Supporting Information

Hepatitis B Virus Capsid Completion Occurs through Error Correction

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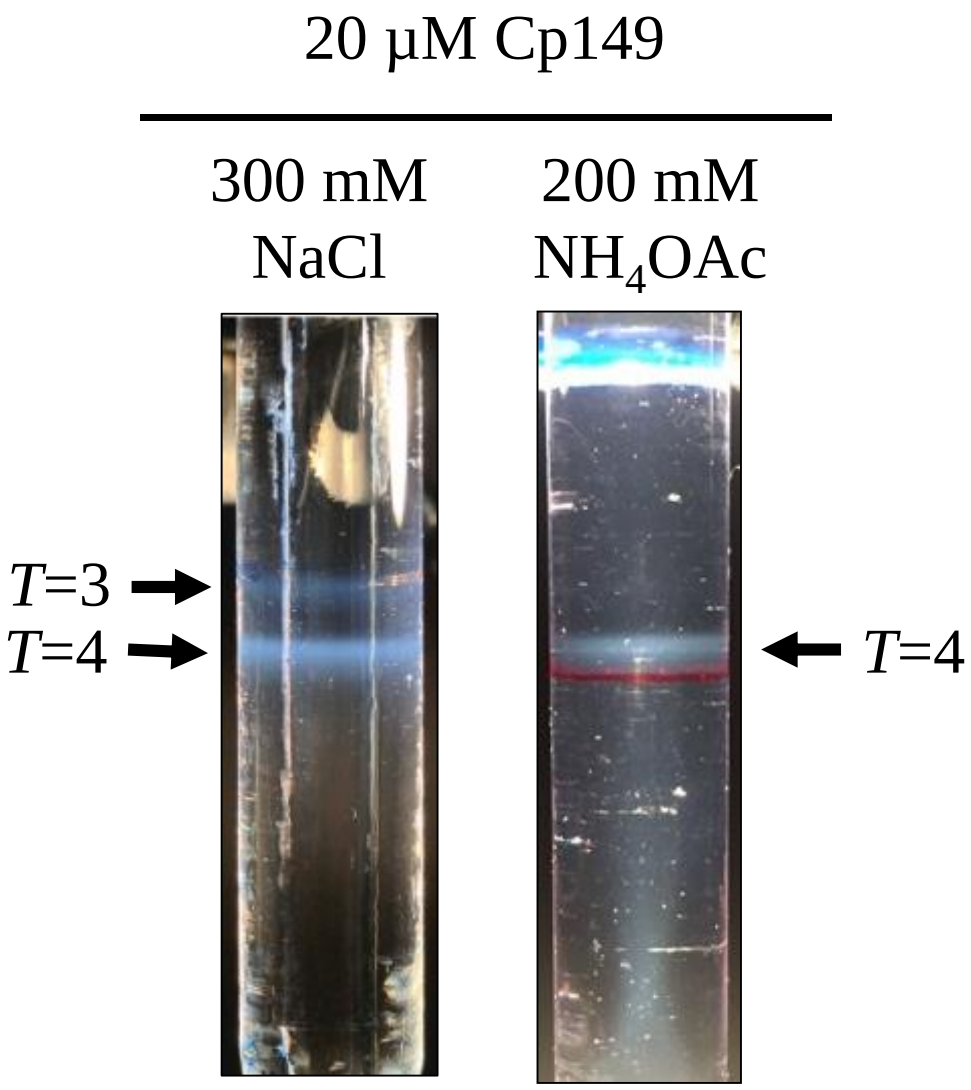
Further Evidence for Negligible Concentration of *T*=3 Capsid from Assembly in Ammonium Acetate



Supplemental Figure 1. Sucrose gradient centrifugation shows two distinct bands corresponding to *T*=3 and *T*=4 capsids when 20 µM Cp149 is assembled in 300 mM NaCl. Assembly in 200 mM NH_4OAc shows that mostly *T*=4 capsids are formed as indicated by the single band observed.

Analysis of Non-Specific Aggregation During Electrospray

The large ions studied in this work are thought to be generated by the charge residue mechanism where the solvent evaporates from the electrospray droplet leaving behind a charged ion.[1] The electrospray droplet may contain more than one species (such as a dimer and a capsid) that can form a complex when the solvent evaporates. Such complex formation could contribute to the higher than expected masses determined for the capsid. To explore this possibility, we estimated the probability that a dimer is incorporated into the same droplet as a capsid. Ignoring the excluded volume of the capsid, this is the same as the probability that a dimer is incorporated into any droplet, which is given by a Poisson distribution:

$$P(k,\lambda) = \frac{e^{-\lambda}\lambda^{k}}{k!} \quad \textbf{(1)}$$

$P(k,\lambda)$ is the probability that a droplet contains k dimers, and λ is the mean number of dimers per droplet (the dimer concentration in molecules/m^3 divided by the droplet volume). The diameter of the primary electrospray droplet (68 nm) was estimated from the solution and electrospray conditions (density, surface tension, conductivity and flow rate).[2,3] With a free dimer concentration of 3.55 µM, the probability of one or more dimers occupying a primary droplet is 0.30. Summing the contributions for one or more dimers in the droplet, the average mass shift is expected to be 12 kDa. This assumes that the extra dimers form a complex with the capsid and are not lost during the transition into the gas phase. In the experiments, the center mass of the capsid peak decreased, after around 36 hours, to around 4.05 MDa. This mass still exceeds the sequence mass by around 25 kDa. Based on the preceding estimate it appears that up to half of this extra mass could be attributed to a capsid-dimer complex. The balance is presumably due to solvent (water and ammonium acetate) that is trapped in or on the capsid.

References

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