Electronic Supplementary Material (ESI) for Soft Matter.

SUPPLEMENTARY INFORMATION

The Effect of Linker DNA on the Structure and Interaction of Nucleosome Core Particles

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DNA sequences used in nucleosome reconstitutions.

145 bp 601, (net charge –288e):

TGGAGAATCC CGGTGCCGAG GCCGCTCAAT TGGTCGTAGA CAGCTCTAGC ACCGCTTAAA CGCACGTACG CGCTGTCCCC CGCGTTTTAA CCGCCAAGGG GATTACTCCC TAGTCTCCAG GCACGTGTCA GATATATACA TCCTG

157 bp, (net charge –312e):

ACTCCC **--- 145bp 601 ---** TGCAGT

177 bp, (net charge –352e):

ACTTACGCGG CCGCCC **--- 145bp 601 ---** TGCATGTATT GAAAGT

601-207; net charge -412e

ATCTCGGGGA TGGACCCTAT ACGCGGCCGC CC **--- 145bp 601 ---** TGCATGTATT GAACAGCGAC CTCTCGGGAT

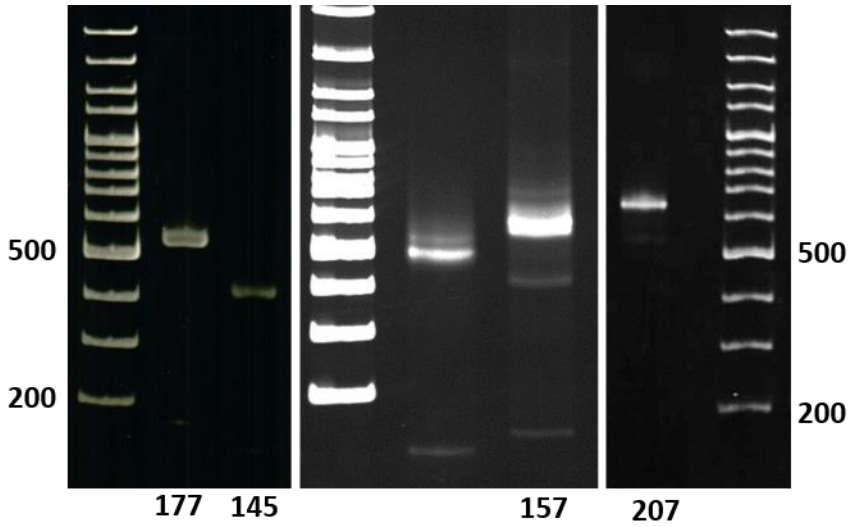

Fig. S1. Polyacrylamide gel electrophoretograms of nucleosomes with 145, 157, 177 and 207 bp DNA. The 100 bp DNA marker ladder with 500 bp and 200 bp DNA fragments are indicated for reference. The unlabeled middle lane in the middle panel is NCP-145 with a different DNA sequence not studied in this work. The figure was assembled from three gels that were run separately, imaged and cropped to show the areas of interest. The original gel images are available upon request.

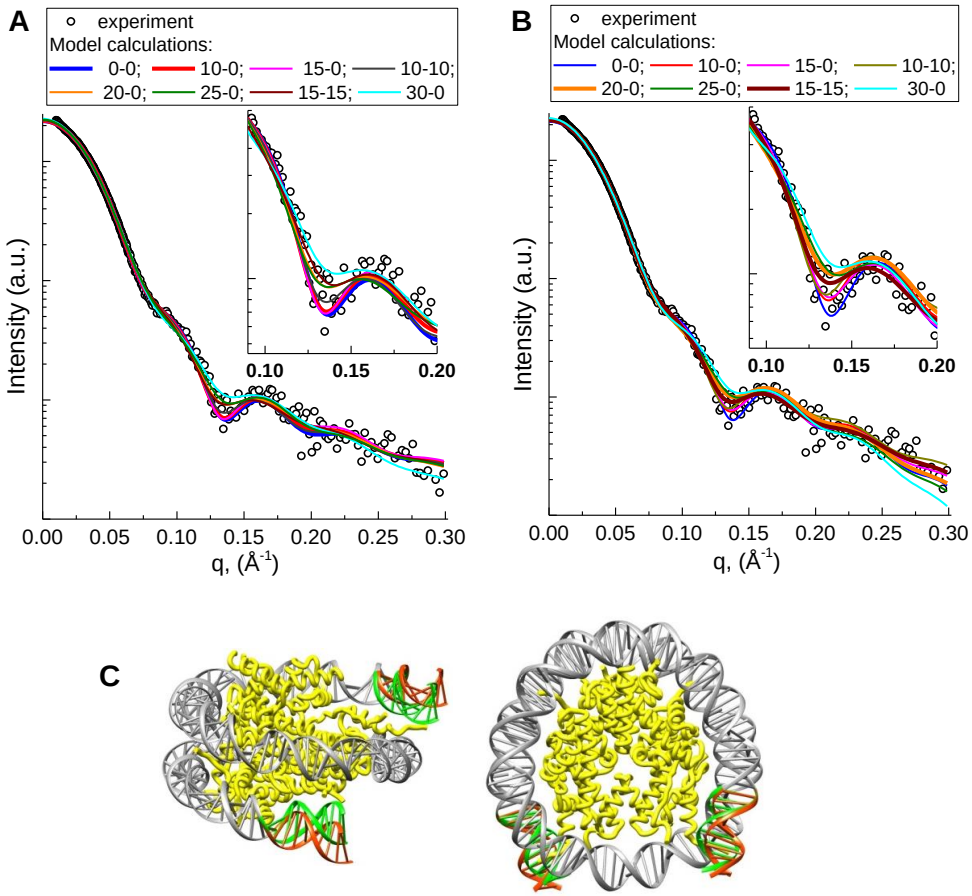

Figure S2. Comparison of SAXS spectra measured in experiment (points) and simulated from molecular structures (curves). Experimental data are for the NCP-145 (1.25 mg/mL). Curves show the SAXS profiles calculated from the molecular structures modelling different degrees of unwinding from one or two ends of the NCPs as indicated in the legend above the graphs. Calculated SAXS profiles were determined using two different NCP structures: (**A**) A snapshot from the MD simulations with histone tails collapsed on the DNA was used as initial structure to build models with DNA unwrapping. **(B).** 145 bp with '601' DNA sequence (3LZ0.pdb (1); histone source and DNA sequence are similar to the ones used in experiment but with significant number of the amino acids from the histone tails missing from the structure). Inserts in (A) and (B) show superimposed experimental SAXS spectra to highlight the local minimum at q = 0.14 $Å^{-1}$. **(C).** An illustration that unwrapping of 10 bp DNA from each end of the NCP-145 results in relatively small alteration of the nucleosome structure.

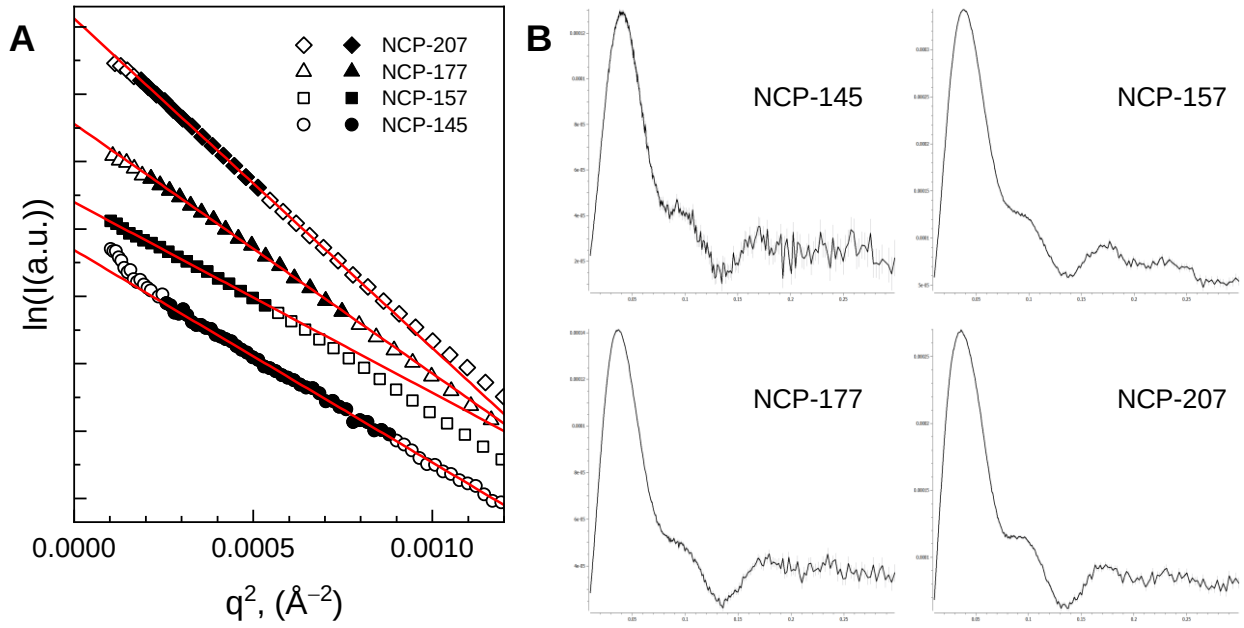

Figure S3. Examples of Guinier (**A**) and Kratky (**B**) plots calculated from the SAXS spectra of the nucleosome solutions shown in Fig. 1 of the main text (NCP-145, 1.25 mg/mL; NCP-157, 3 mg/mL; NCP-177, 1.5 mg/mL; and NCP-207, 3 mg/mL). In (**A**) solid points used for linear fitting shown by red lines; Rg values calculated from the Guinier plots are given in Tables S1-S4. In (**B**) four panels are output from the ATSAS software with identity of the NCP sample indicated in the panels. Bell-shaped form of all curves indicates that all samples of the NCP are folded and globular.

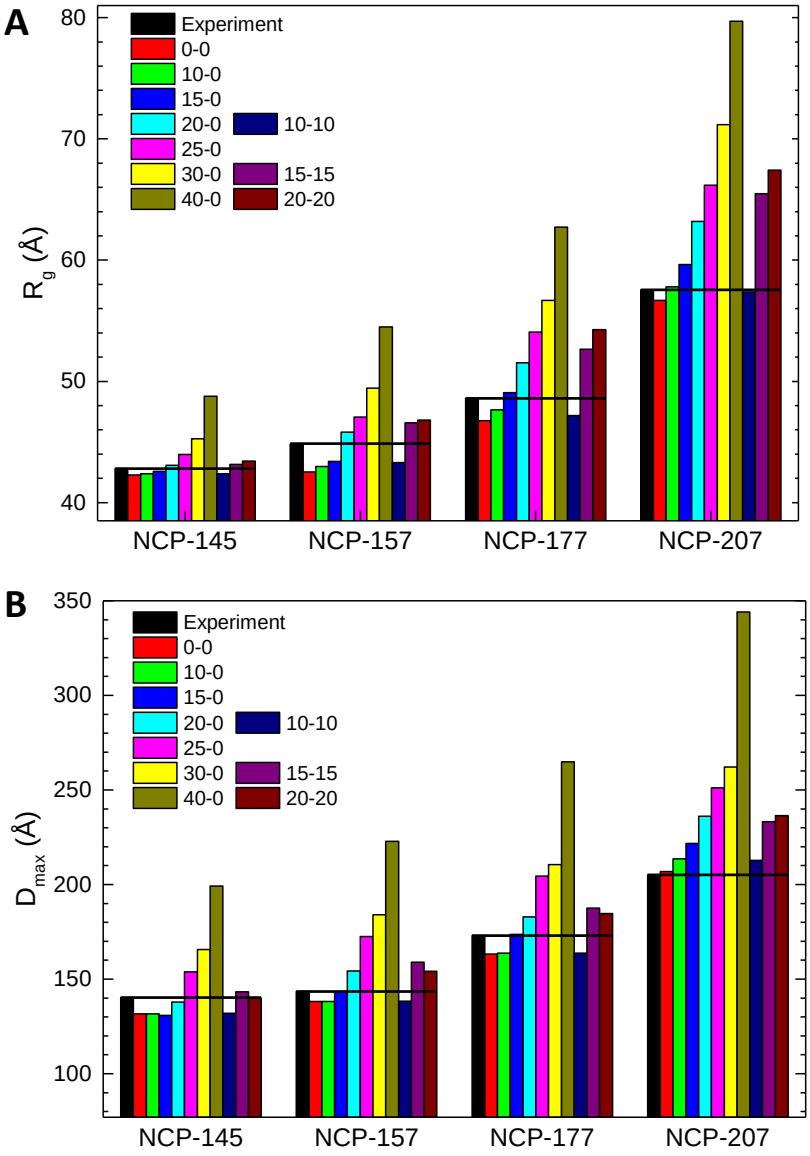

Figure S4. Comparison of the R_g (**A**) and D_{max} (**B**) values calculated from the SAXS solution spectra (in black) with the corresponding values from the form factor profiles (in colours) obtained for the atomic structures modelling DNA unwrapping from the histone octamer. The simulated data is grouped according to the number of DNA base pairs unwrapped in asymmetric (from one end) or symmetric fashion (from two ends). To guide an eye, horizontal lines are drawn on the levels of the experimental R_g and D_{max} values. In (**A**) and (**B**) origins of the y-axis are placed at position of the respective radius and diameter of the sphere with volume equal to the all-atom volume of the dry NCP-145 that corresponds to the minimal possible R_g (38.5 Å) and D_{max} (77 Å) values for all nucleosome structures.

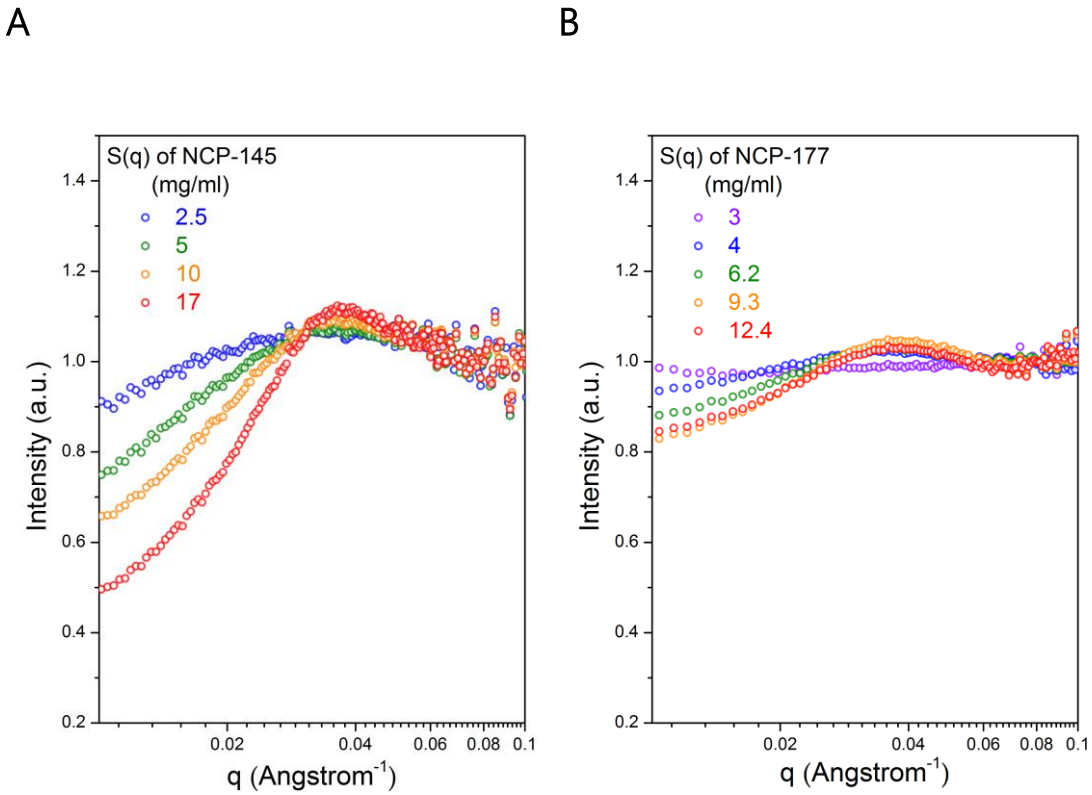

Figure S5. The structure factors, *S(q)*, of NCP-145(A) and NCP-177(B) at various concentrations obtained by dividing the intensity profiles by their respective form factors. Over the concentration range studied, the structure factor of NCP-145 displayed a much more pronounced concentration dependence and deviation from the value of 1.0 in the low-q region than that of NCP-177, conforming that NCP-145 exhibited a stronger interparticle interaction than NCP-177 under a given concentration.

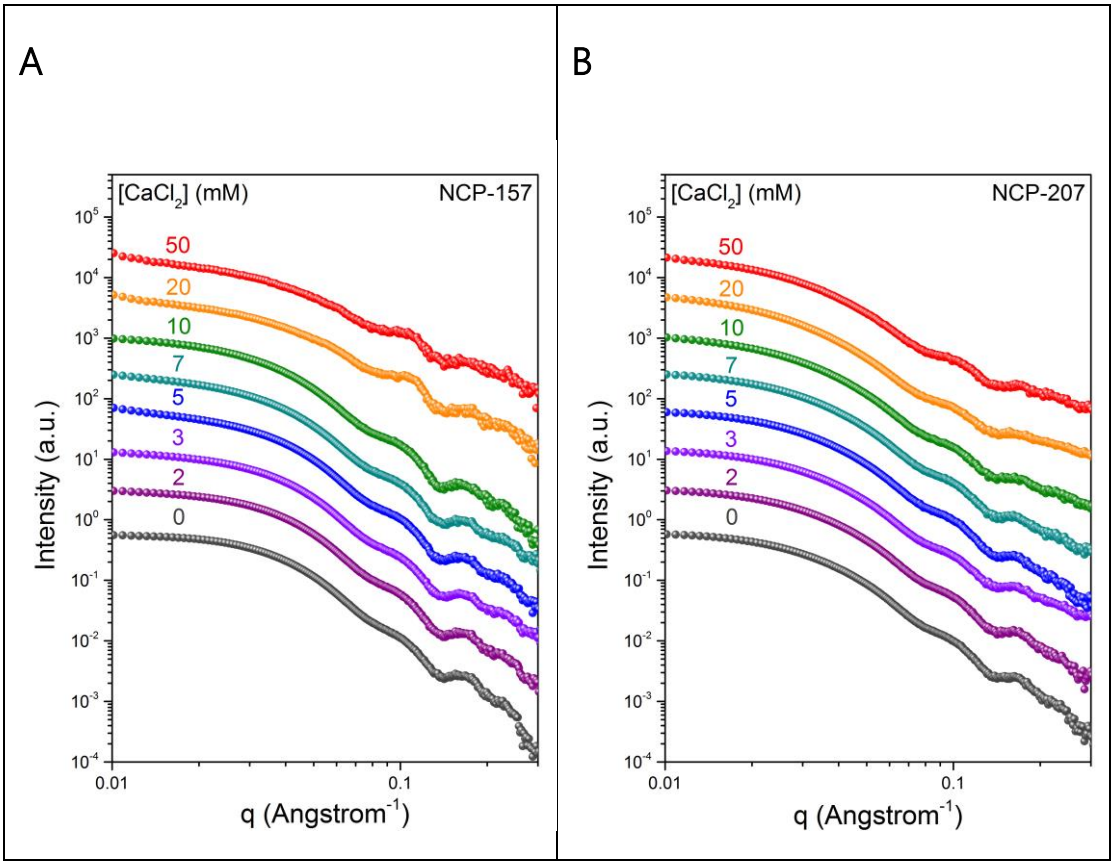

Figure S6. Influence of $CaCl_2$ on the structures and interparticle interactions of the NCP-157(A) and NCP-207 (B). Concentrations of added $CaCl_2$ (in mM) are indicated in the graphs. The initial NCP concentrations are 2.5 and 3 mg/ml for NCP-157 and NCP-207, respectively.

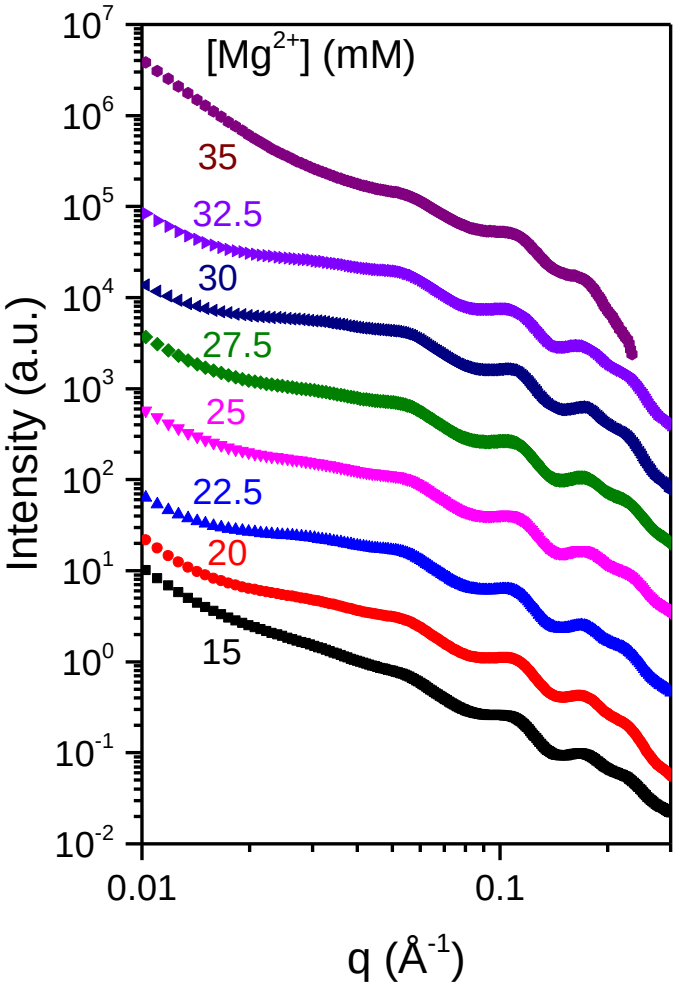

Figure S7. SAXS spectra of NCP-177 at 8 mg/mL precipitated by Mg^{2+}. Concentrations of Mg^{2+} are indicated next to the corresponding spectrum of the same color.

Table S1. Comparison of the outputs from SAXS profiles obtained the NCP-145 in experiment and simulated from modelled molecular structures, including the radius of gyration *(R_g)* and the maximal intra-atom distances within a particle (D_{max}) determined from the distance distribution function *P(r)*. Top row of numbers shows parameters obtained from the experiment; two sections below present output of the analysis of molecular structures constructed from stretches of straight DNA and crystal structure 1KX5.

Number of unwrapped bp	R_g from Guinier plot, Å	R_g from P(r), Å	D_{max}, Å	Fit quality*, χ^2
Data extracted from SAXS measurement NCP-145, 1.25 mg/mL				
	42.85 +/– 0.22	**42.80** +/– 0.15	**140.2**	0.952 excellent
Structure with collapsed tails generated MD simulations using 1KX5 coordinates				
0-0	42.49	42.29	131.6	**2.12**
10-0	42.59	42.38	131.7	**2.10**
15-0	42.76	42.58	130.8	**2.16**
10-10	42.56	42.37	131.9	**2.14**
20-0	43.26	43.08	137.8	2.33
25-0	44.10	43.96	153.8	2.60
15-15	43.31	43.14	143.3	2.60
30-0	45.33	45.26	165.6	3.16
20-20	43.54	43.43	140.0	2.79
40-0	48.76	48.77	199.2	6.62
Crystal structure 3LZ0 with '601' DNA sequence				
0-0	41.42	41.43	117.0	4.68
10-0	41.91	41.92	121.5	2.88
15-0	42.08	42.09	125.5	2.63
10-10	42.14	42.18	122.4	2.84
20-0	43.08	43.11	139.9	**2.10**
25-0	43.57	43.62	145.8	2.42
15-15	42.58	42.61	127.5	**2.10**
30-0	45.06	45.16	159.9	3.00
20-20	44.14	44.28	153.2	3.24
40-0	50.09	50.40	199.9	9.51

*For the experimental data, fit quality value represents the pair distance distribution fit from the GNOM module of the ATSAS package. For the NCP models, the fit quality is defined by χ^2 value calculated for the difference between experimental and simulated spectra.

Table S2. Comparison of output from SAXS profiles obtained the NCP-157 in experiment and simulated from modelled molecular structures, including the radius of gyration *(R_g)* and the maximal intra-atom distances within a particle (D_{max}) determined from the distance distribution function *P(r)*.. Top row of numbers shows parameters obtained from the experiment; a section below presents output of the analysis of molecular structures constructed from stretches of straight DNA and MD-generated NCP-145 structure.

Number of unwrapped bp	R_g from Guinier plot, Å	R_g from P(r), Å	D_{max}, Å	Fit quality*, χ^2
NCP-157; 3.0 mg/mL; data extracted from SAXS profile				
	44.86 +/– 0.63	**44.85** +/– 0.63	**143.5**	0.669 reasonable
Structures generated by combining MD-generated NCP and straight B-DNA fragments				
0-0	42.59	42.52	138.2	32.09
10-0	43.01	42.97	138.2	33.17
15-0	43.46	43.41	143.4	35.99
10-10	43.35	43.29	138.4	44.81
20-0	45.88	45.80	154.3	32.41
25-0	47.06	47.05	172.6	9.87
15-15	46.69	46.57	158.9	38.27
30-0	49.35	49.43	184.1	**4.61**
20-20	46.80	46.80	154.1	37.89
40-0	54.21	54.48	222.9	8.81

*For the experimental data, fit quality value represents the pair distance distribution fit from the GNOM module of the ATSAS package. For the NCP models, the fit quality is defined by χ^2 value calculated for the difference between experimental and simulated spectra

Table S3. Comparison of output from SAXS profiles obtained the NCP-177 in experiment and simulated from modelled molecular structures, including the radius of gyration *(R_g)* and the maximal intra-atom distances within a particle (D_{max}) determined from the distance distribution function *P(r)*. Top row of numbers shows parameters obtained from the experiment; a section below presents output of the analysis of molecular structures constructed from stretches of straight DNA and MD-generated NCP-145 structure.

Number of unwrapped bp	R_g from Guinier plot, Å	R_g from P(r), Å	D_{max}, Å	Fitting quality*, χ^2
NCP-177; 1.5 mg/mL; data extracted from SAXS profile				
	48.57 +/– 0.30	**48.60** +/– 0.30	**173.0**	0.783 good
Structures generated by combining MD-generated NCP and straight B-DNA fragments				
0-0	46.69	46.75	163.2	**2.20**
10-0	47.61	47.65	163.7	**3.50**
15-0	49.04	49.06	173.7	5.79
10-10	47.08	47.17	163.7	5.78
20-0	51.42	51.52	182.9	15.19
25-0	53.86	54.07	204.4	7.16
15-15	52.52	52.66	187.5	14.57
30-0	56.36	56.69	210.6	9.47
20-20	54.05	54.26	184.6	12.24
40-0	62.31	62.72	264.8	11.05

*For the experimental data, fit quality value represents the pair distance distribution fit from the GNOM module of the ATSAS package. For the NCP models, the fit quality is defined by χ^2 value calculated for the difference between experimental and simulated spectra

Table S4. Comparison of output from SAXS profiles obtained the NCP-207 in experiment and simulated from modelled molecular structures, including the radius of gyration *(R_g)* and the maximal intra-atom distances within a particle (D_{max}) determined from the distance distribution function *P(r)*.. Top row of numbers shows parameters obtained from the experiment; a section below presents output of the analysis of molecular structures constructed from stretches of straight DNA and MD-generated NCP-145 structure.

Number of unwrapped bp	R_g from Guinier plot, Å	R_g from P(r), Å	D_{max}, Å	Fit quality*, χ^2
NCP-207; 3.0 mg/mL; data extracted from SAXS profile				
	54.18 +/– 0.59	**57.54** +/– 0.08	**205.1**	0.695 reasonable
Structures generated by combining MD-generated NCP and straight B-DNA fragments				
0-0	56.25	56.69	206.8	**5.04**
10-0	57.42	57.80	213.6	6.91
15-0	59.14	59.63	221.8	9.61
10-10	57.00	57.38	212.8	51.67
20-0	62.63	63.20	236.1	78.44
25-0	65.48	66.17	251.1	23.07
15-15	64.79	65.47	233.2	25.80
30-0	70.46	71.18	262.2	13.93
20-20	66.60	67.42	236.5	14.80
40-0	78.86	79.71	344.1	24.12

*For the experimental data, fit quality value represents the pair distance distribution fit from the GNOM module of the ATSAS package. For the NCP models, the fit quality is defined by χ^2 value calculated for the difference between experimental and simulated spectra.

Table S5. Analysis of SAXS solution spectra obtained for NCP-145 and NCP-177 at different NCP concentrations

Concentration, mg/mL:	R_g from Guinier plot, Å	R_g from P(r), Å	D_{max}, Å	Fit quality*
NCP-145				
1.25	42.85 +/– 0.22	42.80 +/– 0.22	140.2	0.952 excellent
2.5	41.84 +/– 0.48	41.71+/–0.48	119.1	0.878, good
5.0	41.83 +/– 1.08	41.71+/–1.08	121.3	0.684, reasonable
10.0	41.65+/– 2.19	41.53+/– 2.19	117.8	0.679, reasonable
17.0	--	--	--	No solution
NCP-177				
1.5	48.57 +/– 0.30	48.60 +/– 0.30	173.0	0.783 good
3.0	47.79 +/– 0.61	47.80+/– 0.61	156.6	0.655, reasonable
4.0	47.60+/– 0.88	47.61+/– 0.88	156.1	0.657, reasonable
6.2	46.85+/–1.41	46.83+/– 1.41	151.6	0.660, reasonable
9.3	46.00+/–2.28	45.96+/– 2.28	146.0	0.663, reasonable
12.4	46.13+/– 2.81	46.09+/– 2.81	146.6	0.664, reasonable

*For the experimental data, fit quality value represents the pair distance distribution fit from the GNOM module of the ATSAS package.

Supporting References

1. Vasudevan, D., Chua, E.Y. and Davey, C.A. (2010) Crystal structures of nucleosome core particles containing the '601' strong positioning sequence. *J.Mol.Biol.*, **403**, 1-10.