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Title: Biophysical Characterization of Extracellular Domains 1-4 of E- and N-cadherins and Designing Protein Molecular Weight Ladder for SDS-PAGE

Authors: Nivsarkar, Vaishnavi Niraj (/jspui/browse?type=author&value=Nivsarkar%2C+Vaishnavi+Niraj)

Keywords: Biology
Biophysics
Protein

Issue Date: 12-Jul-2017

Publisher: IISER-M

Abstract: We describe the expression and partial characterization of four of the five extracellular domains of both human E- and N-cadherins (i.e. E1-E2-E3-E4 and N1-N2-N3-N4). Both proteins are aggregation-prone with the bulk of the expressed population found in inclusion bodies. However, we have developed a method for recovery using partial denaturing conditions and partial refolding of these forms into soluble entities using on-column refolding and dialysis. Here we describe the structural characterization of these constructs using Circular dichroism and fluorescence spectroscopy. As these are calcium dependent adhesion molecules, we observe the changes in conformation upon Ca²⁺ binding and report the binding affinity for Ca²⁺-cadherin complex. The tendency of these constructs to adhere and form aggregates was also studied using dynamic light scattering.

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