

# CONFORMATIONAL ANALYSIS OF ELECTROSTATIC INTERACTIONS

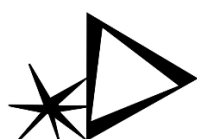
## IN AN INTRINSICALLY DISORDERED PROTEIN

(Short title)

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Electrostatic interactions play important roles in the functional mechanisms exploited by intrinsically disordered proteins (IDPs). The atomic resolution description of long-range and local structural propensities that can both be crucial for the function of highly charged IDPs present significant experimental challenges. Here we investigate the conformational behavior of the delta subunit of RNA polymerase from *Bacillus subtilis* whose unfolded domain is highly charged, with 7 positively charged amino acids followed by 51 acidic amino acids. Using a specifically designed analytical strategy, we identify transient contacts between the two regions using a combination of NMR paramagnetic relaxation enhancements, residual dipolar couplings (RDCs), chemical shifts and small angle scattering. This strategy allows the resolution of long-range and local ensemble averaged structural contributions to the experimental RDCs, and reveals that the negatively charged segment folds back onto the positively charged strand, compacting the conformational sampling of the protein while remaining highly flexible in solution. Mutation of the positively charged region abrogates the long-range contact, leaving the disordered domain in an extended conformation, possibly due to local repulsion of like-charges along the chain. Remarkably, *in vitro* studies show that this mutation also has a significant effect on transcription activity, and results in diminished cell fitness of the mutated bacteria *in vivo*. This study highlights the importance of accurately describing electrostatic interactions for understanding the functional mechanisms of IDPs.



SPEKTROSKOPICKÁ SPOLEČNOST JANA MARKA MARCI



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