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Use of fibrillar collagens in the study of extracellular matrix: analysis of protein-protein and cell-protein interactions

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Collagen is the main protein of connective tissue in animals and the most abundant protein in mammals. In order to better understand formation, morphology and other functions of extracellular matrix, we studied the interactions of fibrillar collagens with a number of proteins.
One aspect of our approach deals with the binding features of type I and type II collagens with three small leucine-rich proteoglycans (SLRPs), decorin, fibromodulin and biglycan. We performed solid-phase assays (ELISA) in different conditions, by using: non pathological and pathological collagens, N-acetylated and N-methylated collagens and CNBr peptides.
The results demonstrated that the triple helical conformation of the collagenous samples is essential for the interaction; the binding of collagens to SLRPs has a ionic nature; collagen Lys/Hyl residues are fundamental since their N-acetylation inhibits the binding to SLRPs. In addition, we have found that type I collagen has multiple binding sites for SLRPs; in fact, at least four CNBr peptides from this collagen have the capacity to interact with the three SLRPs. Type II collagen probably shares this feature. The affinity constants of collagens/CNBr peptides with SLRPs is in the nanomolar range (1, 2, 3).
In this work we have also analyzed the role of fibrillar collagens (and their chemically modified forms) in megakaryocyte (MK) spreading and proplatelet formation (PPF). We found that adhesion of MKs to type I collagen inhibited PPF, but not MK spreading; on the contrary type III collagen was permessive for PPF. Furthermore, in presence of type I collagen, we assisted to translocation of endogenous fibronectin to cell periphery. This process was substrate-dependent: in fact, the exposure of endogenous fibronectin to the cell membrane, determined by immunofluorescence staining, occurred on MKs spreaded on collagen I, but not on fibrinogen. The interaction of MK with type I collagen seemed to be influenced by its biochemical properties: in fact, chemical N-acetylation of collagen I reduced fibronectin interaction and permitted PPF, while completely inhibited MK spreading (4).

References:

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