International Symposium on Polyelectrolytes
From Basic Science to Biological Applications
Research Workshop of the Israel Science Foundation
Ein Gedi, Israel, January 20-23, 2014

Heparin Binds Growth Factors and Oligolysines by Different Electrostatics

Dubin Paul, Minsky Burcu Baykal, Kaltashov Igor A.

Chemistry, UMass, Anherst, MA, USA

The presence of multiple protein binding sites on single heparin chains could allow them to bind non-covalently to oligopeptide components of tissue scaffolds while retaining growth factor affinity. However, the extent to which oligopeptide coupling interferes with cognate protein binding is unknown. To elucidate such simultaneous specific and non-specific interactions, we examined a well-defined ternary system comprising acidic fibroblast growth factor (FGF-1) and tetralysine (K_4), with a heparin (Hp) decamer (dp10) acting as non-covalent coupler. Electrospray ionization mass spectrometry was used to assess binding affinities and complex stoichiometries as a function of ionic strength *I* for K_4-dp10 and FGF-dp10. Formation of K_4-dp10 is qualitatively consistent with binding driven by the release of condensed counterions previously suggested for native heparin with divalent oliogpeptides (Mascotti, D. P.; Lohman, T. M. *Biochemistry* **1995**, *34*, 2908-2915); whileFGF binding displays more salt resistance and complex ionic strength dependence more consistent with screened electrostatics. Hp dp10 (5 nm length) can bind two FGF's, but notably only a single two nm K_4. This "specificity" of K_4 binding arises from its affinity for central oligoheparin units with higher concentrations of condensed counterions (Minsky, B. B.; Atmuri, A.; Kaltashov, I. A.; Dubin, P. L. *Biomacromoleculs* **2013**, *14*, 1113-1121). While K_4-heparin complexes vanish when the bulk salt concentration exceeds the local concentration of condensed counterions, FGF binding persists to higher ionic strength. Thus, K_4 and FGF with different modes of binding do not compete for dp10, and the ternary K_4-dp10-FGF complex is readily abundant. Observation of the compositional distribution of free and FGF-bound dp10 suggest a correlation between heparin degree of sulfation and FGF affinity, indicating a more nuanced approach than the use of heterogeneous native heparin for growth factor sequestration and release.