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DNA Deformability at the Base Pair Level

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It is now well established that DNA−protein interactions are governed not only by specific chemical contacts but also by the structure of the particular DNA sequence and/or its propensity to adopt a suitable conformation. To understand these phenomena, the energetic cost associated with DNA mechanical deformation has to be known. A range of experimental and theoretical approaches has been used to study DNA deformability at the flexible-rod or dinucleotide level, but as far as the base pair level is concerned, only base pair opening has recently been investigated in more detail.[1−7] Very little is known about the deformation energetics of other base-pair degrees of freedom.

To shed more light onto the problem, we performed unrestrained molecular dynamics simulations of selected DNA duplex oligomers and analyzed the structural fluctuations to obtain harmonic elastic potentials for the six base pair conformational parameters. These describe the mutual position and orientation of one base in the pair with respect to the other and include the displacement of the bases along the pair's long axis (stretch), short axis (shear), and DNA helical axis (stagger), as well as the base−base opening angle toward the grooves (opening), around the pair's short axis (buckle), and around the long axis (propeller twist).[8,9] We build on our experience by applying the method to study global elastic properties of DNA oligomers,[10] the effect of nucleotide exocyclic groups,[11] and sequence-dependent base-pair step deformability.[12]

Two 18-bp oligomers have been simulated. The first one, d(GCCTATAAACGCCTATAA), contains a strong nucleosome positioning motif[13] and was found to have exceptional global elastic properties, while the other simulated oligonucleotide, d(CTAGGTGGATGACTCATT), exhibits properties close to those of the generic B-DNA.[14] Both sequences include multiple AT and GC pairs in different sequence contexts. We have chosen the two oligomers because the substantial differences in their global deformability may be reflected at the local level, which would enable us to cover a wide range of possible local elastic behavior.

The simulation protocol[12] is described in the Supporting Information. Briefly, each oligomer was simulated (using the AMBER6 package) in NPT ensemble in the presence of water and Na^+ counterions plus 100 mM NaCl added salt (total 40 000 atoms). The Cornell et al.[15] force field, truncated octahedron box, and random ion placement were used, and the simulations were extended to 20 ns each. The snapshots were recorded every ps, and the 3DNA algorithm[16] was used to obtain the time courses of the conformational parameters. The method of fluctuations[17] was then applied: assuming that the parameters have a multidimensional Gaussian distribution, their correlation matrix is related to the inverse of the stiffness matrix by a simple equation, $\langle x_i x_j \rangle = kT(F^{-1})_{ij}$, where x_i is the value of the ith parameter (with the average value subtracted) and F is the stiffness matrix. The effect of the Jacobian arising from the transformation of variables to noncanonical (angular) ones[18] was neglected due to small angular fluctuations. The first nanosecond of the simulations and three base pairs at each end were excluded from the analysis. Thus, 13 AT pairs and 11 GC pairs were considered.

The "diagonal" force constants (averages and ranges of observed values) are summarized in Table 1. They describe the energy cost associated with the change of only one parameter, while the others retain their equilibrium values. The full list of force constants including coupling terms and energy errors is available as Supporting Information.

As can be seen from Table 1, the GC base pair is, on average, stiffer than AT in all parameters except propeller twist and shear for which the values are comparable. The ratios of average force constants for GC and AT pairs with respect to buckle, stretch, and stagger span the range of 1.4−1.7 while the ratio for opening is 3.9. However, the range of observed values (about 10% of the average) blurs this difference, and only stretch and opening remain distinctly stiffer for GC than for AT in all instances. Note that the ratio of the stretch stiffness of the GC and AT base pairs (1.7) is closer to the ratio of the number of their H-bonds (1.5) than to the ratio of their intrinsic (gas phase) interaction energies (2.15 with the Cornell et al. force field).[19−21]

An important insight into base pair interactions in B-DNA emerged from studies of base pair opening. Using atomic-resolution molecular dynamics with umbrella sampling, Lavery and co-workers[3,6,7] investigated the opening of AT and GC pairs in various sequences. The data indicate marginal differences in the opening stiffness for GC and AT base pairs within the harmonic range (ca. ±25°). However, their opening angle was defined, roughly speaking, as the angle between the C1′−C1′ vector and the glycosidic bond projected onto the base pair plane, rather than as the symmetric opening angle between base-fixed coordinate systems and a mid-base triad as implemented in 3DNA.[22] Lavery and co-workers also simulated the distortion of only one base at a time, which corresponds to a simultaneous change of our opening and shear values.

Nevertheless, our results conform with their "opening" force constant (0.054 kcal/mol·deg^2, deduced from Figure 1 in Giudice et al.[6]).The same applies to the results of MacKerell and co-workers[4,5] (force constant 0.075 kcal/mol·deg^2 for a GC pair, from Figure 3 in Banavali et al.[4]). Similarly, Fuxreiter et al.[2] reported values of 0.028−0.047 kcal/mol·deg^2 for a combination of base pair opening and bending derived via unrestrained molecular dynamics.

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Table 1. Diagonal Force Constants for Rotational and Translational Base Pair Conformational Parameters (in kcal/mol·deg^2 and kcal/mol·Å^2, Respectively)[a]

	buckle	propeller	opening	shear	stretch	stagger
AT	0.0066 ± 0.0011	0.0098 ± 0.0016	0.022 ± 0.002	8.5 ± 0.3	42 ± 3	4.0 ± 0.5
GC	0.0090 ± 0.0019	0.0105 ± 0.0017	0.085 ± 0.007	8.1 ± 0.6	72 ± 6	5.9 ± 0.7
GC/AT ratio	1.36	1.07	3.86	0.95	1.7	1.48

[a] The average values and range of observed values are shown.

It is informative to compare the individual base pair force constants to those obtained at the base pair *step* level.[12] Buckle and propeller deformations of both pair types are softer than the most flexible diagonal angular bp-step parameter, that is the bp roll in the TpA dinucleotide steps (0.0136 kcal/mol·deg^2). The values of opening stiffness are comparable (AT) or higher (GC) than the tilt, roll, or twist stiffness (typically 0.02−0.05 kcal/mol·deg^2), while shear and stagger are approximately as rigid as rise (4−10 kcal/mol·Å^2, depending on the dinucleotide sequence). Stretch, which characterizes the elongation of the base pair parallel to its long axis, is much stiffer than any dinucleotide step translational parameter.

On the basis of our observations to date, one may ask whether the base pair deformation energetics can be understood using the obvious simple model where bases are represented as rigid plates with their hydrogen bonds as harmonic springs connecting them. A straightforward geometrical analysis of this model shows that, in the case of buckle, propeller, shear, and stagger deformation of a planar pair of parallel bases, the elongation of the springs depends only on the square of the corresponding conformational parameter. Since the deformation energy is proportional to the square of the spring elongation, it depends on the fourth power of the parameter change, which in view of their small fluctuations would be a minute contribution. Thus, factors other than base−base interactions within the pair (such as backbone flexibility or stacking interactions) may have substantial effect on these force constants. The span of their values for different instances of a given base pair type can then obscure the differences between the two base pair types, as seen in Table 1. By contrast, opening and stretch deformations result in proportional changes in the elongation of the springs and are thus supposed to depend more on the base−base interactions within the pair. This is again in line with our findings in Table 1, namely, the opening and stretch stiffness unambiguously depend on the base pair type. Further discussion of the results is included in the Supporting Information.

In summary, using atomic-resolution unrestrained molecular dynamics simulations and the method of fluctuations, we established a complete description of the sequence-dependent base pair deformation energetics in the harmonic approximation. We found that the dependence of the elastic properties on the base pair identity (AT vs GC) in B-DNA can be related to a simple "plates-and-springs" model. In our calculations, we assumed that base pair and base-pair step deformations are uncoupled. However, evidence is accumulating that underlines the importance of this coupling, especially between base pair opening and DNA bending.[2,6,7] Thus, a more general model including both levels would be desirable.

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Supporting Information Available: Full list of harmonic elastic constants and maximum energy errors for the AT and GC base pair deformation, details for the simulation and analysis protocol, and further discussion of the results. This material is available free of charge via the Internet at http://pubs.acs.org.

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