# Measurements of Pressurized DNA in Phage Capsids

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#### Abstract

Bacteriophage  $\lambda$  packages its genome into a tiny capsid, creating an internal pressure of 10–20 atm. This pressure may be required during ejection to overcome the 3 atm of osmotic pressure inside the host cell. We induce ejection *in vitro* in the presence of an external osmotic pressure that mimics the cell interior. Using strains of  $\lambda$  with different genome length, we quantify the relationship

genome length  $\rightarrow$  ejection pressure

We find that a parameter-free theoretical model can predict this relationship fairly well.

#### Phages are champion DNA packers

Bacteriophage DNA packaging:



(assuming dsDNA is a 2 nm diameter cylinder and using published genome lengths and capsid sizes)

#### Phages with several genome lengths

We use two  $\lambda$  mutants differing only in genome length to reveal the relationship between genome length and ejection pressure.



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## The experiment: Osmotic pressure stops ejection

LamB triggers ejection of DNA

PEG8000 creates osmotic pressure, stopping ejection

DNase I cuts DNA into small fragments

Capsids are spun out in centrifuge, leaving only ejected DNA

Measure DNA with an ultraviolet spectrophotometer



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# **Theoretical Model**

1: Electrostatic forces

 $\Pi = F_0 \exp\left(\frac{d}{C}\right)$ 

(X-ray scattering experiments by Rau, Lee, & Parsegian 1984)

2: Bending forces

E = L - E

Minimize free energy as a function of d, differentiate energy to get the ejection force. Pressure = Force  $\times \pi (R_{DNA} + R_{DEC})^2$ 

## Results: pressure vs. percent **DNA** ejected

The theoretical model makes quantitative predictions for the amount of DNA ejected at every pressure, with no free parameters, so we can compare it directly to the experiment:



The theory predicts the right magnitude for the pressures and explains the dependence of force on genome length.

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osmotic pressure (atm)