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Viruses

Seminar

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Abstract:

In this seminar we first take a look at the viruses in general, then we learn something about viral self-assembly and about a model of a viral capsid. Finally, we describe the compression of virus-like particles carrying surface dipole.

Contents

1 Introduction

Viruses are often considered as something dangerous and unnecessary, because they cause a lot of diseases. However, if we know well their properties, they can be used in many positive roles, even in cancer treatment.

Viruses are really interesting objects and a lot about them is still unknown, so there is enough work for all researches (physicists, biologists …).

Physical virology deals with thermodynamic of self-assembly, mechanical properties of viral shells etc.

2 Viruses

A virus is a very small infectious object. Viruses can infect animals, plants, bacteria, human beings etc.

They consist of two parts: genetic material (DNA or RNA) and a capsid. The capsid is a protein coat which protects genome. Viruses can be found in many shapes. The most common are sphere-like and rod-like, but spherocylinders, cones and other shapes can also be seen. A lot of viral families have icosahedral symmetry (half of all families), but genomes of these families are not similar.
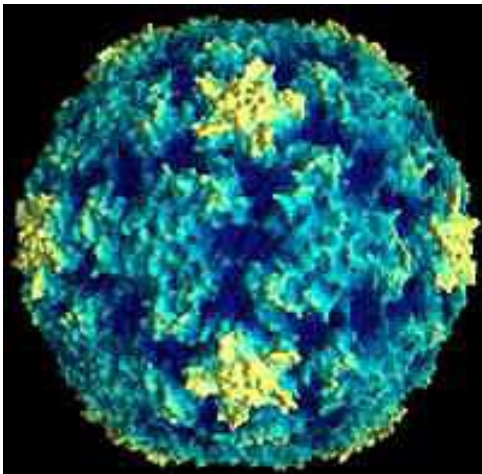

Figure 1: Cold virus
A scanning electron microscope image of rhinovirus (number 14). They are extremely small (few tents of nanometer). [1]
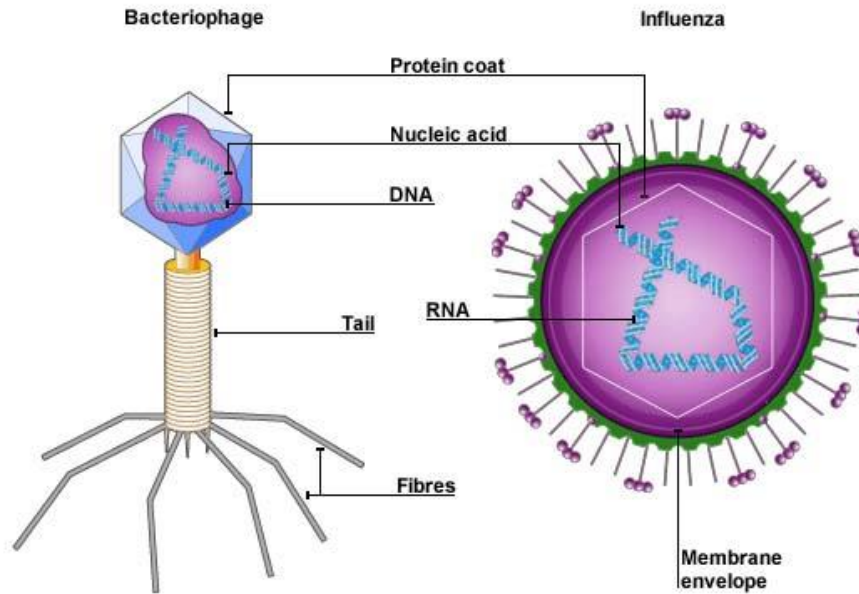
Figure 2: Two more examples of viruses: a bacteriophage (which attacts only bacteria) and an influenza virus. [2]

There is the Baltimore classification which places viruses in seven groups, based on the type of genome [3]:
I: dsDNA viruses (e.g. Herpesviruses)
II: ssDNA viruses (e.g. Parvoviruses)
III: dsRNA viruses (e.g. Reoviruses)
IV: (+)ssRNA viruses (e.g. Picornaviruses, Togaviruses)
V: (−)ssRNA viruses (e.g. Orthomyxoviruses, Rhabdoviruses)
VI: ssRNA-RT viruses (RNA with DNA intermediate in life-cycle e.g. Retroviruses)
VII: dsDNA-RT viruses (e.g. Hepadnaviruses)

We have a classification system for icosahedral viruses, which was introduced by Caspar and Klug (CK) and it is based on the 'T number', which is defined [4]:

$$T = m^2 + n^2 + mn, \qquad\qquad (1)$$

m and n represent the number of steps along crystallographic directions of a hexagonal lattice.
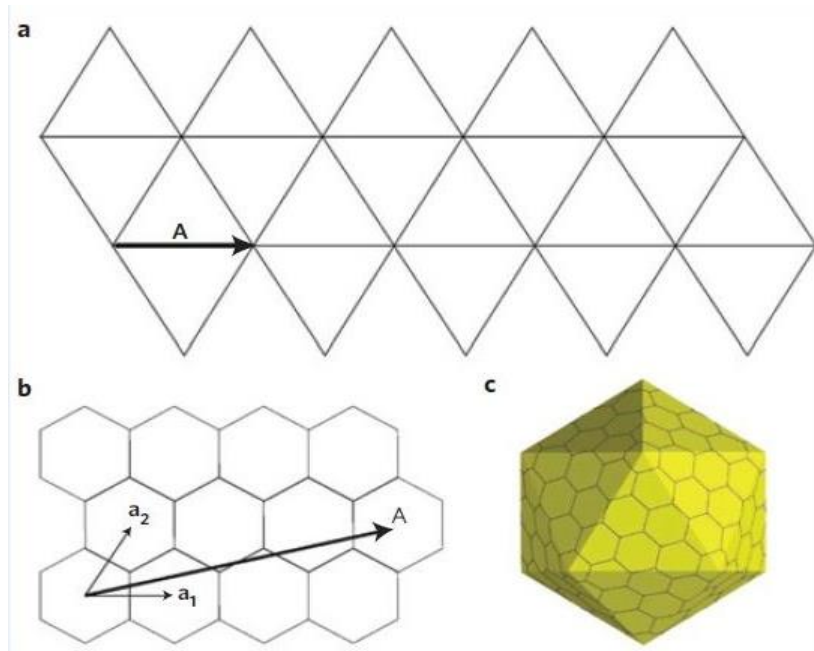
Figure 3: Icosahedral viral shells

a, The network of equilateral triangles which make an icosahedron with the lattice vector $A = ma_1 + na_2$. **b,** An example for $m = 3$ and $n = 1$. **c,** An icosahedron from **a.** It has $T = 13$. [4]

An icosahedral shell consists of 12 pentamers and $10(T - 1)$ hexamers, with $T = 1, 3, 4, 7, ...$ located between pentamers.

More than 80 different viruses (with T number from 1 to 25) were analysed and it was found out that area of a protein capsid is quite conserved ($\sim 25 \ nm^2$). The thickness of the protein is from 2 to 5 nm. [5]
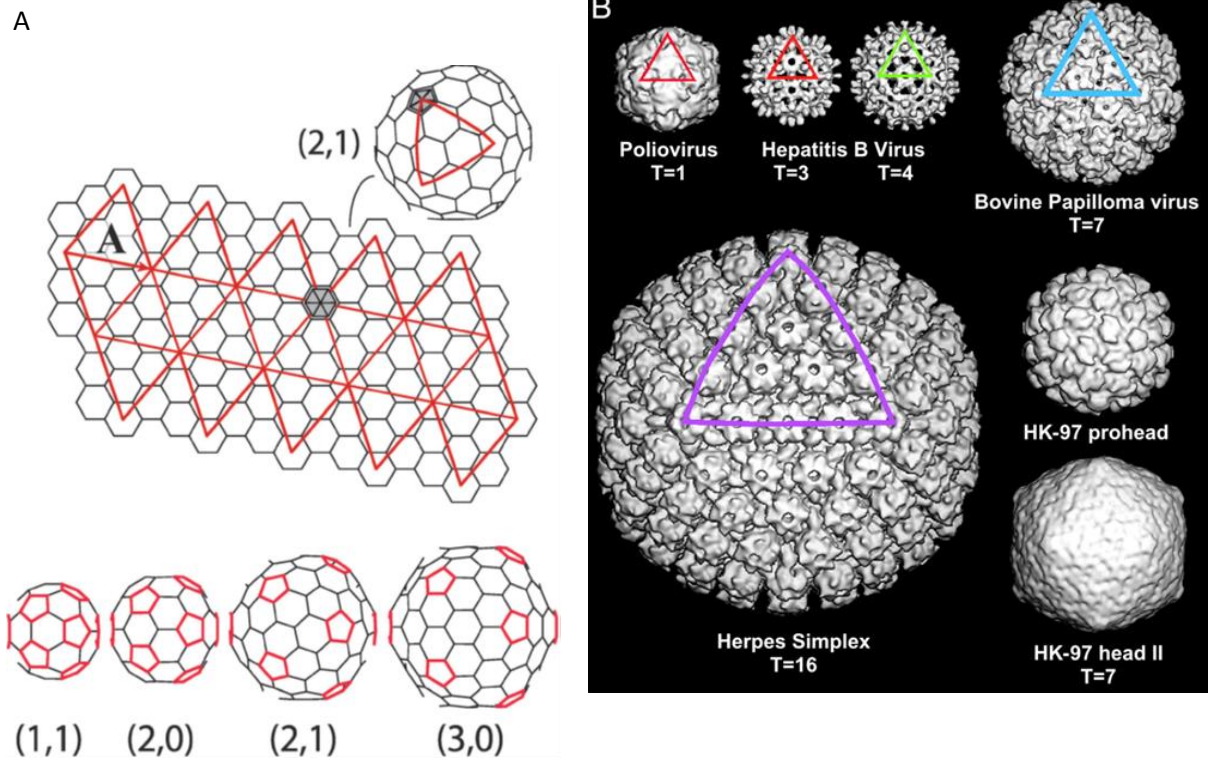
Figure 4: Some more examples of viruses with different *m* and *n* (**A**, [5]) and *T* (**B**, [6])

3 Viral self-assembly

Viruses do not have any metabolic activity and rely only on host cell and its metabolism. In 1955 some scientists discovered that tobacco mosaic virus (TMV) self-assembles into a fully infectious virus from solution containing the components of the virus: the capsid proteins and ssRNA genome molecules.
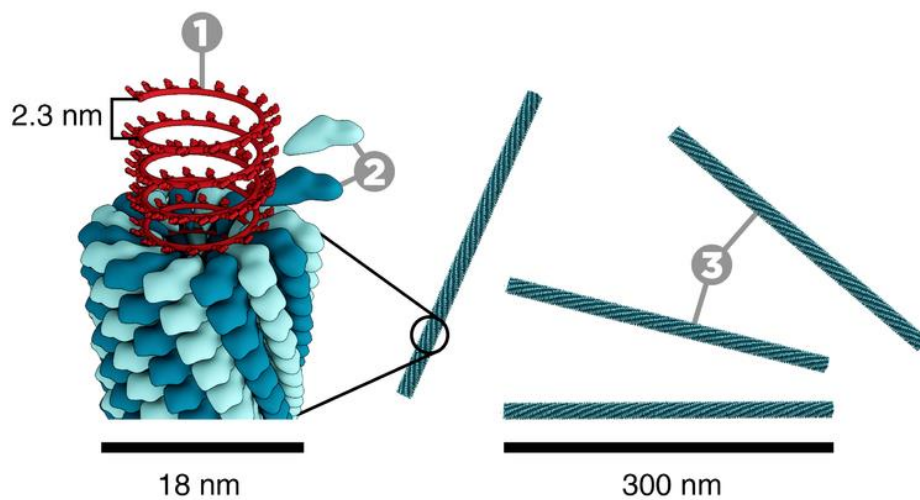

Figure 5: Tobacco mosaic virus (TMV), 1 represents RNA, 2 is a capsomer and 3 is a capsid. [7]

They supposed that this process could be understood on the basis of equilibrium thermodynamics. In 1967 they showed that viruses with icosahedral symmetry can also be self-assembled *in vitro*.[4] Later, the thermodynamic phase diagram of solutions of TMV components in terms of acidity and salinity was determined. There is a combination of different interactions interacting between capsid proteins: electrostatic repulsion, hydrophobic attraction and specific contacts between certain pairs of amino acids (Caspar pairs). If the conditions (acidity, salinity and temperature) are right, self-assembly can start. Firstly, protein clusters are held together by hydrophobic attractive interactions and double-discs or double-rings are formed. They are positively charged and so electrostatic repulsion prevents their aggregation. When oppositely charged ssRNA genome molecules are added, protein discs combine into rod-like cylinders with RNA running along the central axis. Most of the RNA viruses need the presence of the genome molecules to bring self-assembly to the conclusion, at least under ambient conditions. However, this is different for DNA viruses, where the presence of genome molecules is not required. Their genome is usually inserted by a rotary molecular motor.

At the end of self-assembly there is also a process called maturation. This means that the capsid is strengthen by chemical reactions because viruses have to stand large internal pressure during their life.

Self-assembly is possible only if the assembled virus has lower free energy than disassembled structure. This means that viruses are thermodynamically optimal structures.

If we have a shell composed of N subunits, the concentration of assembled capsids is proportional to c^N. c represents the concentration of free subunits and c_T is the total protein concentration. The fraction of proteins $f(c_T)$ incorporated into capsids is

$$f(c_T) \sim 1 - \frac{c_{crit}}{c_T} \tag{2}$$

for $c_T > c_{crit}$. c_{crit} is a minimum concentration of the ingredients required for assembly and it is defined

$$c_{crit} \propto \exp(\frac{\beta \Delta G_0}{N}) \tag{3}$$

and it is determined by the standard Gibbs free energy (ΔG_0) of the assembly reaction. This describes well the self-assembly of Cowpea chlorotic mottle virus (CCMV) and Hepatitis B virus (HBV) with a c_{crit} in the μM range. [4]

However, there are viruses which cannot self-assemble (for example bacteriophage viruses). They can have all components available, but self-assembly occurs only inside the host cell, because ATP energy is needed for this process.

4   A simple model of a capsid and its energy

As we find out in previous section the protein charge is very important in viral self-assembly, thus we have to take a look at its energy.

The simplest distribution of the protein charge would be homogenously distributed on a protein. This means that proteins would not want to be near each other because of the electrostatic repulsion. However, we know that proteins assemble in a capsid so there has to be an attractive interaction between them. It turns out that this is a combination of hydrophobic and van der Waals interactions.

We take an uniformly positively charged (with surface charge density σ) permeable infinitely thin sphere of radius R and this is one really simple model of a capsid.[5] This can be solved by treating the ions as ideal gas which adjust to the external potential and contribute to it via their charge density. [5] This is Poisson-Boltzmann approach where we get a nonlinear differential equation for the electrostatic potential ϕ. It can be derived by minimizing the appropriate free energy which has the form [5]

$$F_{PB}[\phi(r), \nabla\phi(r), c^i(r)] = \int f_{PB}\left(\phi(r), \nabla\phi(r), c^i(r)\right) d^3r \tag{4}$$

and the free energy density f_{PB} is

$$f_{PB}\left(\phi(r), \nabla\phi(r), c^i(r)\right) \tag{5}$$
$$= \frac{1}{2}\varepsilon_0\varepsilon\nabla\phi(r)^2 + \sum_{i=\pm} e_i\, c^i(r)\phi(r)$$
$$+ \sum_{i=\pm} k_B T\left[c^i(r)\ln c^i(r) - c^i(r) - \left(c_0^i \ln c_0^i - c_0^i\right)\right]$$
$$+ e_0\rho_p(r)\phi(r)$$

$e_0\rho_p(r)$ is the charge density of the capsid, T is the temperature, k_B is the Boltzmann constant, c^i are the concentrations of salt ions, c_0^i their bulk concentrations, $\varepsilon_0\varepsilon$ is the permittivity of water and e_0 is the electron charge. When we minimize the equation (5), we get a new form of Poisson-Boltzmann equation

$$-\varepsilon\varepsilon_0\nabla^2\phi(r) = \sum_{i=\pm} e_i c_0^i e^{-\beta e_i \phi(r)} + e_0\rho_p(r) \tag{6}$$

with $\beta = k_B T$ and $e_i = \pm e_0$. If the electrostatic potentials in the solution are small ($e_0\beta\phi \ll 1$) and we have a symmetric electrolyte ($c_0^+ = c_0^- = c_0$), equation (6) becomes

$$-\nabla^2\phi(r) = \frac{\beta}{\varepsilon\varepsilon_0}\left(\sum_{i\pm} e_i^2 c_0^i\right)\phi(r) + \frac{e_0\rho_p(r)}{\varepsilon\varepsilon_0} + \cdots \tag{7}$$

And this is called Debye-Hückel equation. We introduce Debye-Hückel length κ^{-1}, κ is defined:

$$\kappa^2 = \frac{\beta}{\varepsilon\varepsilon_0}\left(\sum_{i\pm} e_i^2 c_0^i\right). \tag{8}$$

Our viruses are typically of the order 20 nm and $c_0 \approx 150\ mM$. This gives us $\kappa^{-1} \approx 1\ nm$, so the limit of $\kappa R \gg 1$ is often implied. This helps us a lot to solve the equation (7) and we get the expression for free energy [5]:

$$F_{DH} = \frac{\pi\sigma^2 R^2}{\varepsilon\varepsilon_0\kappa}. \tag{9}$$

To estimate free energy we need some information. It is estimated that the surface charge density is typically $\sigma \approx 1\ e_0\ /nm^2$, $R \approx 20\ nm$ and $c_0 \approx 100\ mM$. This gives us the result $F_{DH} \approx 10^4 k_B T$.

We can estimated the hydrophobic energy from the area of proteins engaged in protein-protein contacts. The capsid with $R \approx 20\ nm$ typically has triangulation number $T = 3$, this means that it consists of 180 protein subunits. The length of protein contacts in a capsid is approximately $3000\ nm$ and the thickness of capsid is about $2\ nm$. This gives us the total area of about $6000\ nm^2$. The energy of attractive protein interactions for such cases is of the order of $10\ \frac{mJ}{m^2}$ and if we multiply this with the total area of protein contacts, we get $F_{HP} \approx 10^4 k_B T$.

We can see that the electrostatic free energy and the hydrophobic are of the same order of magnitude so the assembly and disassembly are in a balance. Viruses are able to self-assemble, but they can disassemble too; when they want to put their genome into host cell.

It turns out that our simple model is quite good. When the model with the finite thickness of a capsid is introduced, it turns out that the free energy is of the same order of magnitude as in the previous case (with infinite thickness).

5 Analysis of thin elastic cylindrical tube

We are often interested in elastic deformations of a system, but most of the studies are made in the linear regime where we have small deformations. However, if we have bigger deformations, nonlinear regime should be considered too.

It is interesting to take a look at experiment in which can be studied both small and large deformations. This experiment is meant to model large deformations in materials made of thin sheets such as carbon nanotubes, fullerenes, cellular membranes, virus capsids etc. However, the experiment is made on macroscopic level, but it has been shown that is useful and accurate for nanolevel as well.
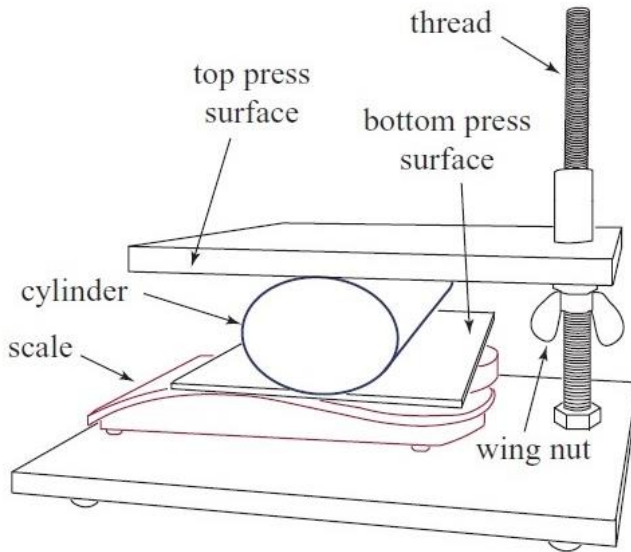

Figure 6: Illustration of the experiment [8]

As we can see in the picture, there is a tube pressed between two parallel plates. The problem is one dimensional, because the length of the parallel plates is larger than the length of the tube. The elastic energy of the pressed tube is

$$E_{el} = \frac{\hbar}{2} \int_S K^2 dS,$$

(10)

where $\hbar$ is the bending rigidity of the sheet and K is defined

$$K = R_1^{-1} + R_2^{-1}$$

(11)

with R_1 and R_2 being principal radii of curvature at some point. We assume that both radii are much bigger than the thickness of the sheet d, so we can use next equation for $\hbar$:

$$k = \frac{Ed^3}{12(1 - v^2)} \tag{12}$$

where E is Young modulus and v is Poisson ratio.

6 The compression of virus-like particles carrying surface dipoles

The findings from section 5 can now be used for our further research about viruses.

We are interested in what happens when viruses are compressed under certain conditions. We take N viruses and put them between two rigid plates. In this case a virus is treated as a perfect sphere carrying a certain dipole moment distribution across the capsid thickness.
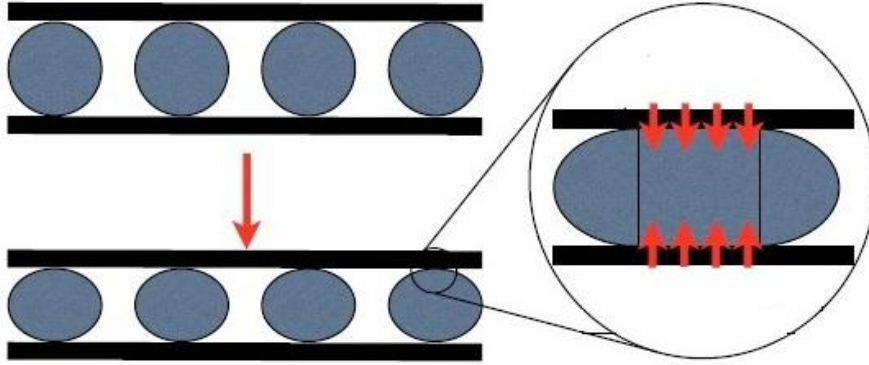

Figure 7: Illustration of a problem

R is the capsid radius and h is the thickness of capsid (h is from $0.1R$ to $0.2R$). As we see in the picture 7, at first each sphere has two contact points with plates and the distance between them is $2R$. Later when plates are compressed, the distance becomes $2(R - d)$. We also have a free part of the capsid which is not in contact with plates. This part can be described with the curve $r = r(z)$ in cylindrical coordinates. The curve has a flat part: a disc of radius b and this quantity b is the most important for us, because we would like to know the relation $b = b(d)$ (how the size of the flat part changes with the compression of the plates) and energy of the compressed capsid.

We use the equation (10) from previous section for energy and another form of equation (11)

$$K(z) = \frac{r(z)r''(z) - [1 + r'^2(z)]}{r(z)[1 + r'^2(z)]^{3/2}}. \tag{13}$$

This expression is then inserted in equation (E_{el}) for elastic energy. For obtaining total energy functional we need to add the contribution of the deformation of the shell caused by pressing the plates which, at the end, results in a complicated expression to be solved.

The radial dipole moment across the capsid thickness (in the figure 7 is marked red) introduces jumps in the electric field at both plates and exactly this jumps and their size are our main interest.

7 Conclusion

Even though viruses are nano objects, there are a lot of things we can examine about them; some of them are presented in this seminar. If we knew all their properties, our lives would be much simpler, especially the treatment of diseases they cause. However, there is still a lot of work to do because we do not understand many things, although we are daily surrounded by various viruses. They are small so they can go almost everywhere and reach every cell. We still do not know how to cope with many of them, therefore only further studies would help.

There is a quote which shows us how special creatures they are: "An inefficient virus kills its host. A clever virus stays with it." (James Lovelock) Not only that, they are able to self-assemble, to survive in really unfriendly environment and to cause us many problems; we should respect them because of their qualities.

8 References

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