Following the phase transitions (from hexagonal to cholesteric and isotropic) of a unique DNA chain in the capsid of the bacteriophage T5

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Tailed bacteriophages are complex macromolecular machineries that deliver their genome into the host cytoplasm while their capsid and tail remain bound to the cell surface. Although several models have been proposed, DNA organization is still unknown in the phage head but dimensions of the capsids are adjusted to keep DNA at a concentration close to 500mg/ml independent of the species. DNA ejection from the capsid is triggered by specific interaction of a phage tail protein with a receptor inserted in the wall of the bacteria. DNA progresses in the tail and is injected into the cytoplasm. The bacterial receptor of bacteriophage T5 has been isolated, allowing to reconstitute the ejection process *in vitro* (1) and to investigate the underlying mechanisms. The T5 genome is almost 40µm long (113.9kbp) and confined in a capsid 80nm in diameter.

Using cryoElectron microscopy (cryoEM), we followed the organization of DNA inside the capsid at different steps of the ejection process and correlated these observations with the lengths and concentrations of encapsidated DNA. We show that the DNA chain always occupies the total volume of the capsid and reorganizes under confinement. The single DNA chain undergoes several phase transitions, from constrained hexagonal to hexagonal, cholesteric and isotropic phases (2) following the sequence reported for solutions of short DNA fragments (3).

The confinement and bending of the long double-stranded DNA chain inside the small volume of the capsid is considered to be responsible for DNA release after interaction with the receptor protein, in the absence of any external source of energy. We explored the effect of opposing an pressure (7-16 Atm) outside of the bacteriophage and showed that in T5, the ejection is more complex than an equilibrium of pressures between the inside and the outside of the capsid (4).

The interactions between DNA strands can also be monitored and turned from repulsive to attractive by addition of multivalent cations that diffuse through the capsid. After partial ejection of DNA, we induce the collapse of each individual DNA chain (3000 to 55000bp long (1.4-18µm)) inside the volume of the bacteriophage capsid. We form toroids and related shapes whose structures are analyzed. We show how the frustration arising between chirality and hexagonal packing combined with the strong curvature imposed by the small volume of the container impose phasing of the helices and variations the DNA helical pitch.

References

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