Poster Board P6-101

0554 THE GENERATION OF BRADYKININ IN CLINICAL BLOOD SAMPLES

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Kallikrein specifically cleaves bradykinin (BK) from high molecular weight kininogen. Although measurement of BK in blood samples is potentially a sensitive index of intravascular kallikrein action, the rapid inactivation of BK in the circulation ($t_{\frac{1}{2}}$=20 sec) limits the practical usefulness of BK assays. The present study was designed to develop a method which permits rapid measurement of circulating kallikrein in a large number of samples. Blood is collected in a mixture of kininase inhibitors and incubated for different periods of time before addition of soybean- and ovomucoid trypsin inhibitor. BK was measured by a radioimmunoassay. Cross-reacting kininogens were quantitatively separated from BK by precipitation with polyethyleneglycol. In 25 normal individuals this assay revealed a mean BK plasma level of 0.5 ng/ml±0.5(2SD) and the BK generation in vitro (ΔBK) of 1.5 ng/ml/20 min ±1.0 (2SD). Upon activation of plasma with kaolin or dextransulphate, BK increased up to 800 ng/ml/20 min. In 3 out of 5 patients with asymptomatic hereditary C1-INH deficiency, ΔBK was increased (range 7.0-11.1 ng/ml/20 min) whereas basal BK levels were normal. Increased ΔBK was found in 3 patients with pancreatitis and in 9 out of 26 patients with a malignancy (range 2.8-13.8 ng/ml/20 min) the basal BK plasma levels being normal. 2 of the latter patients showed signs of low grade DIC. It is thought that the BK-generation test detects circulating kallikrein.

P6-102

0555 EVALUATION OF THE BLOOD COMPATIBILITY OF BIOMATERIALS BY PLASMA KALLIKREIN DERIVED FROM CONTACT ACTIVATION

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Contact activation is considered as the crucial step to trigger blood coagulation on artificial surfaces. It is characterized by the conversion of proenzymes in plasma to active enzymes. Kallikrein is one of the proteinases which are generated in the course of contact activation. Consequently, its proteolytic activity can be used for monitoring the velocity of contact activation. This is demonstrated by recording the cleavage rate of the synthetic chromogenic substrate Bz-Pro-Phe-Arg-pNA. Polymer surfaces are known to be only weak activators of the contact phase. Therefore, the aim of the study was to elaborate a system sensitive enough to differentiate between various polymer surfaces. Two different approaches are shown to be effective:

1. In a one-step assay diluted plasma is incubated with the materials to be tested in the presence of the chromogenic substrate.
2. In a two-step assay whole blood or undiluted plasma is extensively incubated with materials before the residual contact activation capacity is determined by addition of ellagic acid.

The release of kallikrein is shown to be specifically surface mediated by using plasma deficient in single clotting factors and by its sensitivity against various inhibitors. The methods were applied to the evaluation of biomaterials of different shape like beads and tubings. The results correspond to those obtained with coagulation tests. Thus the method may provide a useful tool for the development of blood compatible biomaterials.

P6-103

0556 STUDIES ON COMPONENTS OF THE PLASMA KALLIKREIN-KININ SYSTEM IN NORMAL SUBJECTS AND PATIENTS WITH SEPTICEMIA

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Over the past decade evidence that the plasma kallikrein-kinin system is activated during septicemia has accumulated. We have used several tests, including newly developed chromogenic peptide substrate assays, to study components of this proteolytic enzyme system in plasma samples from normal subjects and patients with septicemia. The parameters studied were Hageman factor (HF), plasma kallikrein (KK), plasma prekallikrein (PKK), high molecular weight kininogen (HMwK), functional kallikrein inhibition (KKI), C^1-esterase inhibitor (CIINH), α_2-macroglobulin (α_2M) and α_1-antitrypsin (α_1AT). In samples from patients with fatal sepsis, levels of HF, PKK, HMwK, α_2M and KKI were all markedly reduced and spontaneous KK activity was detected. CIINH and α_1AT levels were much higher than normal. In plasma samples from three patients with septicemia who subsequently recovered mean plasma levels of PKK, KKI, HMwK and CIINH were higher than in the samples from the patients who died. Our results emphasize that the plasma kallikrein-kinin system becomes activated during septicemia and suggest that functional kallikrein inhibition contributes to survival.