ence of potentially interfering substances was examined by adding known
tions to each of two serum pools with 3.24 and 6.43 mg/dL hsCRP. Intralipid
eride-equivalent concentrations of 600 and 1000 mg/dL caused biases of -
-6.5% for hsCRP and -3.4% to -8.6% for wrCRP. Ditauro-bilirubin at
tions of 20 and 50 mg/dL resulted in biases of 2.4% to 6.6% for hsCRP, and
9.0% for wrCRP. Hemolysate from red blood cells with hemoglobin
ations of 250 and 500 mg/dL resulted in biases of -0.8% to -3.8% for hsCRP.
% to -5.9% for wrCRP.

sion, the hsCRP assay had acceptable performance for measurement of low
centrations required for cardiac risk assessment. The wrCRP assay exhibited
formance at lower concentrations.

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ntibodies for dPAPP-A immunoassays

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cy associated plasma protein (PAPP-A) is a high-molecular weight
otein initially identified in blood of pregnant women as heterotetrameric (2:2)
x (htPAPP-A) with proform of eosinophil major basic protein (proMBP).
ly it was revealed that PAPP-A is also expressed in unstable atherosclerotic
s and can be used as a blood marker of adverse outcome in acute coronary
me (ACS) patients. It was shown that atherosclerotic form of PAPP-A was free
roMBP and had lower molecular weight than htPAPP-A. Thus it was suggested
APP-A from plaques as well as PAPP-A from blood of ACS patients is a
limeric form of PAPP-A (dPAPP-A). Immunoassays that do not discriminate
P-A and dPAPP-A are used now for dPAPP-A measurements in blood of ASC
ts. However, low but appreciable levels of htPAPP-A in normal and ACS
ts' blood could significantly influence dPAPP-A measurements by these non-
ic assays. Thus, design of novel dPAPP-A-specific immunoassay seems to be
romising for accurate marker quantification.

im of our study was to generate monoclonal antibodies (MAb) specific to
P-A and to design the prototype immunoassays suitable for dPAPP-A
rements in ACS patients' blood.

mice immunization with dPAPP-A purified from human atherosclerotic
ary vessels we have obtained nine hybridoma cell lines producing monoclonal
odies, specific to endogenous dPAPP-A and recombinant dPAPP-A (produced in
nalian cell line) and having no cross-reaction with htPAPP-A. All new
odies were tested in pairs with panel of PAPP-A antibodies able to recognize all
forms of PAPP-A - htPAPP-A, endogenous dPAPP-A and recombinant dPAPP-
ut of 300 tested two-site combinations two (pairs PAPP8-PAPP24 and PAPP52-
30) were selected for the development of dPAPP-A specific sandwich
moassays. In these assays one antibody (MAb PAPP8 or MAb PAPP30) was
fic to dPAPP-A (without cross-reaction with htPAPP-A), whereas another one
b PAPP24 or MAb PAPP52) was able to recognize all three forms of PAPP-A.
ction MAbs PAPP24 and PAPP30 were labeled with stable Eu(3+) chelate.

assays recognized endogenous and recombinant PAPP-A and showed very low
s-reactivity (< 1%) with htPAPP-A. The analytical detection limit of assays was
than 2 mIU/L, when purified recombinant dPAPP-A (HyTest, Finland) was used
calibrator. dPAPP-A level in plasma samples of 43 ACS patients (acute
cardial infarction, unstable angina, 3-20 hours after chest pain onset) and 34 non-
patients control group was measured. dPAPP-A level in plasma from ACS
ents being measured by PAPP8-PAPP24 and PAPP52-PAPP30 assays was 2.60
2.54-fold, respectively (p<0.01), higher than in plasma samples of control group.
clusions: here for the first time we are describing new generation of dPAPP-A
cific MAbs and new type of immunoassay suitable for direct measurement of
PP-A in ACS patients' blood.

Peripheral blood cells transcriptome to study new biomarkers for myocardial infarction follow up

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Introduction: The main clinical manifestation of atherosclerosis is the acute
myocardial infarction (MI), which is a medical emergency that requires a prompt
diagnosis and efficient therapy. New markers to predict MI have been scarcely
described in the last decade.
Objectives: A transcriptomic experiment was carried out to study the global gene
expression in peripheral blood cells (PBC) to study surrogate biomarkers for the early
diagnosis of myocardial infarction.
Methods: 5 MI patients and 6 healthy individuals were selected in a coronary
emergency facility. MI patients were followed up 8h after MI (T1), after angioplasty
(T2) and each 12 h during 48 h (T3 to T6). The control group (GC) was compose by
healthy individuals and this 5 MI after one year past of the event. RNA was extracted
by PAXgene system (Qiagen). PBC mRNA expression profiles were assessed by
Affymetrix GeneChip Human Exon 1.0 ST array. Signals were quantile-normalized
using iterPLIER algorithm, filtering (IQR, DABG and CROSS HYBR) and bayesian
adjusted t-statistics from the linear models for Microarray data (limma) package
interfaced on oneChannelGUI was applied to evaluate the differential gene
expression. The comparisons of interest where each time of MI versus GC, and the
evolution of the genes selected in the first moment during the time investigated. The
functional interpretation in terms of biological process, molecular interaction and
disease processes were performed using Ingenuity Systems program.
Results: After normalization and filtering were available 16869 genes. 25 genes were
differently expressed between MI T1 and GC with Benjamini & Hochberg adjusted p-
value < 0.05 and fold change 1.5, follow by 596, 138, 24 and 6 in T2, T3, T5 and T6,
respectively. No differences were found after 24 hours (T4).
Only 13 genes were common of the first comparison when T2 (after angioplasty) was
compared with CG and 3 genes after 12 h of MI (T3), which were involved in cellular
growth/proliferation and lipid metabolism. Nine genes involved with cell-mediated
immune response were selected only in T1.
Conclusion: The PBC transcriptome of MI follow up displayed significant variations
on expression of genes involved in immune-mediated response, growth/proliferation
and lipid metabolism. Immune response-related genes selected in this study may
predict the initial response to the MI lesion.

New insights into human proBNP processing: the evidence for furin-mediated proBNP processing in vitro.

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Brain natriuretic peptide (BNP) is a peptide hormone that acts to decrease systemic
vascular resistance and central venous pressure and to increase natriuresis. BNP is
secreted into the circulation by cardiomyocytes. Active BNP-32 hormone (32 amino
acid residues, a.a.r.) is formed from the precursor molecule, proBNP (108 a.a.r.),
along with N-terminal fragment (NT-proBNP; 76 a.a.r.), by specific enzyme cleavage.
The cleavage site of the proBNP molecule is located between amino acid residues
Arg76 and Ser77.
At present 2 proprotein convertases, furin and corin, are discussed as possible
candidates responsible for proBNP processing, but still it is not clear which of these
two enzymes is responsible for proBNP cleavage. The data obtained by our group