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Supporting Information

Alpha-Synuclein Modulates the Physical Properties of DNA

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k.j. Investigation: Equal; Writing – review & editing: Supporting
s.r. Investigation: Equal; Supervision: Equal; Writing – review & editing: Supporting
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f.w. Conceptualization: Equal; Funding acquisition: Equal; Methodology: Lead; Supervision: Equal; Writing – original draft: Equal; Writing – review & editing: Equal.

Supplementary Information

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Data analysis with Figures S2-S3 and references

Figure S4

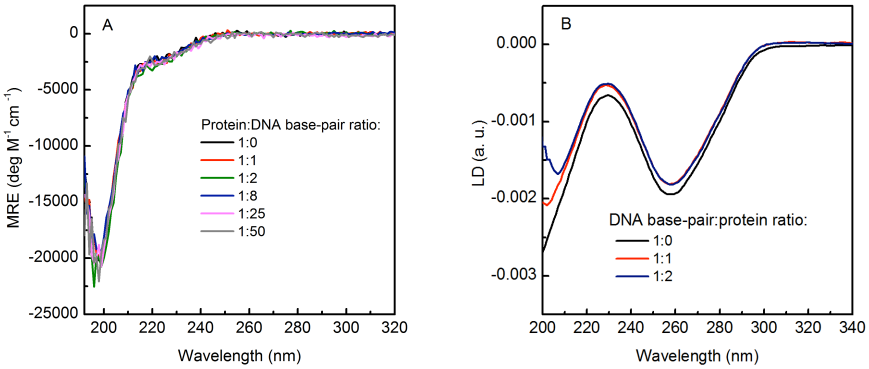

Figure S1. Structure characterization of aS and DNA (calf-thymus) by circular dichroism (CD) and linear dichroism (LD) spectroscopy. A) CD spectra of aS (5 μM) in the absence and presence of DNA. Each spectrum was subtracted from that of only DNA at the same concentration used in the samples with protein. B) LD spectra of DNA (10 μM base pairs) in the absence and presence of aS.

Computation of the persistence length from fractional extension data

When a semiflexible chain of length L, persistence length l_p, and effective width w, is confined to a channel of size D, excluded volume between the segments of the polymer cause it to extend a non-zero distance $R_{||}$ along the channel axis [1]. Werner *et al.* [2] proposed a correlated telegraph model to describe this stretching process. The model considers the confined polymer as a series of hairpin turns separated by a characteristic distance g, known as the global persistence length. The polymer contour between the hairpin turns is not perfectly aligned along the channel axis, due to thermal fluctuations, leading to an average alignment a. Finally, hairpin turns incur an excluded volume penalty ε when two strands of a hairpin attempt to occupy the same position along the channel axis.

The telegraph model predicts that the fractional extension of the confined chain is [2]

$$\frac{R_{||}}{L} = c_X a \alpha^{\frac{1}{3}} \qquad (1)$$

where $c_X = 1.1$ is a constant known to within rigorous bounds [3]. The scaling parameter α, arising from dimensional analysis of the model, quantifies the typical number of contacts between the two strands of a hairpin of length g. In addition to developing this theory, Werner *et al.* [2] provided data for the parameters appearing in Eq. 1 for square channels in the form

$$a^2 = f_1\left(\frac{D}{l_p}\right) \qquad (2)$$

and

$$\frac{\alpha D^2}{w l_p} = f_2\left(\frac{D}{l_p}\right) \qquad (3)$$

where the function f_1 appears in Fig. 3a and the function f_2 appears in Fig. 3d of Ref. [2]. Given the physical properties of the polymer (L, l_p, w) and the channel size (D), Eqs. 1-3 permit a calculation of the stretching $R_{||}$ of the polymer.

The experimental data consist of measurements of DNA extension at different concentrations of aS in two different channel sizes, which we denote as D_1 and D_2, with corresponding extension $R_{||,1}$ and $R_{||,2}$. From Eq. 1, the model predicts that the ratio of the fractional extensions is

$$\frac{R_{||,1}}{R_{||,2}} = \frac{a_1}{a_2}\left(\frac{\alpha_1}{\alpha_2}\right)^{1/3} \qquad (4)$$

Using Eq. 2 and Eq. 3 in the latter, we can write this ratio as

$$\frac{R_{||,1}}{R_{||,2}} = \left(\frac{D_2}{D_1}\right)^{2/3} \frac{f(D_1/l_p)}{f(D_2/l_p)} \qquad (5)$$

where

$$f = f_1^{1/2} f_2^{1/3} \quad (6)$$

Figure S2 provides a plot of Eq. 6 based on the data from Werner *et al.* [2], as well as a spline interpolation used for the subsequent analysis.

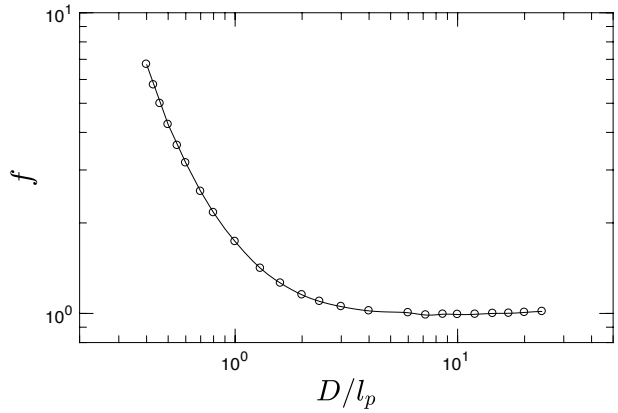

Figure S2. *Plot of the function f(D/l_p). Circles correspond to the data from Ref. [2] using Eq. 6. The solid line is a spline interpolant used to compute the fractional extension by Eq. 5.*

Equation 5 provides an approach to compute the persistence length l_p from a pair of measurements of the fractional extension at different channel sizes. This calculation must be done numerically since there is no simple, invertible form of *f*. **Figure S3** plots Eq. 5 for the channel sizes used in this experiment. While it is possible to read off the values of the persistence length directly from this figure, we found it more robust to first form a residual between Eq. 5 and the experimental measurement, fit that residual with a spline, and then numerically compute the zero of the spline.

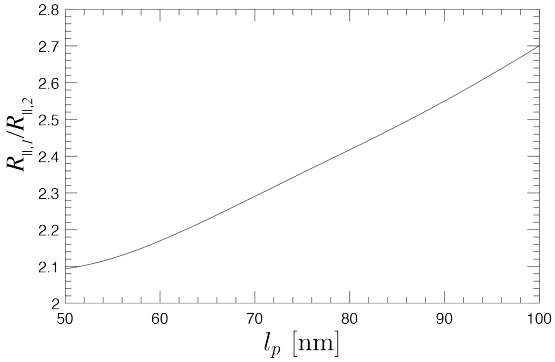

Figure S3. *Plot of the predicted ratio in fractional extension, $R_{||,1}/R_{||,2}$, as a function of the persistence length l_p for the channel sizes used in this experiment, D_1 = 122 nm and D_2 = 324 nm. The channel sizes correspond to the geometric mean of the width and height of the rectangular channels.*

There are several approximations in this analysis that are worth noting. Most importantly, the data used to compute *f* in Ref. [2] were obtained from simulations of ideal semiflexible chains confined in square channels. To approximate the behavior in rectangular channels, we use the geometric mean of the channel size for *D* [4]. For small aspect ratios, this is an excellent approximation.

However, for very large aspect ratios, this approximation can lead to systematic errors since the chain statistics are sensitive to the ratio of the channel size to the persistence length [5]. For the particular channel sizes used here, previous work suggests that the geometric mean provides a reasonable estimate for the fractional extension but would lead to systematic errors when computing the variance about that extension [6]. We have also made two less severe assumptions in the analysis, neglecting the depletion length with the channel wall [4] and any changes in the effective width due to binding of aS to the DNA. These assumptions are expected to have a much smaller impact on the measurement of the persistence length than approximating the rectangular geometry by its geometric mean. Moreover, there is considerably uncertainty in the size of the wall depletion length [7] and the change in effective width due to binding of aS (since the effective width is of electrostatic origin [8]). As a result, attempts to correct for the latter two assumptions may introduce further systematic errors.

References

1. M. Daoud and P.G. de Gennes, "Statistics of macromolecular solutions trapped in small pores." J. Phys. Paris 38, 85-93 (1977).
2. E. Werner, G.K. Cheong, D. Gupta, K.D. Dorfman and B. Mehlig, "One parameter scaling theory for DNA extension in a nanochannel." Phys. Rev. Lett. 119, 268102 (2017).
3. R. van der Hofstad, "The constants in the central limit theorem for the one dimensional Edwards model." J. Stat. Phys. 90, 1295-1310 (1998).
4. Y. Wang, D.R. Tree and K.D. Dorfman, "Simulation of DNA extension in nanochannels." Macromolecules 44, 5694-6604 (2011).
5. E. Werner and B. Mehlig, "Scaling regimes of a semiflexible polymer in a rectangular channel." Phys. Rev. E 91, 050601(R) (2015).
6. D. Gupta, J. Sheats, A. Muralidhar, J.J. Miller, D.E. Huang, S. Mahshid, K.D. Dorfman and W. Reisner, "Mixed confinement regimes during equilibrium confinement spectroscopy of DNA." J. Chem. Phys. 140, 214901 (2014).
7. W. Reisner, J. Pedersen and R.H. Austin, "DNA confinement in nanochannels: physics and biological applications." Rep. Prog. Phys. 75, 106601 (2012).
8. D. Stigter, "Interactions with highly charged colloidal cylinders with applications to double-stranded DNA." Biopolymers 16, 1435-1448 (1977).

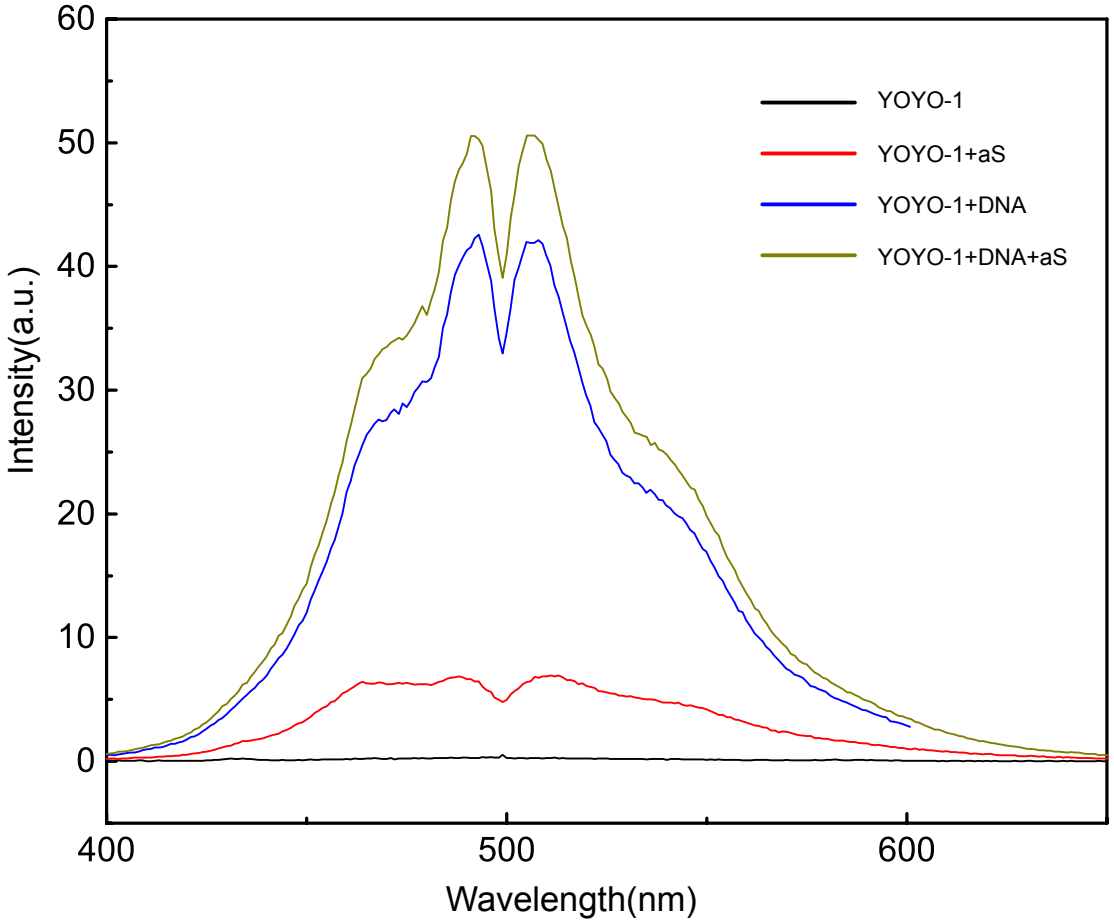

Figure S4. Fluorescence excitation and emission spectra of YOYO-1 (0.2 μM) in the presence of aS (red) or λ-DNA (blue), and aS plus λ-DNA (green). Excitation was at 491 nm (for emissions spectra) and emission at 509 nm (for excitation spectra). The protein concentration was 40 μM and λ-DNA was added to give a ratio of aS-to-base pair of 8:1. The spectrum of YOYO-1 in buffer is also shown for reference (black).