

Non-Specific Interaction between DNA and Protein allows for Co-operativity: A Case Study with Mycobacterium DNA Binding Protein

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Supplementary Methods:

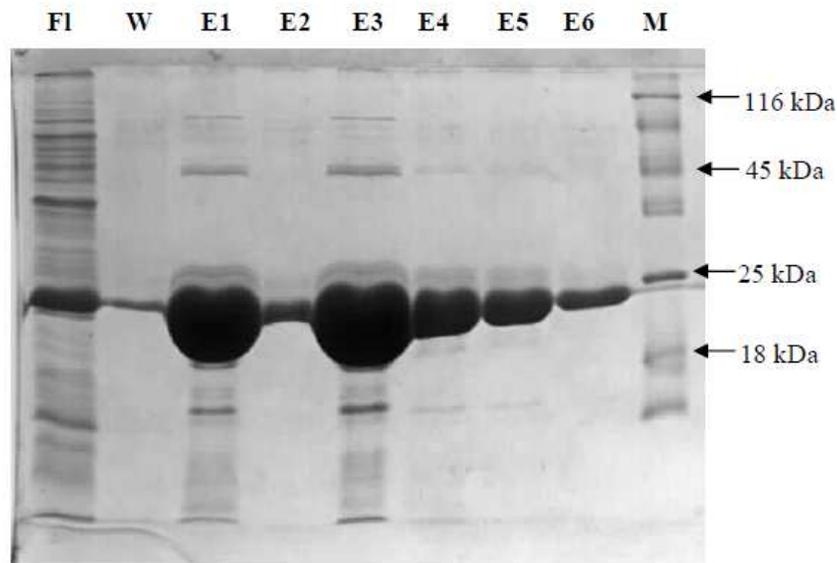
Preparation of Glass Slides and Silicon Slides and Deposition of Langmuir-Blodgett Films

The glass slides (18 mm x 19 mm, Marienfeld, Germany) and silicon slides (10 mm x 10 mm) were cleaned in a 3:7 (v/v) mixture of 30% H₂O₂ and 95% H₂SO₄ for 1 h. They were then rinsed thoroughly with Milli-Q and stored under water until they were used. They were sonicated in Milli-Q water prior to transferring the monolayers. This ensured a hydrophilic transfer of the monolayer components onto the glass/silicon slides (Supplementary Figure S2). The Langmuir-Blodgett films were transferred at a uniform dipper speed of 2 mm/min and the transfer ratio was monitored to be greater than 0.8 in all cases.

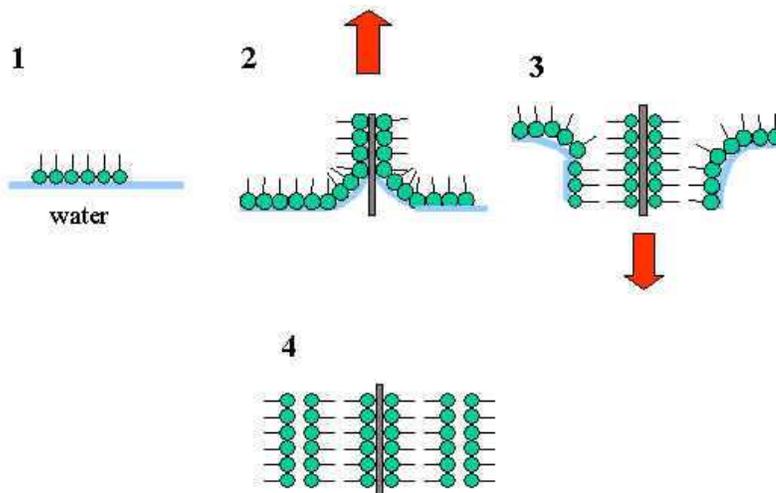
FTIR Spectroscopy:

The LB monolayers containing the immobilized components were transferred onto silicon slides which were used as solid substrate for our FTIR measurements. All the FTIR measurements were done in a Perkin-Elmer FTIR spectrometer, Spectrum 1000, where the incident rays are normal to the sample. The resolution of the spectra was 4 cm⁻¹. The spectra for LB films of the NiA-His-MsDps1 were obtained over the range of 1600 to 1700 cm⁻¹ and from 1500-1800 cm⁻¹ for NiA-His-MsDps1–DNA LB films. Minimally 75-100 monolayers were transferred at a constant surface pressure of 22 mN/m to obtain a good signal.

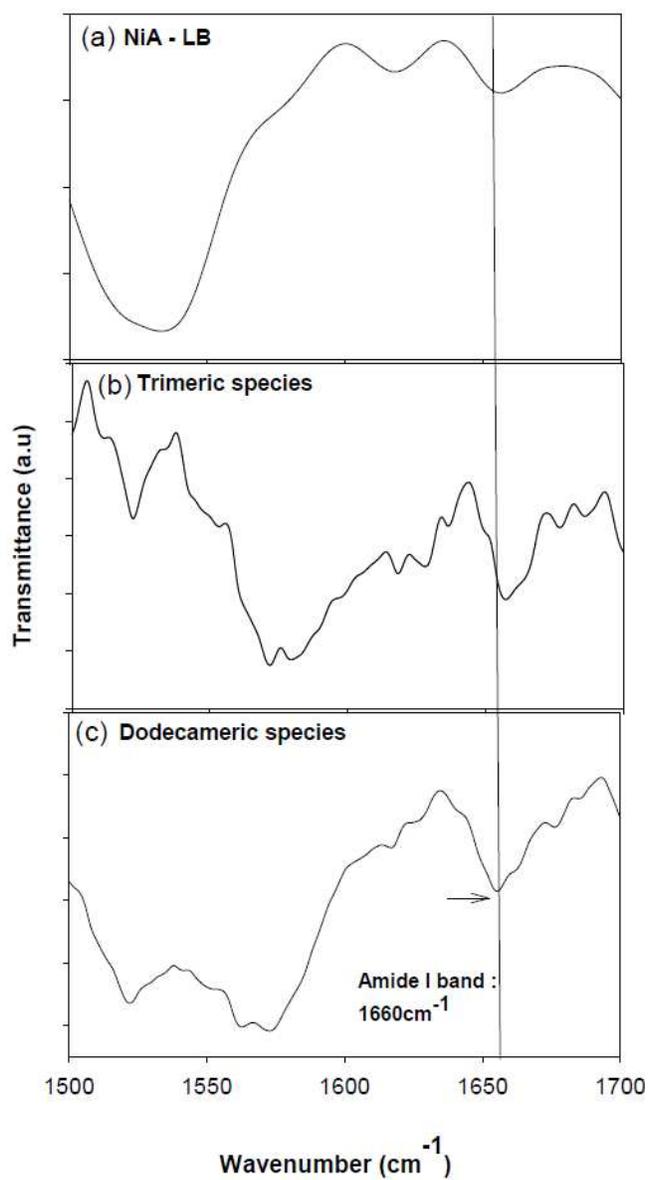
Supplementary Figures



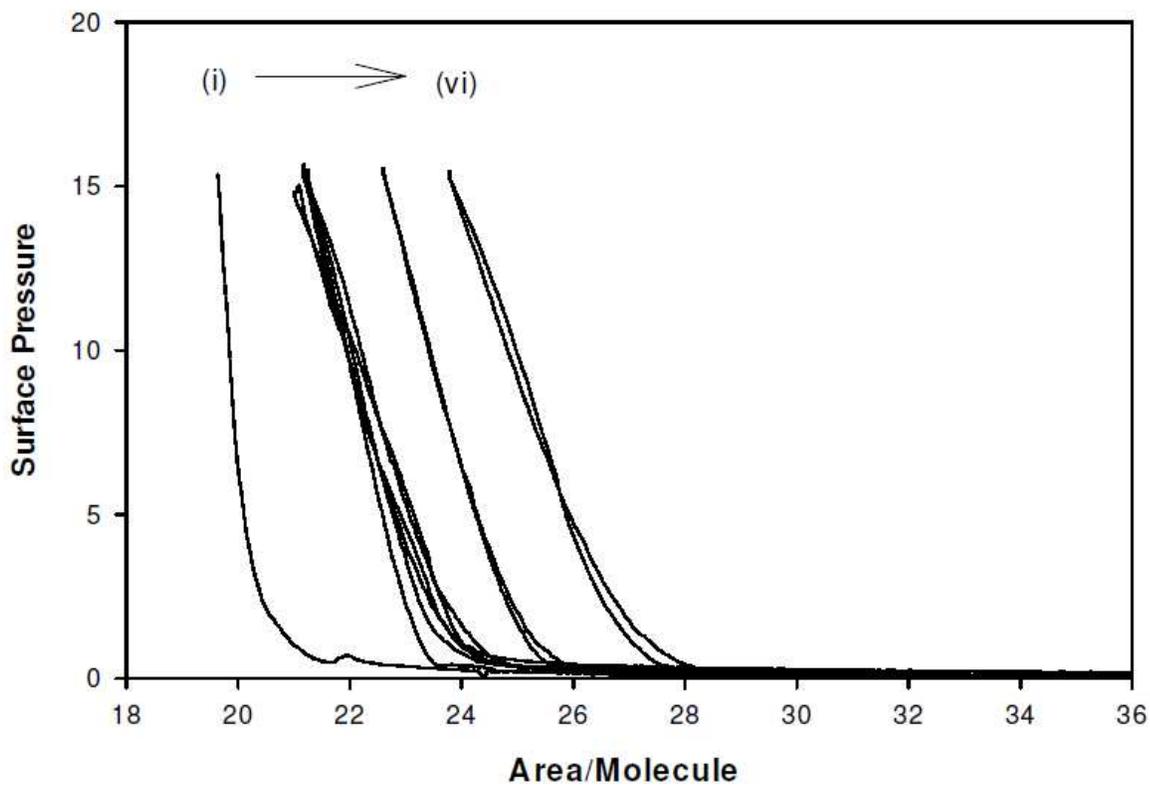
Supplementary Figure S1. Purification profile of His-MsDps1 protein on a 15% SDS-PAGE gel. Lanes FI and W denotes flow through and wash respectively. Lanes E1-E6 are the eluted protein fractions. The migration position of the proteins in the molecular weight marker is shown on the right.



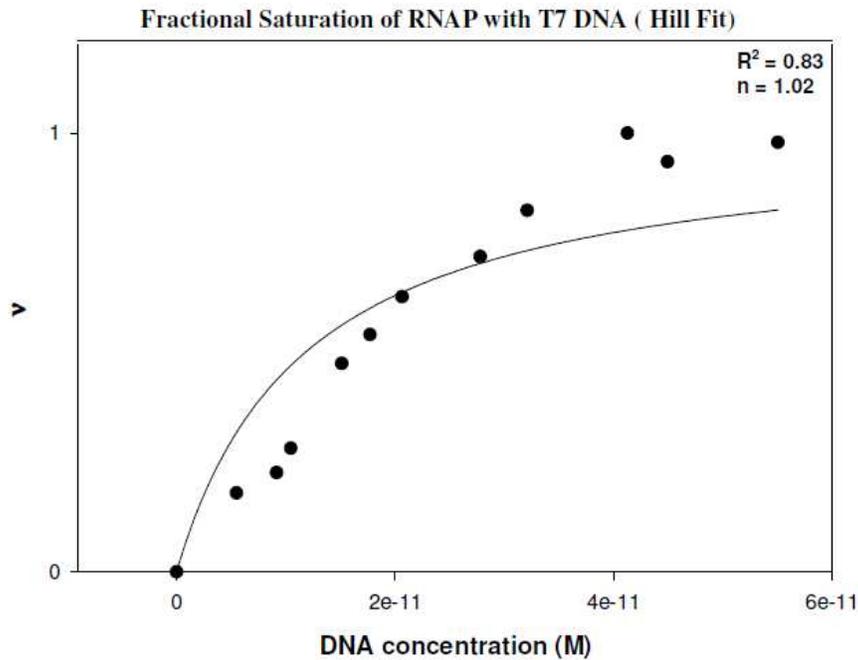
Supplementary Figure S2. A schematic representation of hydrophilic transfer of LB monolayer on a glass slide. The first layer is transferred on the upstroke and the orientation of each subsequent layer depends on the forces of adhesion involved. (adapted from NIMA-LB Manuals 4th edition)



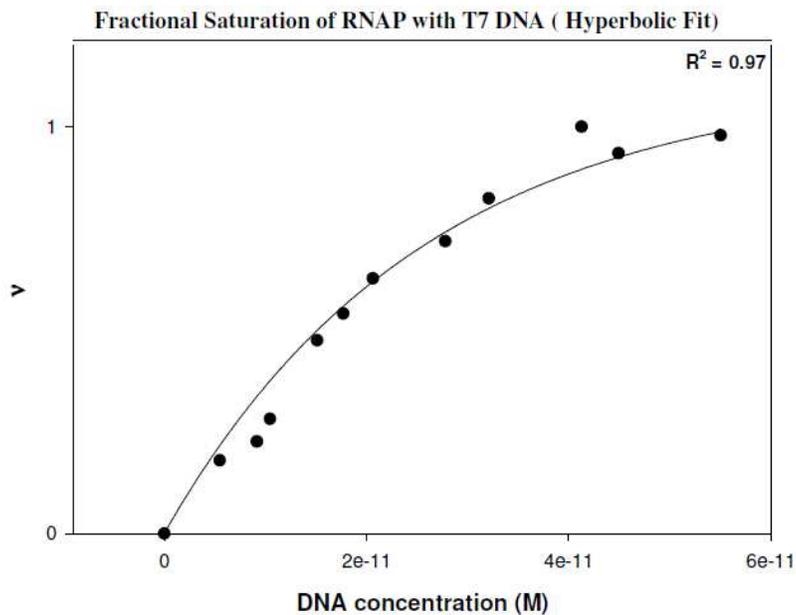
Supplementary Figure S3: FTIR of LB films of (a) NiA, (b) NiA-HisMsDps1 trimer and (c) NiA-HisMsDps1 Dodecamer. The Amide I carbonyl stretching frequency at 1650 cm^{-1} is a signature of the protein backbone. There is also a small peak at 1540 cm^{-1} which is due to the asymmetric COO^- stretching frequency.



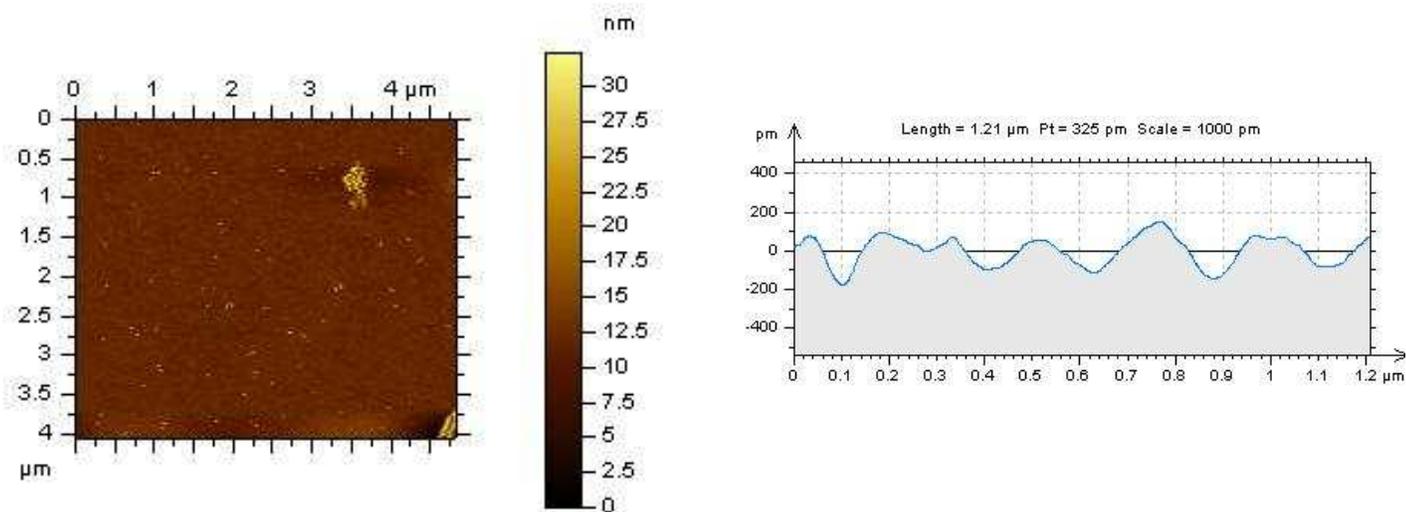
Supplementary Figure S4. From left to right P-A isotherms of (i) NiA and (ii) to (vi) shows the compression cycles of NiA with His-MsDps1 dodecamer (42 pmoles) at 25⁰ C and 20 mM NaCl over a period of 5 h. No hysteresis was observed.



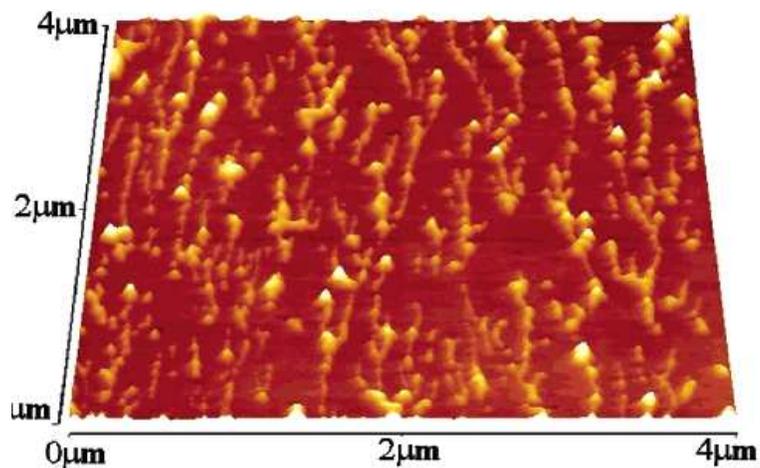
Supplementary Figure S5. A Hill Fit (3 parameter fit) for the fractional saturation of RNA polymerase with T7A1 promoter. The *r.m.s* value is only 0.83, and the Hill's coefficient is still $n = 1.02$



Supplementary Figure S6. A non-cooperative, hyperbolic fit (2 parameter fit) for the fractional saturation of RNA polymerase with T7A1 promoter. The *r.m.s* value is 0.97



Supplementary Figure S7. A 4 x 4 μm AFM image of NiA monolayer along with a height distribution profile of the image. The r.m.s roughness was 0.4-0.71 nm.



Supplementary Figure S8. A 4 x 4 μm HisRNAP-promoter DNA complex on 3 layers of NiA monolayer. (Brar, L. K.; Rajdev, P.; Raychaudhuri, A. K.; Chatterji, D. *Langmuir* **2005**, *21*, 10671-10675).