Structural Responses of DNA-DDAB Films to Environmental Triggers

Thorsten Neumann[1], Surekha Gajria[2], Luc Jaeger[2], Matthew Tirrell[1].
[1]Bioengineering Dept., University of California Berkeley, [2]Chemistry Dept., University of California Santa Barbara.

Statement of Purpose: We recently described the distinctive structural transition of a DNA-dimethyldidodecylammonium bromide (DNA-DDAB) film in response to the water content in the surrounding environment (Neumann *et al.* 2009). Such a unique structural change prompts further investigation into its cause and the extent to which it can be manipulated. We have explored the structural response of DNA-DDAB films to environmental changes, particularly temperature and humidity, in greater detail, and found that we are able to direct the lamellar structure of the film into three distinct states by adjusting parameters of temperature and humidity: double-stranded DNA (dsDNA) paired with interdigitated DDAB bilayers (bDDAB), single-stranded DNA (ssDNA) with DDAB monolayers (mDDAB), and ssDNA with bDDAB. The lamellar structure can be reversibly altered from ssDNA to dsDNA and/or from mDDAB to bDDAB.

We found that the transition of DNA and DDAB is concerted and follows apparent first order kinetics. We also found that the structure and switching behavior of the films are affected by inclusion of either nucleic acid intercalating molecules (e.g. ethidium bromide) and bilayer intercalating molecules (e.g. cholesterol), and that the film can disassemble in the presence of buffer solutions. We conclude that the structure of DNA in the film depends on the water content (as measured by the relative humidity) and temperature of the environment, while the state of DDAB depends essentially only on the water content. The structure of the film is flexible and can be altered by changing environmental conditions as well as the chemical ingredients. We expect that these films will have useful, new applications as responsive materials in drug delivery.

Methods: Aqueous solutions of DNA and DDAB were mixed and the precipitated complex was purified, dried and dissolved in isopropanol. The solution was used to cast Films. Atomic force microscopy (AFM), small-angle X-ray scattering (SAXS), Fourier-Transform infrared- (FT-IR-), fluorescence, and circular dichroism-(CD-) spectroscopy measurements were performed with dry as well as (heated) wet films to determine the structure and its response to various environmental conditions. Degradation experiments were performed with phosphate-buffered saline (PBS).

Results: The structural states of DNA and DDAB in the film were monitored directly by the carbonyl bands at 1650 and 1695 cm^{-1} using FT-IR spectroscopy as the film was wetted and dried (Neumann *et al.* 2009). By this technique we were able to determine the complete transition profile of the film switching as well as the cooperativity of the switching of DNA and DDAB. We determined the apparent rate constant for both components to be 0.014 ± 0.002 $seconds^{-1}$ on average, indicating that their structural transition is cooperative. A similar rate constant within the range of error was also determined in synchrotron SAXS studies. FT-IR and SAXS measurements were used to determine the minimum amount of surrounding relative humidity at 60% R.H. to initiate the switching process. Temperature studies performed by CD spectroscopy and SAXS measurements indicated that the DNA in the wet film denatures to two singles strands in the presence of DDAB when heated. All states from ssDNA/mDDAB, dsDNA/bDDAB, and ssDNA/bDDAB could be repeatedly achieved by changing these environmental parameters. Small molecules which penetrate into either dsDNA or bDDAB can interact with the switching process, as ethidium bromide tends to delay the transition of DDAB from a bilayer to a monolayer, supposedly due to the hydrophobic nature of ethidium bromide which inserts between the DDAB tails and hinders their separation in a drying film. Cholesterol however tends to delay the switching of DNA relative to DNA due to its ability to retain water within the film.

We assume that the rate of the DNA denaturation to ssDNA and annealing back to complementary dsDNA is due to the limited diffusion of long DNA moleucles within the film, ensuring the ability to form the preferred wet or dry structural conformation in a relatively short amount of time.

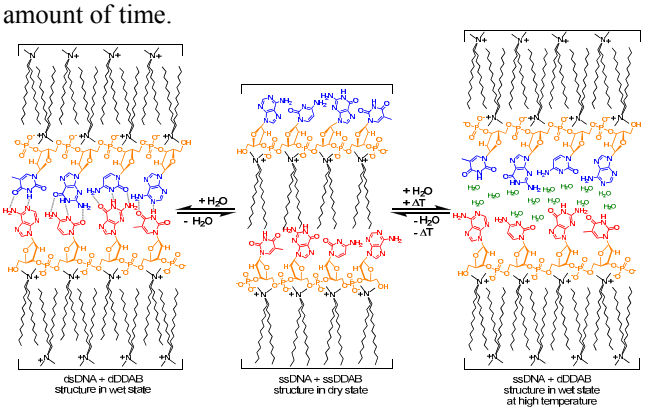

Figure 1. Expected states of DNA and DDAB in the cast film which can be reversibly altered by the environment.

Conclusions: The structure of a DNA-DDAB film can exist in one of three states, depending on the environmental temperature and water content (relative humidity). The apparent rate constants of DNA and DDAB calculated by observations of switching indicate that the switching of the two components is concerted and follows first-order kinetics. The structure and switching behavior of the film can also be altered by the inclusion of small molecules.

References:

Neumann T. J Am Chem Soc. 2009:131:3440-3441.