

Platelet - Collagen Interaction

Purcell Room

Time
14.00

0384 PLATELET MEMBRANE COMPONENTS INVOLVED IN ADHESION TO COLLAGEN FIBERS.

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In an attempt to identify platelet membrane components involved in collagen recognition, rabbit platelets suspended in medium containing 1mM EGTA were passed through sepharose columns containing immobilized collagen fibers. The percent of platelets retained was proportional to the amount of collagen on the column but independent of the platelet concentration applied. Platelets that did not adhere in a single passage were still capable of adhering. Saturation of the column was not achieved. The adhering platelets were lysed with 0.5% triton X-100 and the proteins retained on the collagen were solubilized in 0.3% SDS. Polyacrylamide gel electrophoresis revealed at least 4 bands in the SDS fraction (and in whole platelets solubilized in SDS) that were absent from the triton lysate. None of these bands was of the major glycoproteins. Iodination of the platelet membranes did not interfere with their adhesion to the column. When iodinated platelets were subjected to the treatment described above, the SDS fraction contained one iodinated band not present in the triton lysate and another markedly enriched relative to its amount in the lysate. It is suggested that more than one platelet membrane component, or a complex consisting of several different components, is involved in platelet adhesion to collagen fibers.

14.15

0385 AMINO ACID SEQUENCE OF A PEPTIDE FROM TYPE III COLLAGEN INVOLVED IN PLATELET ADHESION

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The probable importance of the role of type III collagen in the initiation of thrombosis is due to its localization in the subendothelial layers of the vessel wall. The adhesion of platelets to type III has been therefore quantified by a method based on the filtration of non-adhesive ¹⁴C 5HT-labelled platelets through a Sepharose 2B column.

Type III collagen was purified from calf skin by pepsin extraction and salt precipitation. Type III collagen was cleaved by cyanogen bromide and the adhesion induced by the resulting peptides was measured. The activity was attached to the central alpha 1(III) CB4 peptide which was further cleaved by hydroxylamine, chymotrypsin and trypsin. An adhesive potency was linked to three fragments (HA 1 obtained by hydroxylamine, C2 by chymotrypsin and T2 by trypsin) which possess a common portion of 9 amino-acids, localized in the central part of the alpha 1(III) CB4 peptide and of the entire alpha 1(III) chains, which probably represents the active part of type III collagen. Its activity could be due to a particular sterical conformation linked to the presence of 3 imino acid residues in the sequence Gly-Lys-Hyp-Gly-Glu-Hyp-Gly-Pro-Lys of this fragment.