Title:	Investigation of DNA breathing using time resolved emission spectroscopy and Spectroscopic determination of calcium binding affinity of cadherin-23
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Abstract:	DNA breathing is partial opening of double strands into single strands followed by closing into double stranded DNA again due to breakage and formation of H-bonds between the base pairs. HU is a bacterial histone like protein which binds to DNA and as it wraps the DNA it changes the DNA breathing frequency. To determine DNA breathing period time resolved emission spectroscopy (TRES) and time resolved area normalized emission spectroscopy (TRANES) have been used. For ds-DNA correlated TRNAES intensity variation of donor and acceptor have been observed and with addition of HU to ds-DNA, TRANES intensity variation decreases in frequency. But intensity variation has been monitored for small time scale (0-1 ns) due to short lifetime of fluorophores. To get complete information about DNA breathing fluorophores having longer life time will be used in further study. Cadherin-23, a transmembrane, calcium dependent protein, present in the tip-link in inner ear helps in mechanotrunsduction in hearing. We have spectroscopically determined calcium binding affinity of cadherin-23 using competitive chelator method. Four calcium binding affinity values of cadherin-23 are 1.2nM-1, 79.4 μM-1, 45 μM-1 and 40.2μM-1. Cadherin-23 has one tryptophan at 66th position on its backbone. With addition of calcium ions tryptophan fluorescence reveals two stage transition which matches with the MD simulation results which also shows two-stage structural changes of cadherin-23 with addition of calcium ions.
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