Data in brief 24 (2019) 103918

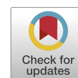
Contents lists available at ScienceDirect

Data in brief

journal homepage: www.elsevier.com/locate/dib

Data Article

Data on the target search by a single protein on DNA measured with ultrafast force-clamp spectroscopy



Carina Monico [a], Alessia Tempestini [a], Lucia Gardini [a, c], Francesco Saverio Pavone [a, b, c], Marco Capitanio [a, b, *]

[a] LENS – European Laboratory for Non-linear Spectroscopy, Via Nello Carrara 1, 50019, Sesto Fiorentino, Italy
[b] Department of Physics and Astronomy, University of Florence, Via Sansone 1, 50019, Sesto Fiorentino, Italy
[c] National Institute of Optics–National Research Council, Largo Fermi 6, 50125, Florence, Italy

A R T I C L E I N F O

Article history:
Received 21 March 2019
Received in revised form 5 April 2019
Accepted 8 April 2019
Available online 28 April 2019

Keywords:
Optical tweezers
Force-clamp spectroscopy
Lactose repressor
DNA
Single molecule biophysics

A B S T R A C T

The mechanism by which proteins are able to find small cognate sequences in the range from few to few tens of base pairs amongst the millions of non-specific chromosomal DNA has been puzzling researchers for decades. Single molecule techniques based on fluorescence have been successfully applied to investigate this process but are inherently limited in terms of spatial and temporal resolution. We previously showed that ultrafast force-clamp spectroscopy, a single molecule technique based on laser tweezers, can be applied to the study of protein-DNA interaction attaining sub-millisecond and few base-pair resolution. Here, we share experimental records of interactions between a single lactose repressor protein and DNA collected under different forces using our technique [1]. The data can be valuable for researchers interested in the study of protein-DNA interaction and the mechanism of DNA target search, both from an experimental and modeling point of view. The data is related to the research article "*Sliding of a single lac repressor protein along DNA is tuned by DNA sequence and molecular switching*" [2].

© 2019 The Authors. Published by Elsevier Inc. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

* Corresponding author. LENS – European Laboratory for Non-linear Spectroscopy, Via Nello Carrara 1, 50019, Sesto Fiorentino, Italy.

E-mail addresses: carina.monico@bioch.ox.ac.uk (C. Monico), tempestini@lens.unifi.it (A. Tempestini), gardini@lens.unifi.it (L. Gardini), francesco.pavone@unifi.it (F.S. Pavone), capitanio@lens.unifi.it (M. Capitanio).

https://doi.org/10.1016/j.dib.2019.103918
2352-3409/© 2019 The Authors. Published by Elsevier Inc. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

Specifications table

Subject area	*Biophysics*
More specific subject area	*Single molecule force spectroscopy*
Type of data	*Position records from an optical tweezers instrument*
How data was acquired	*Custom double optical tweezers instrument*
Data format	*Raw and graph*
Experimental factors	*DNA containing the operator O3 in between two copies of the operator O1 was labeled with biotins at both ends. Wild-type LacI was expressed In e.coli.* *No pretreatment of data*
Experimental features	*A single DNA molecule was stretched between streptavidin-coated beads and interactions with a single LacI molecule were recorded under forces in the range 3-10 pN*
Data source location	*Sesto Fiorentino, Italy, LENS – European Laboratory for Non-linear Spectroscopy*
Data accessibility	The raw data files are provided in the Data in Brief Dataverse, https://doi.org/10.7910/DVN/URGSKI [1]. *All other data is with this article*
Related research article	*Tempestini* et al. *"Sliding of a single lac repressor protein along DNA is tuned by DNA sequence and molecular switching", NAR 46, 5001–5011 (2018)*

Value of the data

- The data presented here is acquired using ultrafast force-clamp spectroscopy [3] and shows a much higher spatial and temporal resolution and much longer observation time compared to analogous single molecule studies using fluorescence microscopy [2,4–7]. Therefore, the data might contain valuable information on rapid (ms) conformational changes of the lactose repressor protein (LacI) during its interaction with cognate and non-cognate DNA sequences, as well as conformational changes occurring on a time scale of several minutes [8].
- Although the authors developed methods to analyze ultrafast force-clamp data on molecular motors [9,10], the analysis of the sliding motion of proteins along DNA during their target search would require advanced analysis tools to fully uncover sequence-dependent changes in sliding velocity. The shared data can be used to develop such tools.
- Models of DNA target search by transcription factors, restriction enzymes and other DNA-processing proteins rely to a great extent on single molecule data [11–13]. Single molecule high-resolution data obtain with ultrafast force-clamp spectroscopy can serve as a benchmark to define and test such models.

1. Data

The shared data are recordings from ultrafast force-clamp spectroscopy experiments in which a single lactose repressor protein (LacI) was interacting with a DNA molecule. The DNA molecule was a 10.2-kbp DNA construct containing the low-affinity operator O3 in between two copies of the high-affinity operator O1 [14]. The three operators were placed in the center of the DNA construct and the O3 operator was separated by 212 and 92 bp from the two O1 operators, respectively (relative distances between the operators are displayed in Fig. 1A). The DNA molecule was labeled with biotins at both ends, to allow the specific binding to streptavidin-coated microspheres. Experimental records,

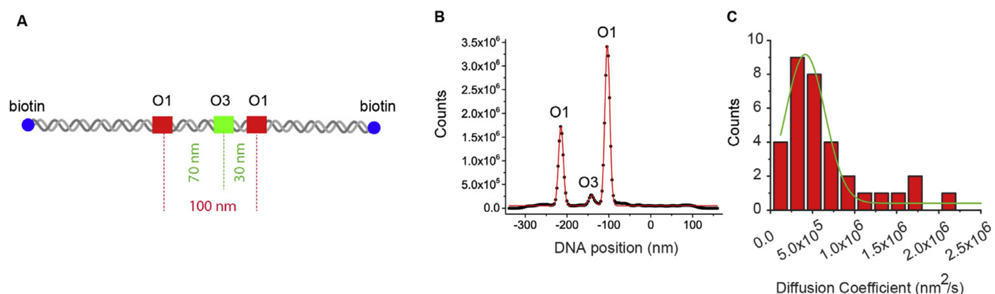


Fig. 1. **A)** Sketch of the DNA construct used in the data reported here. **B)** Distribution of the attachment point of LacI along DNA under negative force for the experimental record "Misclamp1" [1]. **C)** Distribution of the diffusion coefficient of LacI on DNA under ultrafast force-clamp spectroscopy. Measurements in (B) and (C) are under 3 pN force.

acquired as described in the next section, showed specific binding of LacI at specific locations on the DNA, corresponding to the distances between the operators O1 and O3 and with different affinity between O1 and O3, as expected [14] (Fig. 1B). Moreover, we observed sliding of LacI along the DNA construct with a broad distribution of sliding velocities. From those velocities, we calculated the LacI diffusion coefficient on DNA, as described in Ref. [2]. Fig. 1C shows the distribution of diffusion coefficients, which is well fitted by a Gaussian distribution centered around $D_{lac} = 4.1 \times 10^5$ nm^2s^{-1} and with a standard deviation $\sigma = 2.3 \times 10^5$ nm^2s^{-1}. Raw experimental records are composed by several minutes acquired at 200kHz and are therefore extremely large. We share few representative records under 3, 5, and 10 pN force [1], in which LacI shows binding to the three operators and sliding. Matlab scripts to read, plot and convert the raw data, as explained in the following section, are shared as supplemental files. About 156 traces obtained from scanning 22 DNA molecules and 54 LacI molecules with forces in the 3–10 pN range are available upon reasonable request to the authors.

2. Experimental design, materials, and methods

Data are acquired using ultrafast force-clamp spectroscopy, which is extensively described elsewhere [2,3]. Briefly, a single DNA molecule is trapped between two 1 μm diameter beads (forming a structure named herein "dumbbell"), pre-stretched at about 3 pN tension, and brought in close proximity to a third bead attached on the coverslip surface, where a LacI protein is present. Two voltage signals (V_1, V_2) proportional to the displacement of the two beads from the trap center along the DNA direction (x_{bead1}, x_{bead2}) are recorded from two quadrant photodiodes (QPD) (Fig. 2). These signals are also proportional to the applied forces, as $F_{1,2} = -k_{1,2}x_{bead1,2}$, where k is the trap stiffness and indices 1,2 refers to the two traps. The two traps can be moved independently along x by using two acousto-optic deflectors (AOD). The position of the two traps (x_{trap1}, x_{trap2}) is proportional to the AOD frequency (f_1, f_2). A double feedback loop is applied to the two AODs to maintain a constant net force on the beads by changing the AODs frequencies f_1, f_2 to maintain x_{bead1}, x_{bead2} constant. The total force F is alternated back and forth to maintain the dumbbell within a preset spatial interval as the dumbbell moves in a triangular wave.

When LacI binds DNA, interactions are detected from a change in velocity of the dumbbell. The dumbbell movement either slows down when LacI slides on DNA applying a viscous load, or stops when LacI binds strongly to DNA [2]. Differently from ultrafast force-clamp spectroscopy data obtained from the interaction between molecular motors and cytoskeletal filaments, in which interactions are detected from abrupt changes in the filament velocity [3,9,10], LacI-DNA interaction also shows variable changes in filament velocity corresponding to LacI sliding. Description of data analysis for strong interactions to DNA as well as analysis of sliding and how sliding velocity was converted to diffusion coefficients is extensively described in Tempestini et al. [2].

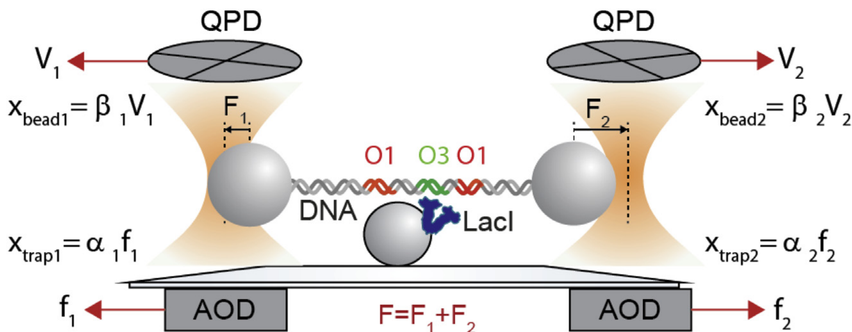


Fig. 2. Sketch of the ultrafast force-clamp spectroscopy configuration to measure interaction between LacI and DNA. The figure shows voltage signals from the QPD (V_1, V_2) and their relation to the bead displacements x_{bead1}, x_{bead2} through the detector calibration factors β_1,β_2. The figure also shows the frequencies f_1, f_2 of the acoustic waves generated inside the two AODs and their relation with the trap positions x_{trap1}, x_{trap2} through the calibration factors α_1,α_2.

The voltage signals V_1, V_2 from the QPDs are recorded together with the frequencies f_1, f_2 of the acoustic waves generated inside the AODs. Data is organized in a proprietary raw format (named "UFCS", from ultrafast force-clamp spectroscopy) starting with a header that contains all the experimental parameters used during data acquisition, followed by raw data formed by 4 channels (2 QPD voltages + 2 AOD frequencies). Calibration of the trap stiffness $k_{1,2}$ and the detector calibration factors β_1,β_2 are obtained before measurements using a power spectrum method [15] and recorded in the file header. We provide a Matlab script to read the header (readUFCSheader.m), a script to read the data (readUFCSdata.m), and a script that uses both readUFCSheader.m and readUFCSdata.m to load and

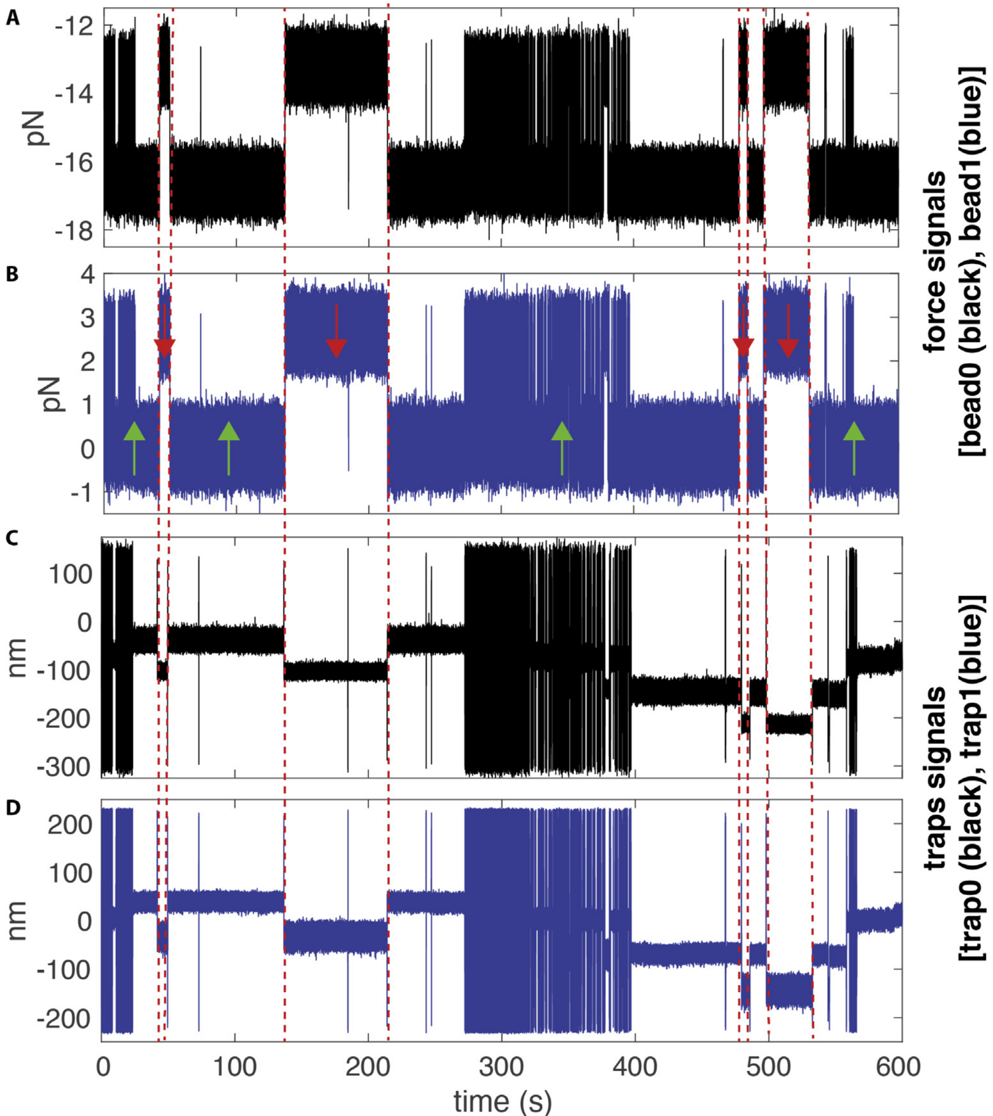


Fig. 3. Example of a representative 10 min record "Misclamp1", acquired under 3 pN force [1]. **A)** and **B)** are force signals obtained from the voltage signals V_1, V_2 as $F_{1,2}$ = -$k_{1,2}$ $xbead_{1,2}$ = - $k_{1,2}$ $\beta_{1,2}$ $V_{1,2}$. **C)** and **D)** are position signals obtained from the frequency signals f_1, f_2 as $x_{trap1,2}$ = $\alpha_{1,2}$ $f_{1,2}$. Vertical red dashed lines delimit regions in which the net force on the DNA was either positive (green arrows) or negative (red arrows).

visualize the data (ViewUFCSFiles.m). Data can be also saved as an ASCII table that can be imported in Excel, Origin, or similar analysis software. The file to be read must be in the current folder of Matlab and the filename written in a text file named UFCSfiles.txt. Table 1 in supplementary materials shows a description of the header. Fig. 3 shows an example of a 10 min record visualized through the View-UFCSFiles.m script. The time scale is large compared to the period of the triangular wave, which appears as vertical lines in the position records (Fig. 3C and D). Long interactions correspond to the binding of LacI to the three operators, as highlighted in Fig. 1B. Binding positions to the same operator under positive (green arrows) and negative (red arrows) force appear displaced by several tens of nanometers in the direction of the force because of the elasticity of the DNA-LacI complex.

Acknowledgments

This work was supported by the European Union's Horizon 2020 research and innovation program under grant agreement no. 654148 Laserlab-Europe and by Ente Cassa di Risparmio di Firenze.

Transparency document

Transparency data associated with this article can be found in the online version at https://doi.org/10.1016/j.dib.2019.103918.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.dib.2019.103918.

References

[1] M. Capitanio, C. Monico, A. Tempestini, L. Gardini, F.S. Pavone, LacI-DNA ultrafast force-clamp spectroscopy, Harvard Dataverse (2019), https://doi.org/10.7910/DVN/URGSKI.
[2] A. Tempestini, C. Monico, L. Gardini, F. Vanzi, F.S. Pavone, M. Capitanio, Sliding of a single lac repressor protein along DNA is tuned by DNA sequence and molecular switching, Nucleic Acids Res. 46 (2018) 5001–5011, https://doi.org/10.1093/nar/gky208.
[3] M. Capitanio, M. Canepari, M. Maffei, D. Beneventi, C. Monico, F. Vanzi, R. Bottinelli, F.S. Pavone, Ultrafast force-clamp spectroscopy of single molecules reveals load dependence of myosin working stroke, Nat. Methods 9 (2012) 1013–1019, https://doi.org/10.1038/nmeth.2152.
[4] Y.M. Wang, R.H. Austin, E.C. Cox, Single molecule measurements of repressor protein 1D diffusion on DNA, Phys. Rev. Lett. 97 (2006) 048302, https://doi.org/10.1103/PhysRevLett.97.048302.
[5] J. Elf, G.-W. Li, X.S. Xie, Probing transcription factor dynamics at the single-molecule level in a living cell, Science 316 (2007) 1191–1194, https://doi.org/10.1126/science.1141967.
[6] P. Hammar, P. Leroy, A. Mahmutovic, E.G. Marklund, O.G. Berg, J. Elf, The lac repressor displays facilitated diffusion in living cells, Science 336 (2012) 1595–1598, https://doi.org/10.1126/science.1221648.
[7] C. Monico, G. Belcastro, F. Vanzi, F.S. Pavone, M. Capitanio, Combining single-molecule manipulation and imaging for the study of protein-DNA interactions, JoVE 90 (2014) e51446, https://doi.org/10.3791/51446.
[8] M. Lewis, Allostery and the lac operon, J. Mol. Biol. 425 (2013) 2309–2316, https://doi.org/10.1016/j.jmb.2013.03.003.
[9] L. Gardini, A. Tempestini, F.S. Pavone, M. Capitanio, High-speed optical tweezers for the study of single molecular motors, in: C. Lavelle (Ed.), Mol. Mot., Methods in Molecular Biology, vol. 1805, Humana Press, New York, NY, 2018, pp. 151–184, https://doi.org/10.1007/978-1-4939-8556-2_9.
[10] L. Gardini, S.M. Heissler, C. Arbore, Y. Yang, J.R. Sellers, F.S. Pavone, M. Capitanio, Dissecting myosin-5B mechanosensitivity and calcium regulation at the single molecule level, Nat. Commun. 9 (2018) 2844, https://doi.org/10.1038/s41467-018-05251-z.
[11] C. Monico, M. Capitanio, G. Belcastro, F. Vanzi, F.S. Pavone, Optical methods to study protein-DNA interactions in vitro and in living cells at the single-molecule level, Int. J. Mol. Sci. 14 (2013) 3961–3992, https://doi.org/10.3390/ijms14023961.
[12] O.G. Berg, R.B. Winter, P.H. Von Hippel, Diffusion-driven mechanisms of protein translocation on nucleic acids. 1. Models and theory, Biochemistry 20 (1981) 6929–6948, https://doi.org/10.1021/bi00527a028.
[13] A. Tafvizi, L.A. Mirny, A.M. van Oijen, Dancing on DNA: kinetic aspects of search processes on DNA, ChemPhysChem 12 (2011) 1481–1489, https://doi.org/10.1002/cphc.201100112.
[14] K.A. Sharp, Allostery in the lac operon: population selection or induced dissociation? Biophys. Chem. 159 (2011) 66–72, https://doi.org/10.1016/j.bpc.2011.05.007.
[15] M. Capitanio, Optical tweezers, in: Y.L. Lyubchenko (Ed.), An Introd. To Single Mol. Biophys., CRC Press, Taylor & Francis group, 2017.