

Fibronectin unfolding plays a major role in fibrils formation and anastellin binding

Delphine Gourdon, Michael L. Smith, Sheila L. Morris, Radmila Vukmirovic, Kris E. Kubow,
Viola Vogel

Laboratory for Biologically Oriented Materials, Department of Materials

Swiss Federal Institute of Technology, ETH Zurich, Switzerland

Smith *et al.* recently showed that a broad range of fibronectin (FN) conformations was present within the elastic FN fibrils forming the extracellular matrix (ECM), and that tension exerted by cells on their surrounding ECM induced the loss of both quaternary and tertiary structures of around half of the FN molecules forming the fibrillar network [1].

Here we investigate the physiological implications of such a broad range of FN conformations present within the matrix, in particular the crucial role of cell-induced unfolding of individual type III FN modules (in addition of quaternary changes) both in the assembly of new FN matrix by cells, i.e., fibrillogenesis and in the binding of cancer ‘drugs’ proteins such as anastellin (AN). We used a fluorescence resonance energy transfer (FRET)-based technique as an indirect indication of FN conformation to address whether newly incorporated FN and AN molecules were affected by the local conformation of the pre-existing matrix. Cells were grown for 24 hours in the presence of FRET-labeled FN (double labeled with Alexa Fluor® 488 and Alexa Fluor® 546), and subsequently the media was exchanged to contain either new FN or AN labeled with a third color (Alexa Fluor® 633) to map the location of newly incorporated

molecules of within the FRET-labeled pre-existing matrix.

Our findings reveal that cell-induced FN unfolding is required for FN fibrillogenesis as well as for anastellin binding which then affects irreversibly mechanotransduction processes involved in the matrix.

[1] Smith *et al.* PLoS Biology, 5, e268
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