plasma levels of both PICP and CITP following myocardial ischemia caused by transient occlusion of a single epicardial coronary artery during PCI. No such changes were demonstrated in the control group undergoing routine angiography. The ECG changes and positive troponin in the PCI group demonstrate that transient occlusion of a coronary artery produced acute ischemia causing a level of necrosis that was sufficient to stimulate activation of the proteolytic enzymes responsible for collagen degradation.

The changes to plasma levels of PICP were heterogeneous within the group undergoing PCI, with no significant change in group mean within the first few hours. However, levels in the group decreased by 6 h with a trough at 16 h and mean values 18% below preprocedure levels. The heterogeneity of the plasma PICP response to ischemia may be a reflection of the wide normal range and SDs of this marker of collagen turnover. Also, it is important to remember that PICP reflects collagen synthesis, and it is collagen breakdown that predominates in the early stages following myocardial ischemia (1,2). As such, one might not expect there to be significant change in plasma levels of PICP in this timescale – a factor confirmed in previous clinical studies (11,12).

In contrast to PICP, the changes in plasma CITP were evident as early as 1 min following the first balloon inflation with further increases at each time point, reaching statistical significance at 6 h before rising further at 16 h at an average of nearly 50% higher than levels before the procedure. No such change was seen in the control

Figure 2) Pattern of change of plasma procollagen type I carboxyterminal propeptide (PICP) in percutaneous coronary intervention (PCI) and control groups demonstrated as mean absolute values at each time point (A) and mean percentage change from baseline (B)

group. Again, these data are entirely consistent with the pathological data on early collagen degradation following ischemia. Ischemia and infarction due to abrupt coronary artery occlusion produce changes in the extracellular matrix (1,19), causing increased production and activation of matrix metalloproteinases, the activity of which determines the extent of collagen breakdown (20-22). Matrix metalloproteinase gene expression is upregulated within a few hours of infarction (21), and the results of the present study suggest that plasma levels of CITP mirror the changes occurring at a tissue level and reflect the early remodelling process. Given the rapid, exponential rise of plasma CITP levels within a short time period, coupled with the fact that admission CITP values predict the subsequent development of wall motion abnormalities in ST elevation myocardial infarction (13), the present study may support the use of early biochemical assessment as a means to identify those at risk of subsequent adverse ventricular remodelling.

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