Nanoindentation studies on viral particles

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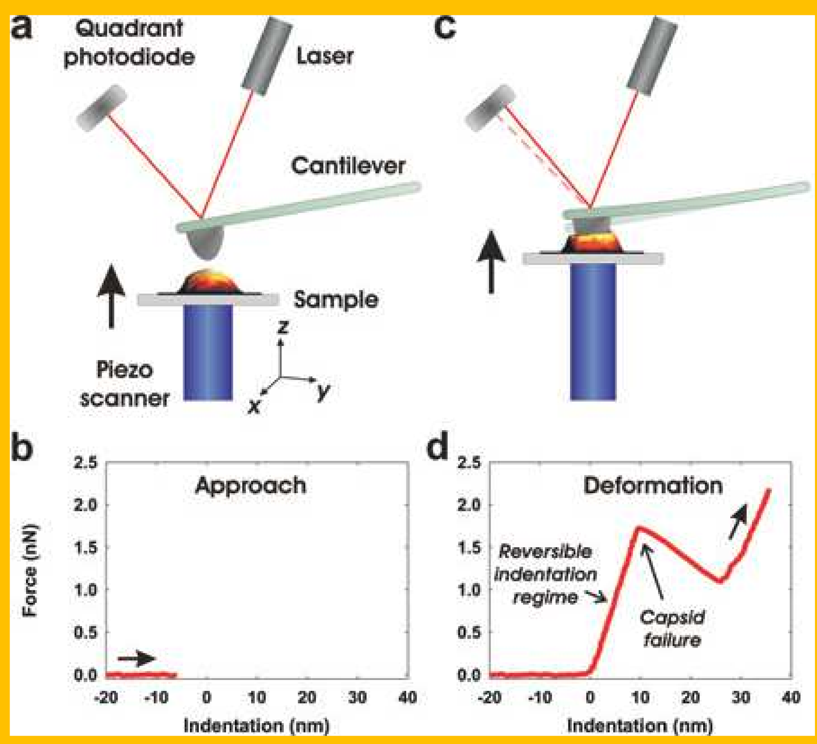
Jumping mode AFM & Force Spectroscopy

In jumping mode the lateral drag forces are avoided by performing the cantilever displacement when the tip and sample are not in contact. During imaging with jumping mode the tip performs a rapid succession of force-distance curves. At each point the tip approaches the object until the set force is reached and then retracts from the object.

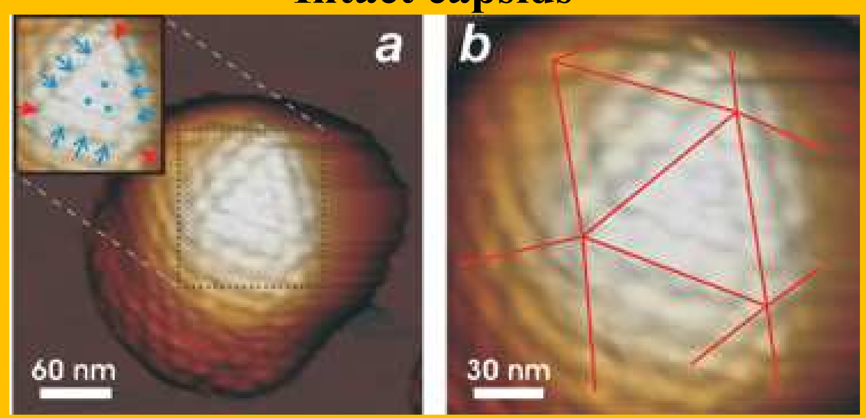
Force Spectroscopy measurements on viruses are performed by indenting a viral capsid, a so-called nanoindentation experiment.



Roos *et al.*,Nature Physics (2010), Vol. 6, p733

Herpes Simplex Virus1

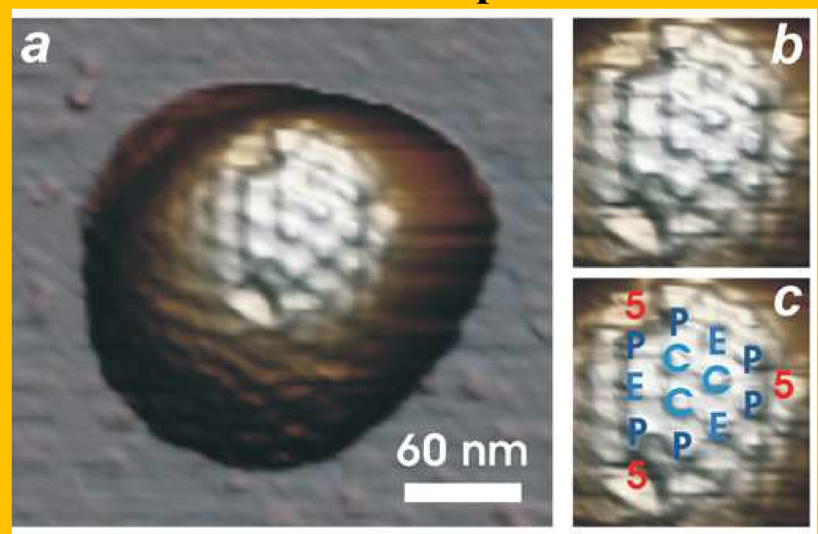
Intact capsids



Roos *et al.* PNAS (2009) Vol. 106, p9673

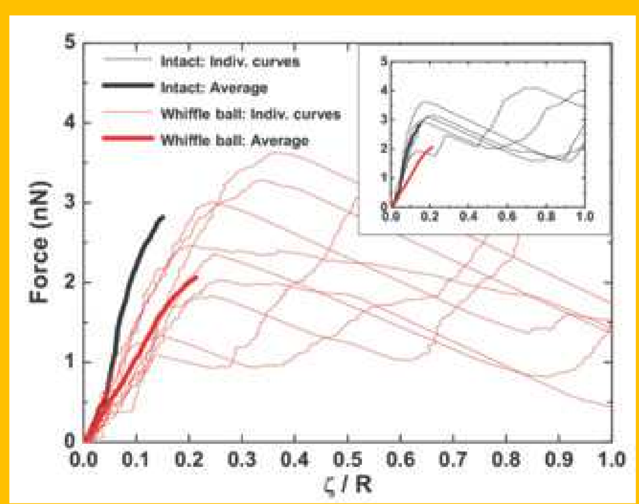
Capsomeres can clearly be observed on the AFM images. The pentamers (red) and hexamers (blue) and the icosahedral faces are shown.

Pentonless capsids

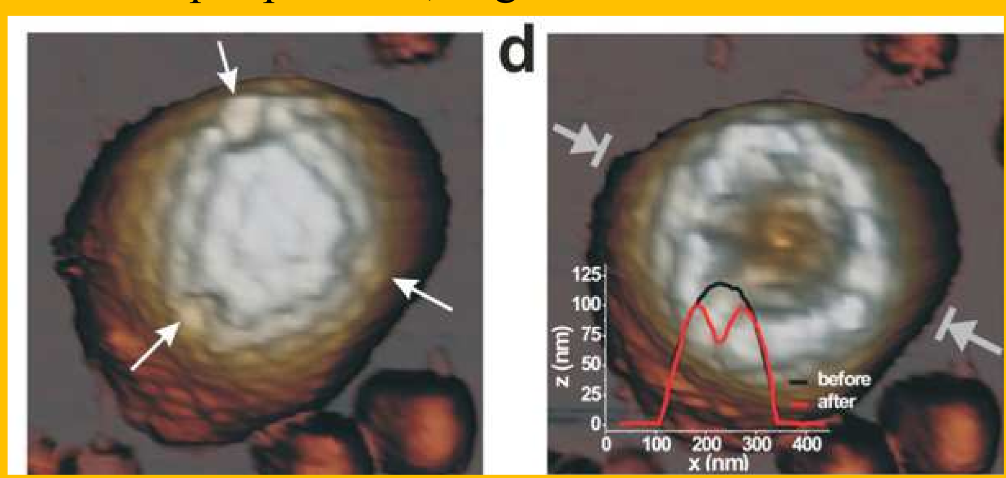


Roos *et al.* PNAS (2009) Vol. 106, p9673

Image showing three holes at the position of the missing pentons (denoted with a 5 in c and d). The pentons are removed with 2 M GuHCl. P, E and C stands for peripentonal, edge and central hexon



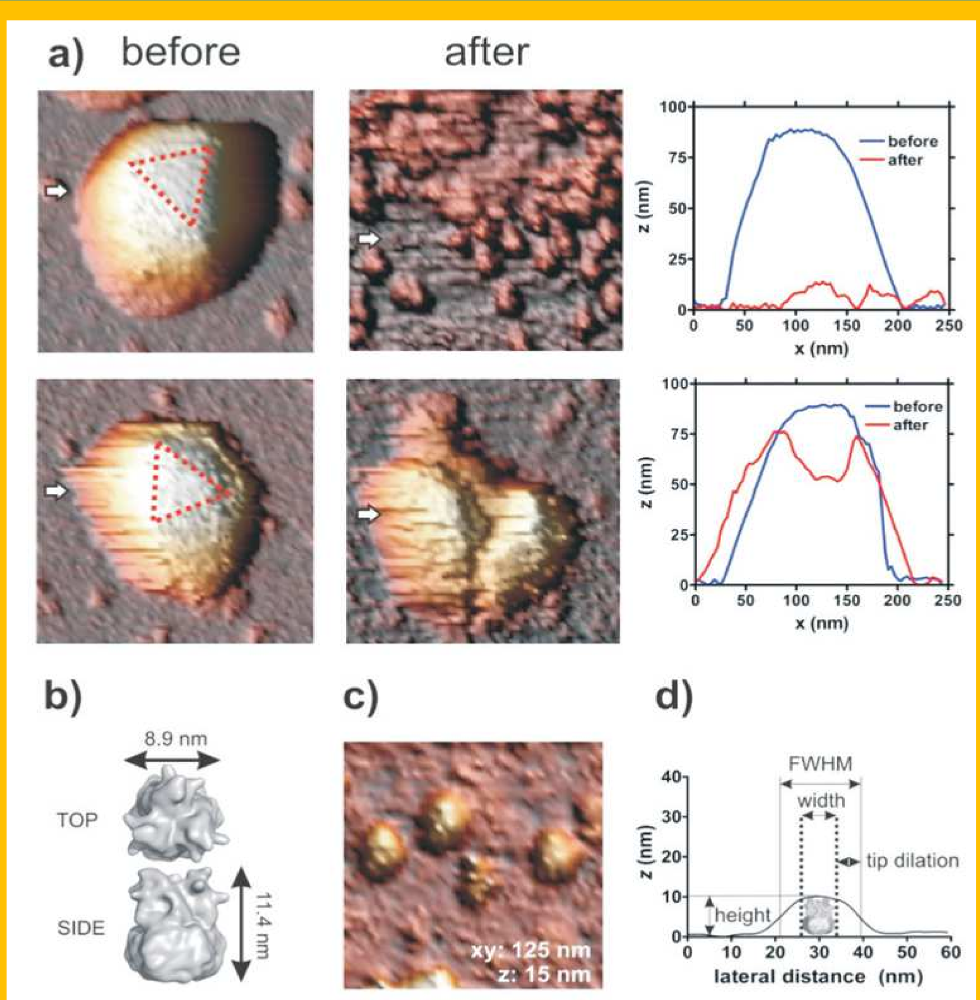
Indentation curves (thin lines show individual curves) and their averages (thick lines) for pentonless and intact capsids



Klug *et al.* PRL (2012) Vol. 109, 168104

Pentonless particle before and after indentation

Adeno Virus

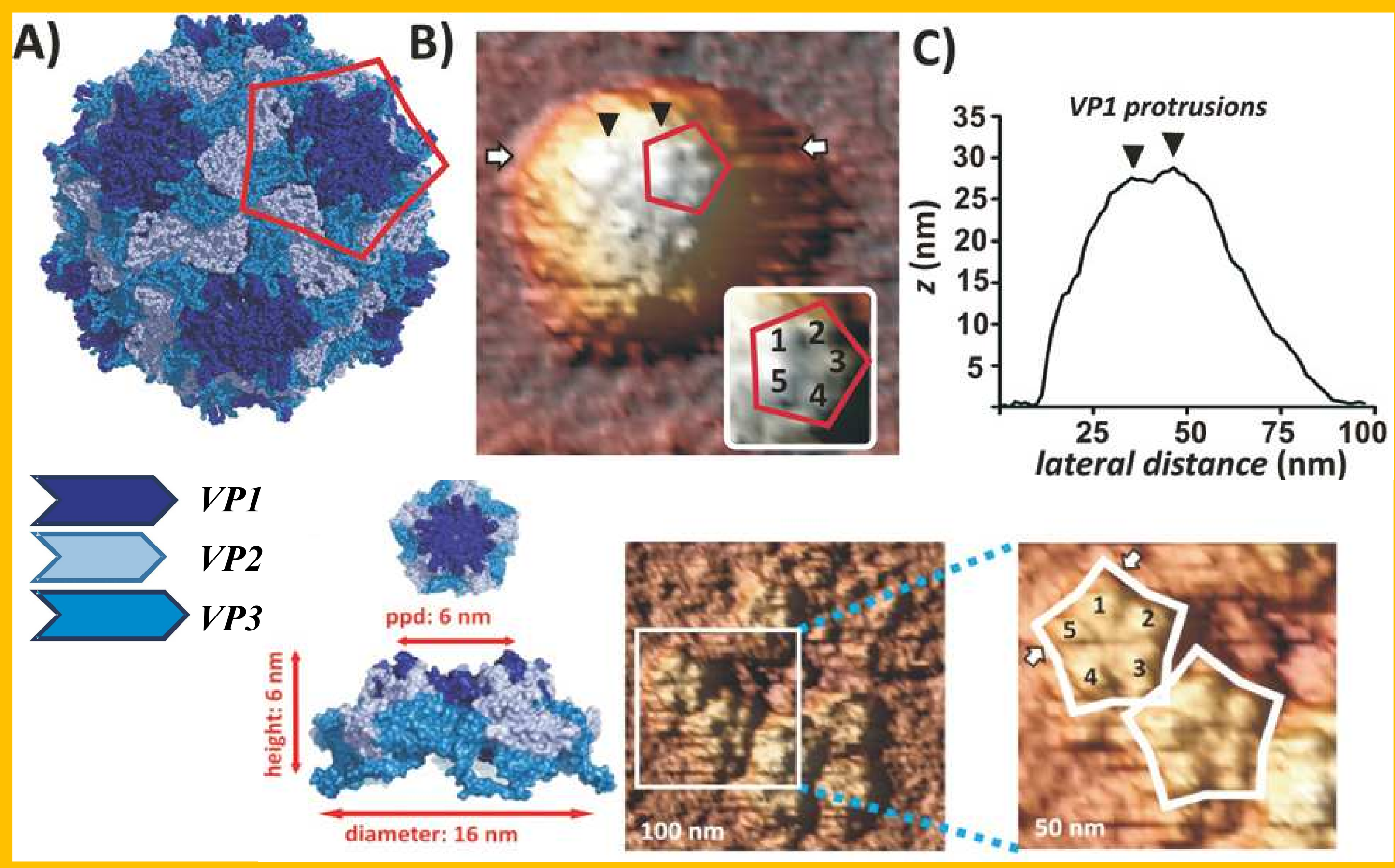


Snijder *et al.* J Virol (2013) Vol. 87, p2756

a) Ad35F particles subjected to mechanical failure. The top row shows a particle that completely disintegrated into individual smaller particles in response to mechanical failure. Left is the particle before, right is the particle after nanoindentation. The corresponding height profiles are taken along the white arrows indicated in the images. The bottom row shows a particle that partially maintained capsid integrity upon mechanical failure. **b)** Surface rendering of an individual hexon trimer as described in the crystal structure of Ad35F; pdb 1VSZ. **c)** AFM images of the smaller particles that arise from mechanical failure of Ad35F. **d)** A typical height profile of force induced disassembly products.

- Virus-host cell molecular interactions impact the mechanical properties of the virus capsid.
- Host cell molecules with opposite effects on capsid disassembly modulate the elastic response of the HAdV virion accordingly.
- We uncovered a direct link between capsid disassembly, genome uncoating and the mechanical properties of virus capsids.

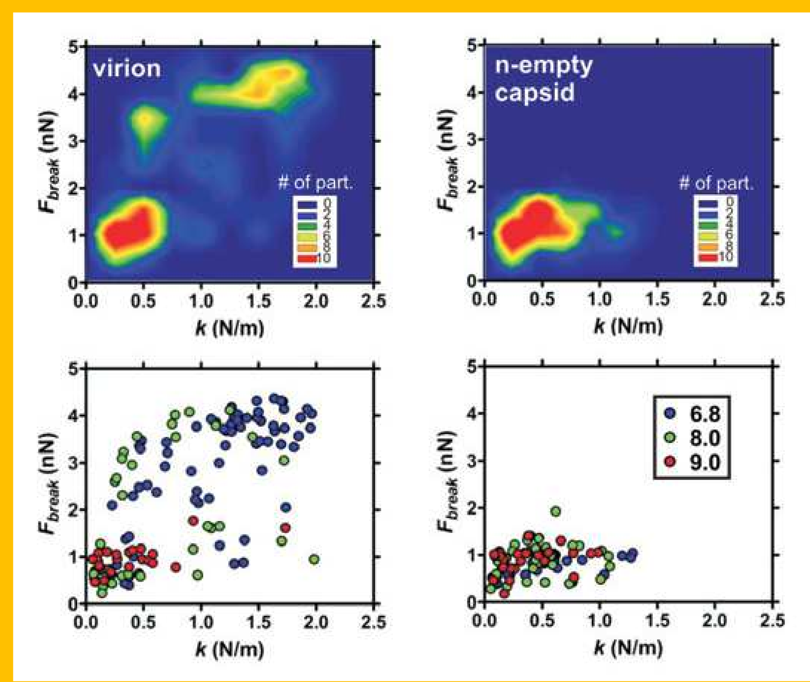
Triatoma Virus



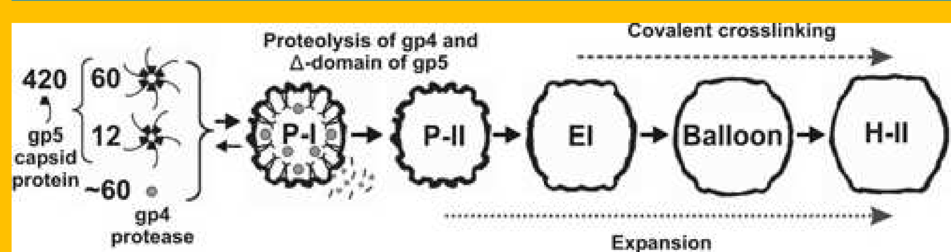
Snijder *et al.* Nat Chem (2013) Vol. 5, p502

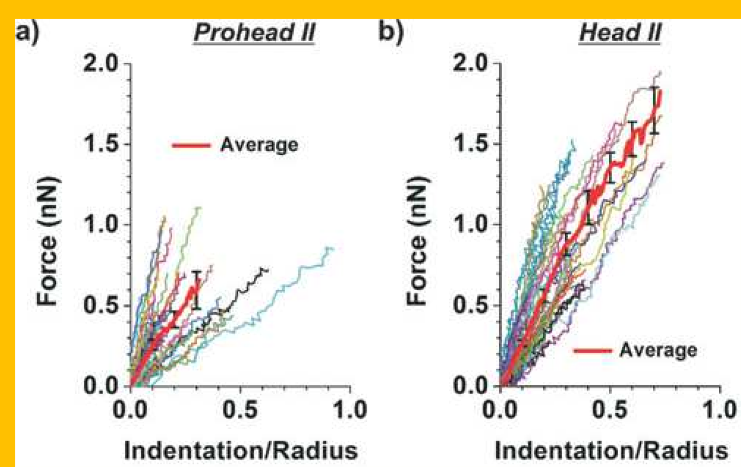
The AFM imaging results show pentamers before and after nanoindentation

Left panels show the 2D density distribution (k vs. F_{break}) of single virions (left) vs. n-empty capsids (right), at pH 6.8, pH 8 and pH 9. Bottom panels show the individual points at different pH, from which the density distributions were calculated. The virion under neutral pH has a higher spring constant and breaking force than the empty capsid. With increasing pH, the spring constant decreases but the high breaking force is maintained, resulting in a discrete intermediate state. Consistent with genome release to yield empty capsids, all particles are mechanically indistinguishable from n-empty capsids at pH 9.

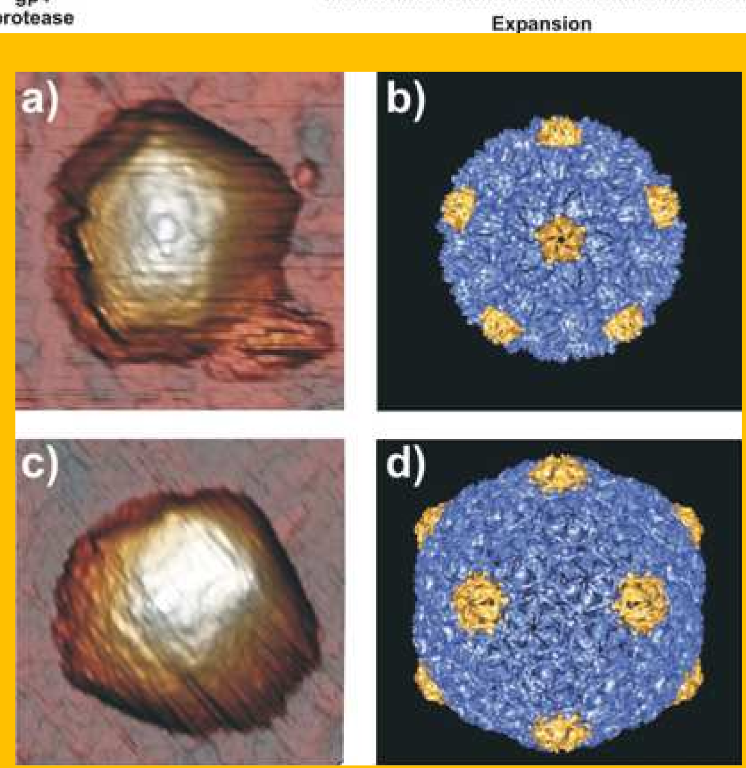


Phage HK97





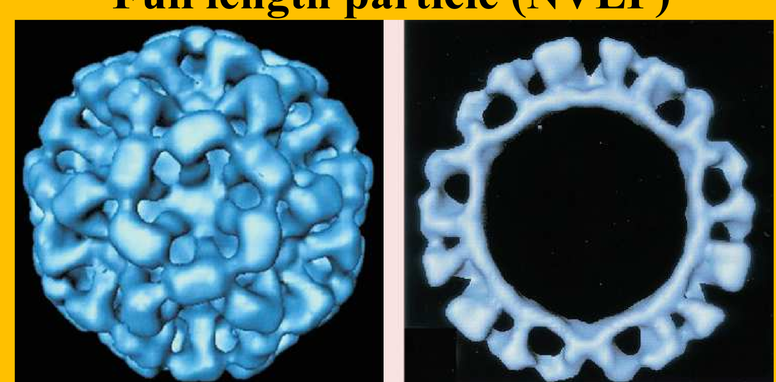
Roos *et al.* PNAS (2012) Vol. 109, 2342

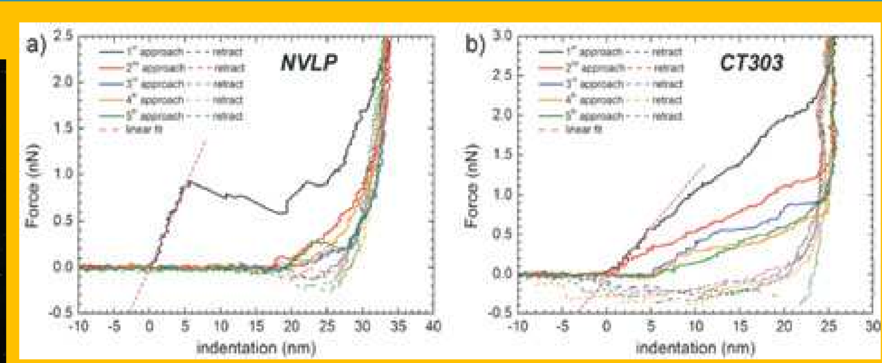


The AFM nanoindentation experiments on bacteriophage HK97 show a surprisingly complex maturation pathway in which the fragile Prohead II (P-II) is being strengthened by: i) an increase in Young's modulus, ii) an increase in ultimate strength and iii) astance against material fatigue. The resulting particle is called Head II (H-II).

Norwalk virus

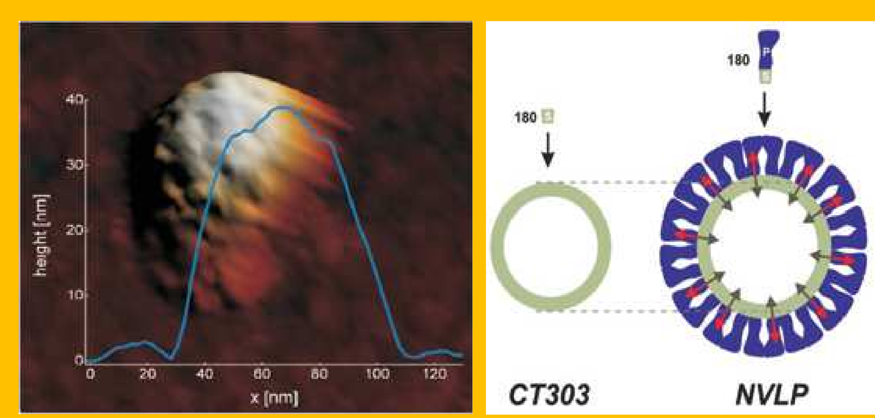
Full length particle (NVLP)





Baclayon *et al.* Nano Letters (2011) Vol. 11, p4865

CT303 Mutant



Bertolotti *et al.* J Virol. 76, p4044

AFM measurements on the Norwalk virus show the unexpected differences in mechanical behaviour of the thin shell mutant particle (CT303) with respect to the NVLP. This is in apparent contradiction to continuum elastic theory. Our results indicate that the "bridge-like" structures on the latter yield a particle under isotropic pre-stress that strenghens the virus.

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