HYDRATION OF ds-DNA ans ss-DNA

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Hydration of DNA plays an important role in its structure, conformation and function, particularly in the process of recognition of drugs. The B form of DNA contains a relatively large number of water molecules: around 18 per nucleotide. Depending on their position along the double helix, the behaviour of each water molecule is different, especially the dynamics of diffusion and rotation.

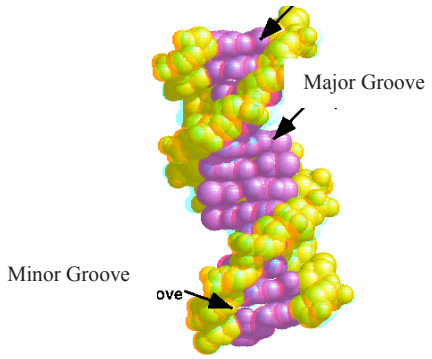

Figure 1. Schematic view of the B form of ds-DNA showing the two grooves.

Studies performed by X-rays have shown that the minor groove (fig. 1) is hydrated in an extensive and regular manner with a spine of first and second hydration shells. In contrast, the hydration of the major groove is reduced to a monolayer [1]. Differential scanning calorimetry studies of samples of double stranded (ds) and single stranded (ss) DNA[2] show that part of the hydration water does not freeze when cooled down at low temperatures. This amount of "unfreezable" water is equal to 0.55 g H_2O/g ds-DNA and 0.40 g H_2O/g ss-DNA. Logically, the difference between this two quantities (0.15 g H_2O/g DNA) corresponds exactly to the dehydration of ds-DNA at the transformation double-stranded helix → single-stranded chains. This difference is also equal to the hydration change during the thermal denaturation of ds-DNA. All these features were interpreted as suggesting that the thermal transition from double helix to single strand is accompanied by a disruption of the ordered part of the water present in the hydration shell of the double helix.
Incoherent quasi-elastic neutron scattering probes the dynamics of hydrogen atoms and is an ideal technique to study the behaviour of hydration water.

It is assumed that different hydrogen atoms can be classified into three classes:
a) a fraction p belonging to DNA, which are immobile in the experimental time window.
b) a fraction $(1-p)q$ belonging to water molecules strongly attached to DNA.
c) the fraction $(1-p)(1-q)$ of remaining hydrogen atoms, belonging to water molecules that can diffuse and rotate in the vicinity of the DNA.
The resulting scattering function $S(Q, \omega)$ is:

$$S(Q, \omega) = [p+(1-p)A_0(Qa)]\delta(\omega)+(1-p) \times(1-A_0(Qa))[qL_1(\omega)+(1-q)L_2(\omega)]$$

where $L_1(\omega)$ and $L_2(\omega)$ represent respectively a narrow and a broad Lorentzian functions, $\delta(\omega)$ is the Dirac distribution and $A_0(Qa)$ represents the confined diffusion of the atom within a sphere of radius a, according to a model due to F. Volino and A.J. Dianoux [3]:

$$A_0(Qa) = [3j_1(Qa)/(Qa)]^2$$

where $j_1(Qa)$ is the first order spherical Bessel function.
Two different configurations, corresponding to two instrument resolutions of the spectrometer Mibémol allow the evaluation of the two Lorentzians. With the lowest resolution, the only motions that can be observed are those of the class c) of hydrogen atoms. In this case, atoms of class b) are seen as if they were immobile and the scattering function is:

$$S(Q, \omega) = [P+(1-P)A_0(Qa)]\delta(\omega)+(1-P) \times (1-A_0(Qa))L_2(\omega)$$

where $P = p+q(1-p)$.
The first term is an experimental elastic incoherent structure factor (EISF). Its asymptotic value at large values of Q is equal to P and, because p is deduced from the chemical composition, the different fractions of water molecules are experimentally determined. The molecular motions are deduced from the width of the Lorentzians. In figure 2 the experimental EISF for hydrated ds-DNA and ss-DNA are plotted as a function of Q.

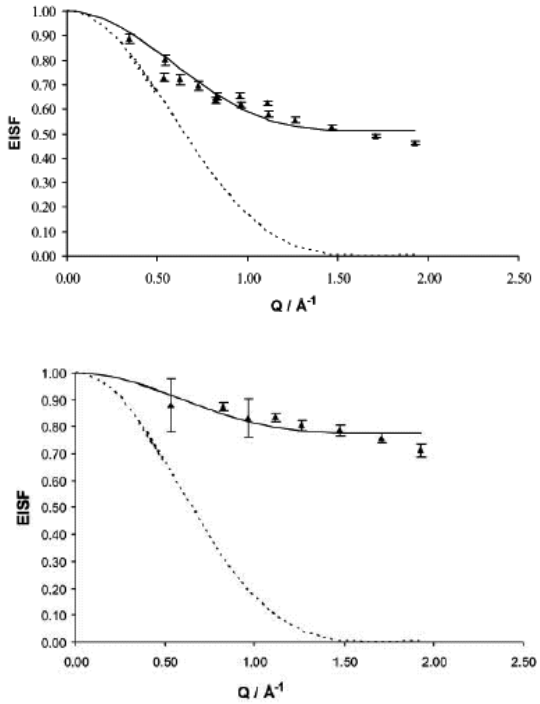

Figure 2. EISF for ds-DNA (upper plot) and ss-DNA (lower plot). Symbols are experimental points, the solid lines are the experimental fit and the dotted lines represent the function $A_0(Q, \omega)$ with a= 2.8 Å.

The following values of p and q can be deduced:

	ds	ss
p	0.32	0.78
q	0.28	0.0

The half-width at half-maximum $\Gamma_{1/2}$ of the Lorentzian $L_2(Q, \omega)$ is plotted in Figure 3 for ds-DNA. The effect of confinement is clearly seen, because at small values of Q the line-width goes to a constant value. The model of confined motion [3] gives the value of the self-diffusion constant $D = 2.37 \times 10^{-5}$ cm^2 s^{-1}, very close to the self-diffusion of bulk water, but the radius of the sphere of confinement is a=2.8 Å, what means that the dynamics of the water molecules is reduced to rotational and diffusive motions along distances of the order of magnitude of the molecular size.

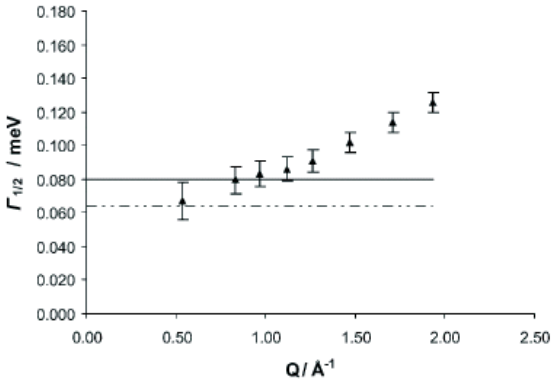

Figure 3. Half-width at half-maximum of the broader Lorentzian for ds-DNA.

Instead, the Lorentzian $L_1(Q, \omega)$ is absent for the sample ss-DNA, what corresponds to the absence of water molecules strongly bounded and is in agreement with the value of $q=0$ deduced from the Q dependence of the EISF.

Finally, the value q=0.28 obtained for ds-DNA (see Table above) corresponds to 0.16 g H_2O/g DNA, i.e. almost exactly the value deduced by calorimetry for the water release in the transformation ds-ss.

We conclude that, at this limiting hydration value (B-DNA) two types of water molecules are present in ds-DNA. About 72% of hydration water is confined, probably inside the grooves, with a local diffusion coefficient D not very different from that of bulk water. The remaining hydration water (28%) is strongly attached to the ds-DNA. Its dynamic is very slow, probably reduced to reorientations resulting from hydrogen bond dynamics. This water is lost upon ds-DNA thermal denaturation, demonstrating that the thermal transition between ds-DNA and ss-DNA is accompanied by disruption of the ordered water fraction in the inner hydration of the double helix and stresses the importance of hydration effects on the maintenance of the double-helix structure.

References

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