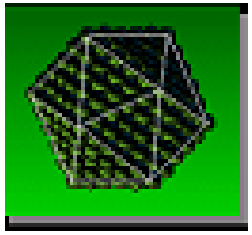

Patogeneze virových nákaz

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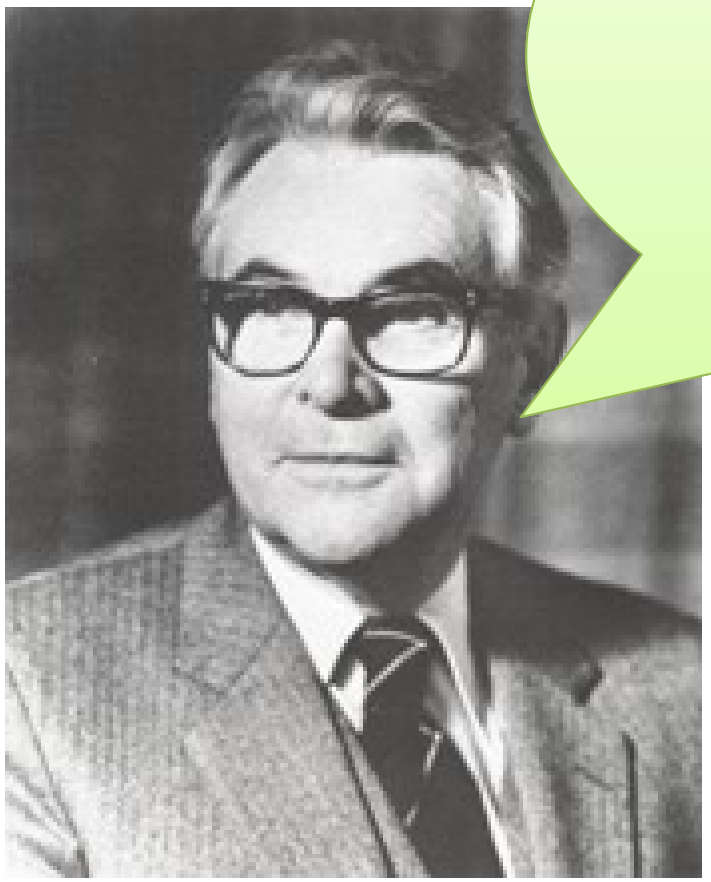
Financováno Evropskou unií NextGenerationEU

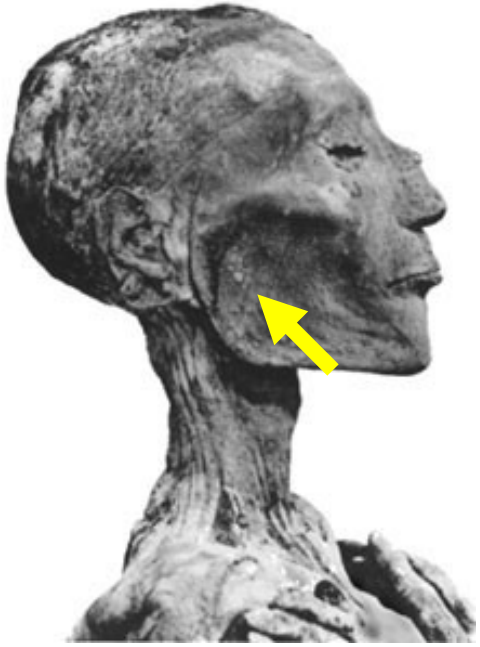
NÁRODNÍ PLÁN OBNOVY

MINISTERSTVO ŠKOLSTVÍ, MLÁDEŽE A TĚLOVÝCHOVY

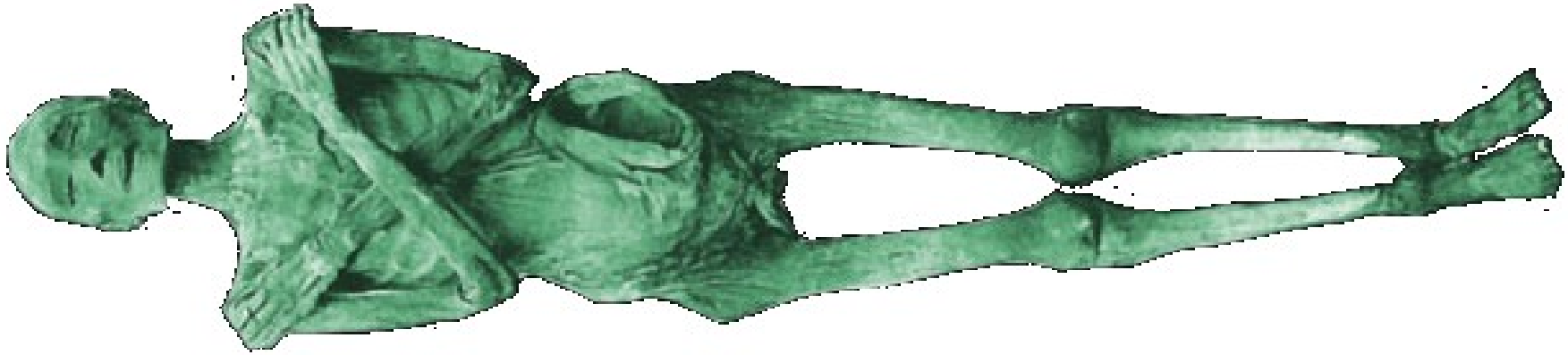


„Infekční nemoci předcházejí vzniku lidstva; budou trvat jako lidstvo samo, a bezpochyby zůstanou i nadále jedním z rozhodujících činitelů lidských dějin, tak jako jím jsou dosud.“

(William McNeill)

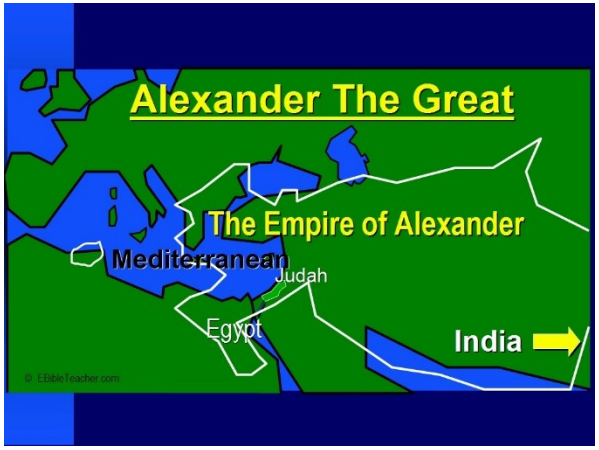


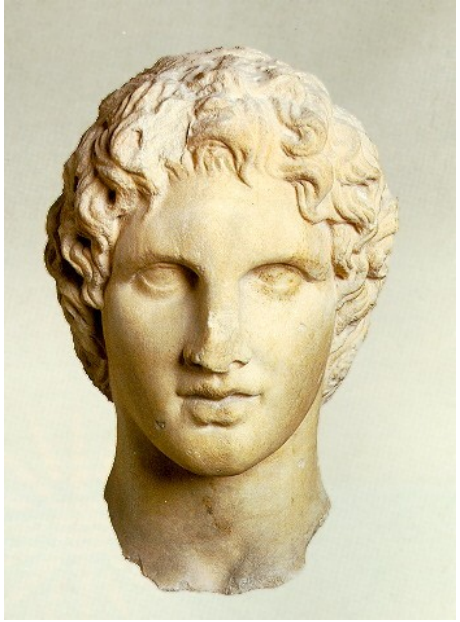
Faraón Ramses V.
– zemřel na pravé neštovice roku 1156 př.n.l.



Egyptská deska z 18. dynastie ukazující osobu postiženou dětskou obrnou

Mummy and clubfoot of 19th Dynasty Pharaoh Siphtah (c. 1300 B.C.)





Alexandr Veliký zemřel na infekci virem západního Nilu v Babylóně roku 323 př.n.l.

Table. Medical history and physical examination of Alexander the Great

Patient characteristics	Medical history	Clinical symptoms
Male	Ten years before death, traveled widely (Mediterranean, North Africa, and Middle East)	Escalating fever associated with chills
Born in Macedonia	Unexplained fever 5 years previously	Excessive thirst, diaphoresis
32 years of age	Penetrating right chest wound one year before final illness	Acute abdominal pain
Soldier	Onset of final illness May 29, 323 BC	Single episode of back pain at onset of fever
Heavy drinking	Death June 10, 323 BC	Increased weakness leading to prostration with intermittent periods of energy
Frequent bathing		Delirium
Married to many wives		Aphonia
One son		Terminal flaccid paralysis

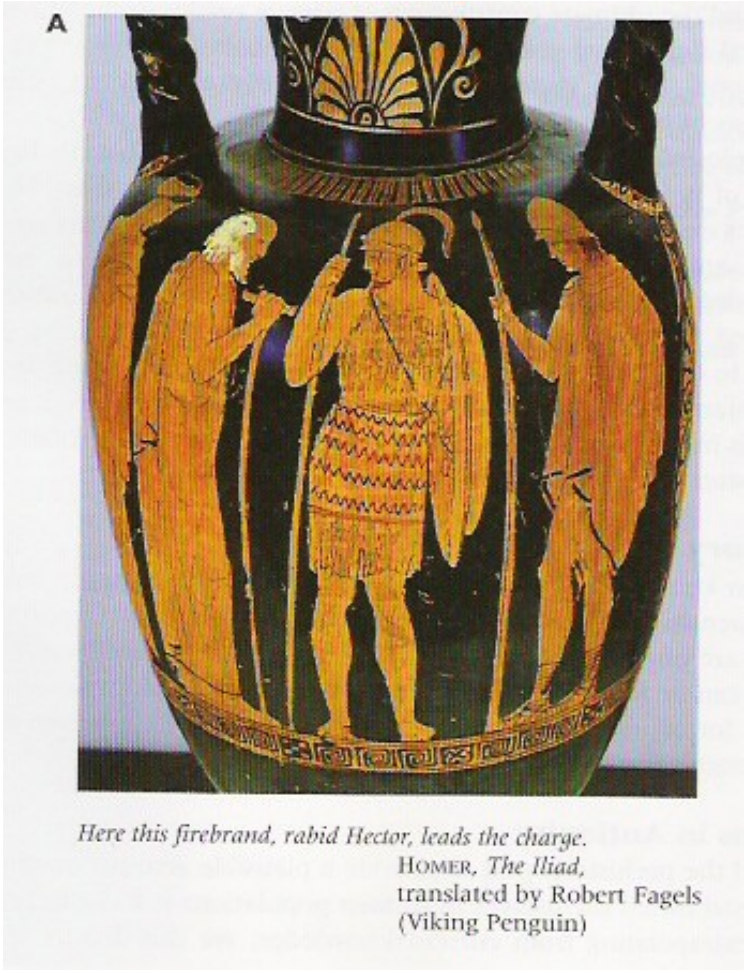
Vzteklina – nejstarší dokumentované virové onemocnění lidstva

Vteklina - Rabies (odvozená ze sanskrtu, “rabhas” znamená “dělat násilnosti, zákeřnosti, prudkost, hrubost...”)

- poprvé popsáno v Babylóně
- lékaři Mezopotámie, Číny, Řecka, Říma a Indie popsali klinické znaky a symptomy nákazy virem vztekliny
- v prvním století našeho letopočtu Celsus a Galen popsali způsob léčby, včetně excize místa pokousání a vypalování místa pokousání horkým železem

Mezopotámské zákony (1000 let př.n.l.) definují vzteklinu jako nakažlivou chorobu a určují povinnosti a odpovědnost majitelů psů postižených vzteklinou.

Starověké Řecko a Řím



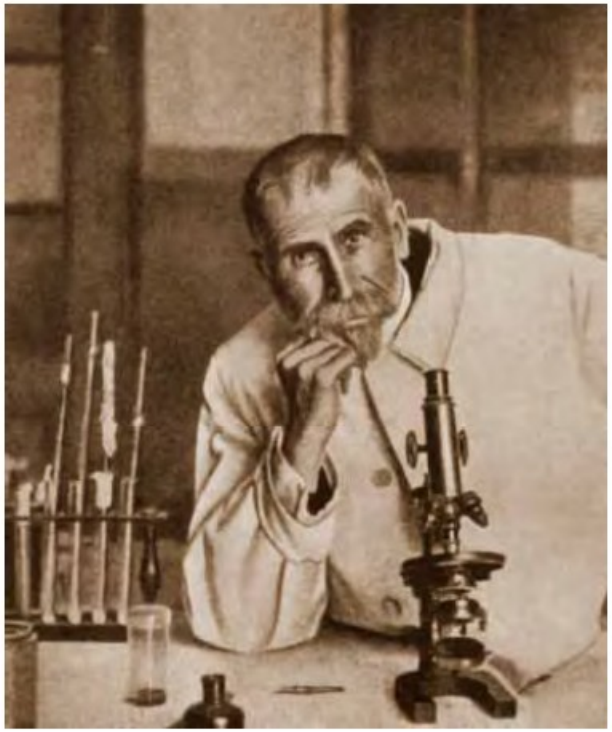
Homérova Iliada: vystupuje zde buřič Hector, který je postižen vzteklinou

Vzteklý pes napadající tele
Reliéf ze zbytků římského opevnění Neumagen-Dhron poblíž Trieru, Německo, 4. století n.l.

Edward Jenner vaccinating his son, who is held by his wife, while the maid prepares for her vaccination
A hand-colored engraving by Jean Claude Manigaud (Paris) after a painting by Edward Hamman

Jubilé de Pasteur a la Sorbonne – 27 December 1892 [Painting by Jean André Rixens (1846-1924)] Pasteur's seventieth birthday was the occasion of a national holiday. The celebration was held in the great hall of the Sorbonne. Pasteur entered on the arm of the President of the Republic, Sadi Carnot. Both were wearing the grand cordon of the Légion d'Honneur. Pasteur was greeted by an immense ovation; he was too weak to speak to the delegates who had gathered from all over the world, so his address was read by his son: *"Gentlemen, you bring me the greatest happiness that can be experienced by a man whose invincible belief is that science and peace will triumph over ignorance and war.... Have faith that in the long run ... the future will belong not to the conquerors but to the saviors of mankind."* The painting shows Joseph Lister coming forward to greet his old friend. He said: *"It is my great privilege to convey to you, tributes, thanks and respect from all involved in medicine and surgery; it is true to say that, of all people in the world today, medical sciences owe you the most. For centuries, infectious diseases have been shrouded, as it were under a dark curtain. In discovering the microbial origin of disease you have raised that dark curtain!"*

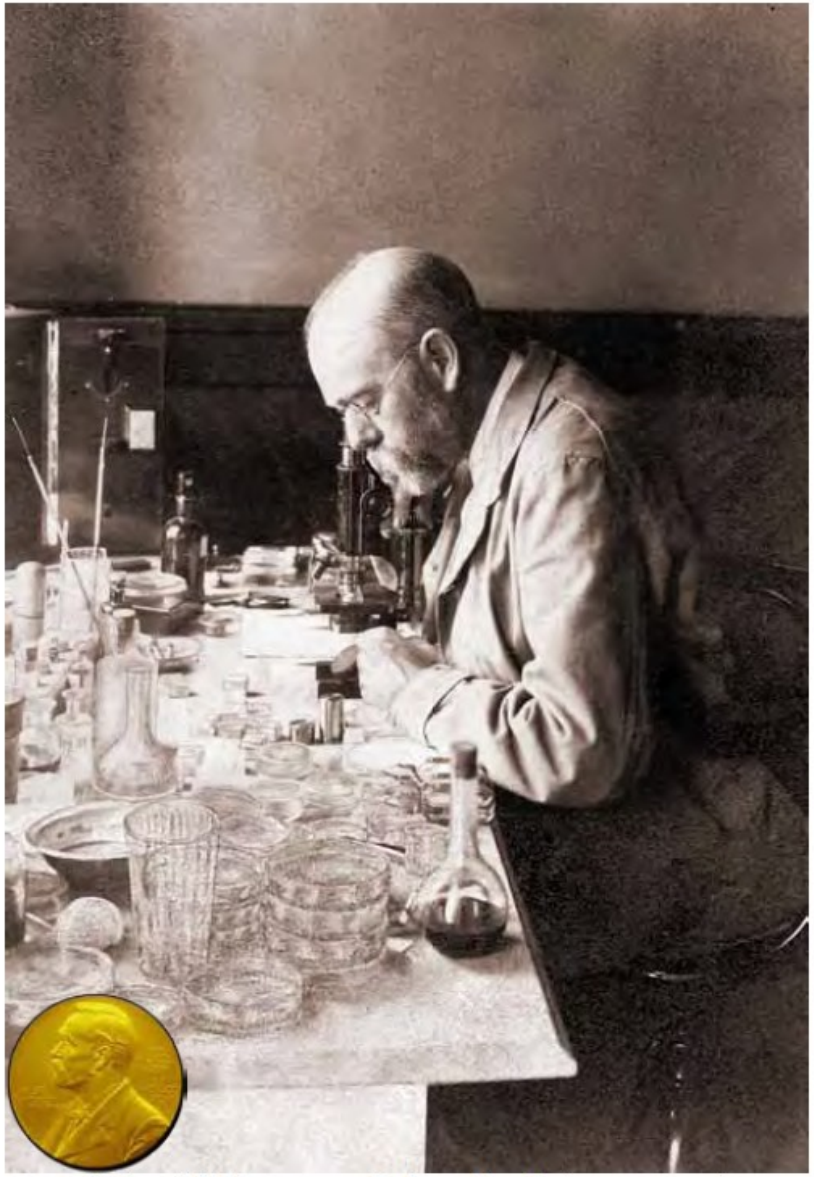
In 1881 and 1882, Louis Pasteur, Charles Chamberland, Emile Roux and Louis Thuillier began their research toward developing a rabies vaccine. They modified Pierre-Victor Galtier's technique by inoculating nervous tissue from a rabid dog directly into the brain of the next dog via trephination. By successive passages in dogs, they obtained a virus of maximum virulence and with a fixed incubation period of about 10 days. They estimated the degree of attenuation of brain tissue from each passage. They then passaged the infectious agent serially in rabbits. This final attenuation procedure consisted of suspending the spinal cord of a rabid rabbit in a flask, in a warm dry atmosphere, to achieve slow desiccation. They succeeded in producing "attenuated viruses of different strengths," that is a standardized range of viruses, the weakest of which could be used to prepare the first dose of a vaccine. Inoculating dogs with a sequence of spinal cords of increasing virulence rendered the recipient resistant to inoculation with fully virulent virus.



Pierre-Paul-Emile Roux (1853-1933)

Louis Pasteur (1822-1895)
Painting byAlbert Edelfeldt, 1885
[from Institut Pasteur, used with permission]

1884 • Louis Pasteur, Emile Roux, Charles Chamberland, Louis Thuillier • development of rabies vaccine



Heinrich Hermann Robert Koch (1843-1910)

Koch in Africa, ~1904, seeking the cause of rinderpest

Robert Koch is credited with the first incontrovertible proof that a microorganism can cause disease (*Bacillus anthracis*—anthrax). After Casimir Davaine showed the direct transmission of the anthrax bacillus between cows, Koch studied anthrax more closely. He invented methods to purify the bacillus from blood and grow pure cultures. He found that, while the anthrax bacillus could not survive outside a host for long, it produces endospores that survive for many years. He developed bacterial staining techniques, bacterial liquid and solid growth media (and agar plates thanks to the advice of Angelina and Walther Hesse), and he developed the Petri dish, named after its inventor, his assistant Julius Richard Petri. With these techniques, he was able to discover the bacterium causing tuberculosis (*Mycobacterium tuberculosis)* in 1882.

With Friedrich Gustav Jakob Henle and Friedrich Loeffler, in 1884, he developed the Henle-Loeffler-Koch Postulates (Koch's Postulates) outlining the criteria required to establish a causal relationship between a microbe and a disease:
1. The microorganism must be found in abundance in all organisms suffering from the disease, but should not be found in healthy animals. 2. The microorganism must be isolated from a diseased organism and grown in pure culture. 3. The cultured microorganism should cause disease when introduced into a healthy organism. 4. The microorganism must be reisolated from the inoculated, diseased experimental host and identified as being identical to the original specific causative agent.

1876 • Robert Koch • along with Louis Pasteur, the founder of microbiology

Adolf Eduard Mayer (1843-1942)

Tobacco mosaic disease, Mayer's work, 1866

Adolf Mayer was a German agricultural chemist and director of the Agricultural Experiment Station at Wageningen in the Netherlands. He is credited as the first person to transmit tobacco mosaic virus by using juice extracts from diseased plants as the inoculum to infect other plants. Mayer published a paper in 1886 describing tobacco mosaic disease in detail. He performed chemical analyses of healthy and diseased leaves to see if a difference in nutrition could explain the disease. He investigated temperature, light, fertilization, and looked for fungi or "animal parasites." He tried to follow Koch's Postulates and was able to culture organisms from his extracts, but none of these would reproduce the disease. He was left with the conclusion that the infectious agent was likely some sort of microorganism. Although Mayer came to the wrong conclusion about his finding, his work laid the groundwork for the experiments of Dmitry Ivanovsky and Martinus Beijerinck, who showed that the etiologic agent of tobacco mosaic disease would pass through an ultrafilter, that is, it is an ultrafilterable virus.

Mayer, A. Concerning the mosaic disease of tobacco. Die Landwirtschaftliche Versuchsstationen 1886;32:451-467. Translation published in English as Phytopathological Classics Number 7 (1942). American Phytopathological Society Press. St. Paul, MN.

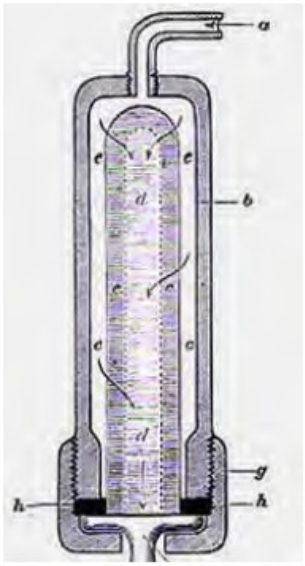
1886 • Adolf Mayer • the concept of transmissibility and the earliest concept of an ultrafilterable virus

100 let virologie jako vědecké disciplíny

Dmitrij Ivanovský, 1892

Fig. 2. Symptoms of tobacco mosaic virus. (Photograph courtesy of the IPO, Wageningen, The Netherlands.)

Charles Chamberland (1851-1908)

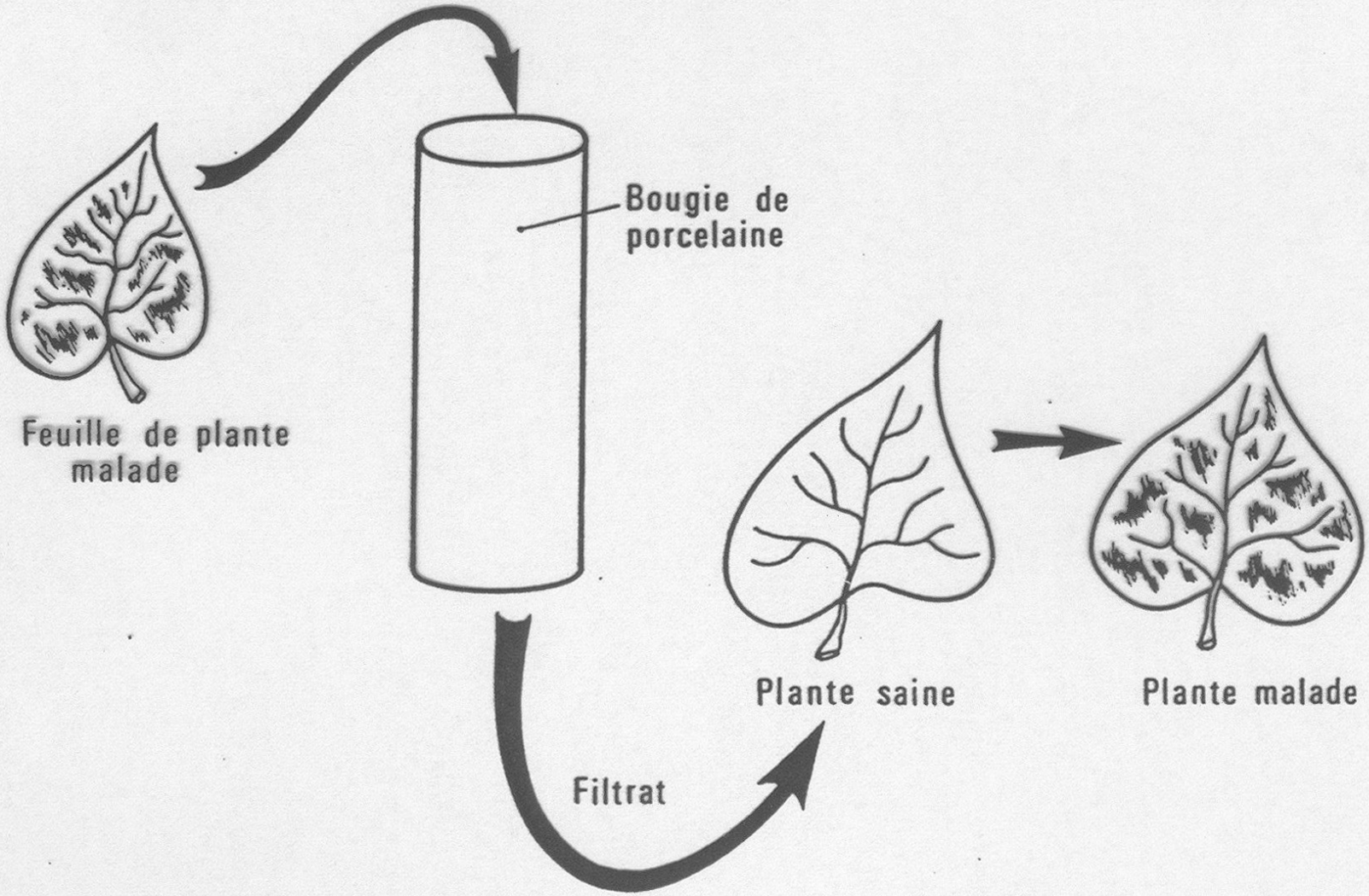


Theodor Albrecht Edwin Klebs (1834-1913) and Ernst Tiegel, in 1871, found that the causative agent of anthrax would not pass through a filter made of unfired clay – it was a non-filterable bacterium. As early as 1876, Louis Pasteur, in collaboration with the French physicist Jules Joubert, used a plaster-of-Paris filter connected to a vacuum pump to confirm Krebs' diacovery. The use of ultrafilters to retain and concentrate bacteria became standard practice after 1884, when Charles Chamberland and Emile Roux, experimenting with a broken clay pipe purchased from Chamberland's tobacconist, developed unglazed porcelain ultrafilters (candles, "bougies de Chamberland") that retained bacteria. Chamberland filters were produced in 13 types: L_1 to L_{13}; L_1 filters having the coarsest pore size, L_{13} the finest.

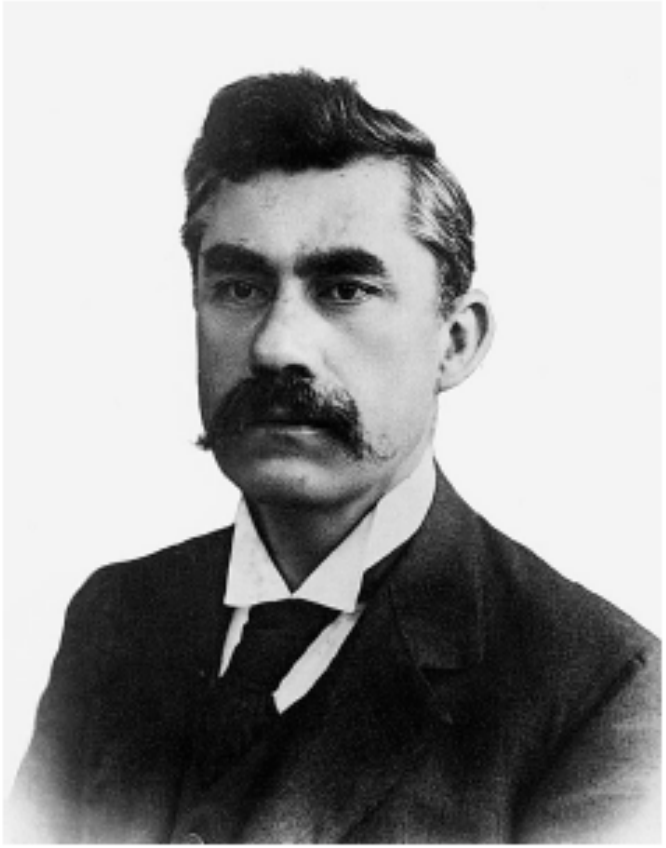
Chamberland C. Sur un filtre donnant de l'eau physiologiquement pure. C. R. Acad. Sci. Paris 1884;99:247-248.

page 51

1884 • Charles Chamberland • development of the porcelain ultrafilter, key to the discovery of the viruses



L'expérience d'Ivanowski.



Martinus W. Beijerinck

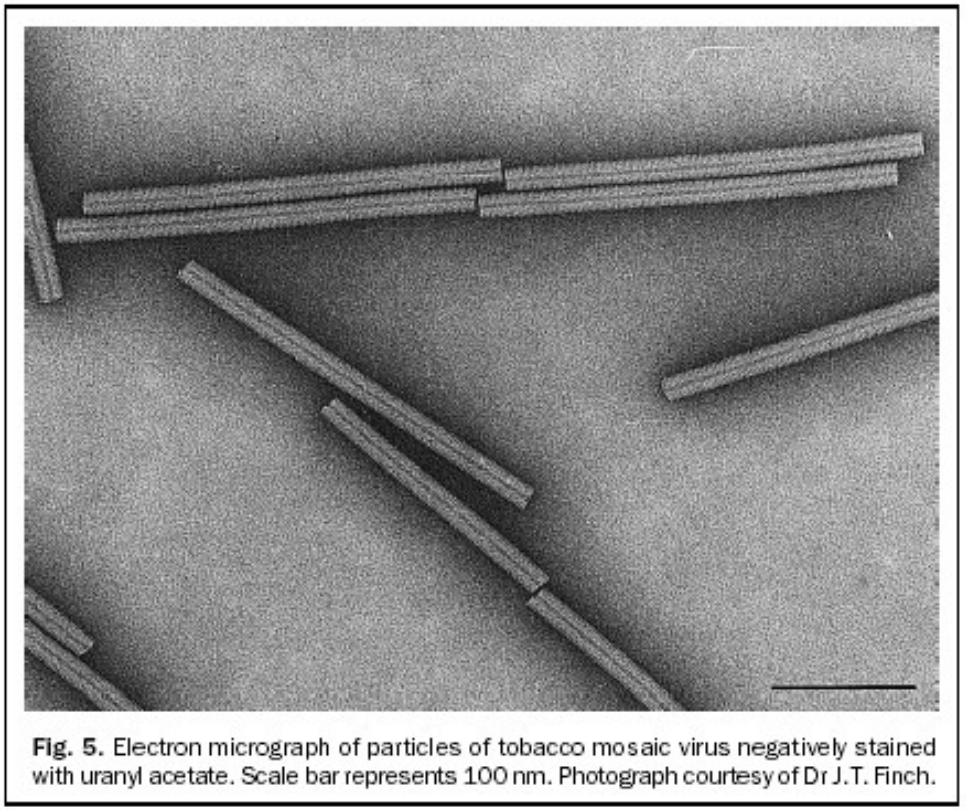
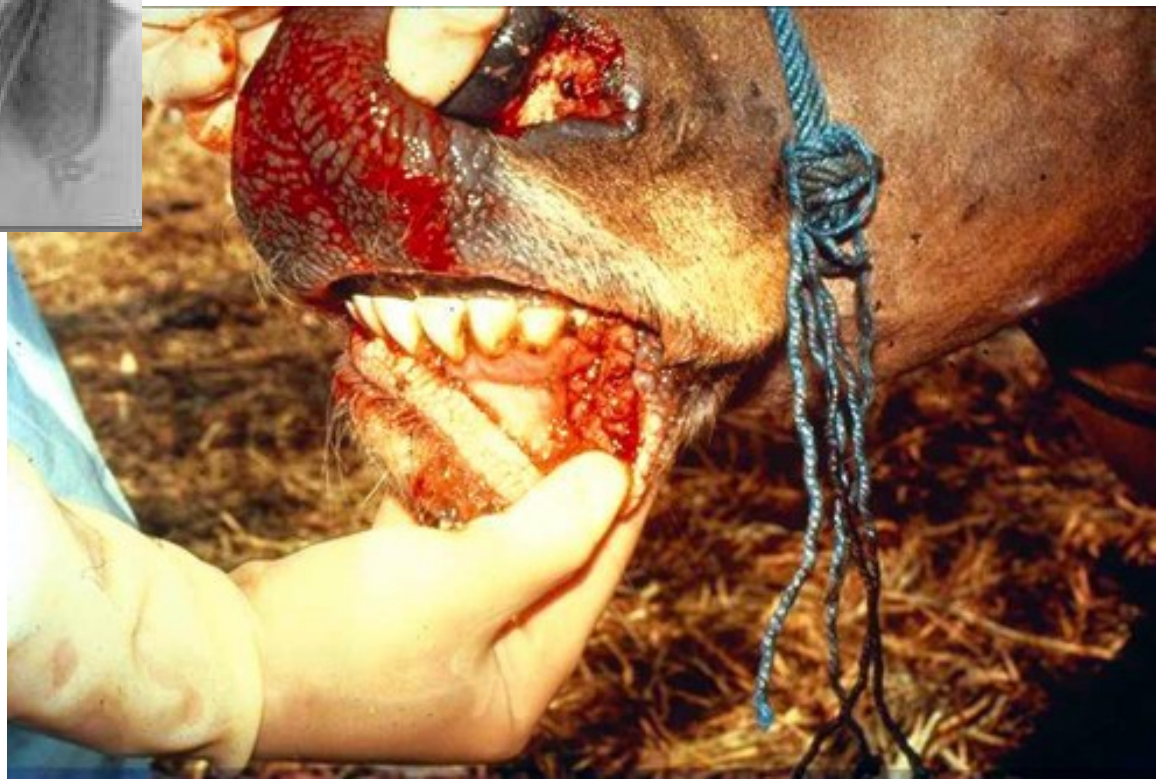


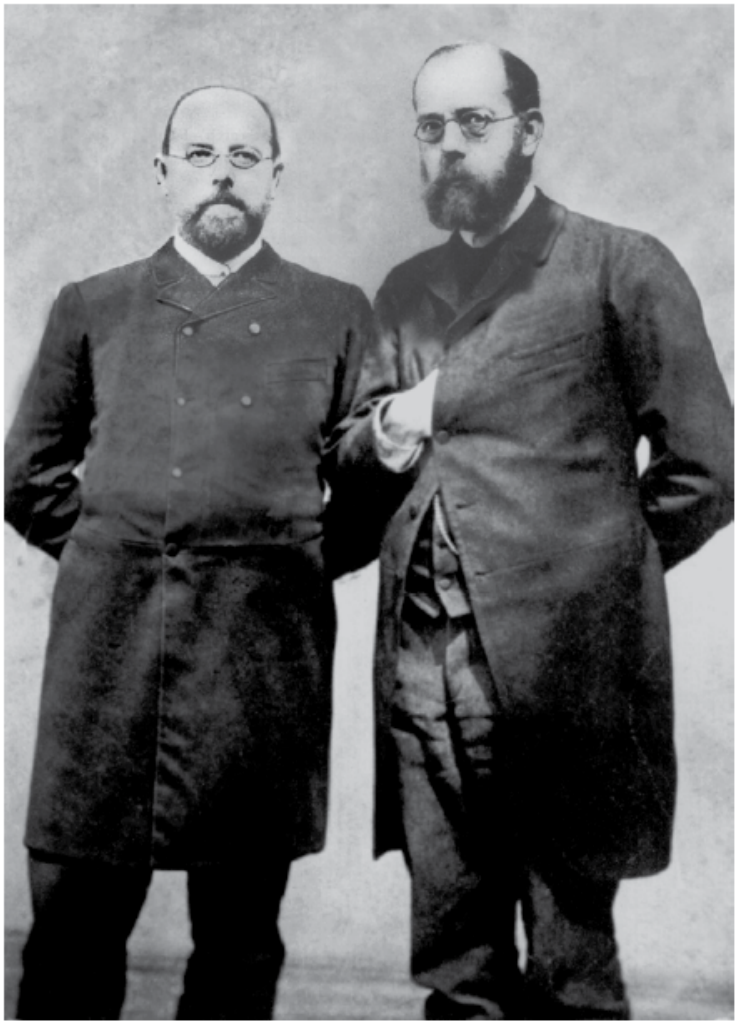
Fig. 5. Electron micrograph of particles of tobacco mosaic virus negatively stained with uranyl acetate. Scale bar represents 100 nm. Photograph courtesy of Dr J.T. Finch.

Contagium vivum fluidum

Fridrich Loeffler a Paul Frosh

Slintavka a kulhavka, 1898





Friedrich Loeffler (1852-1915) Robert Koch (1843-1910)

Who discovered the first virus?

There are many, many articles arguing the priority of the discovery of the first virus – this is a place for strongly held opinions…

Dmitry K. Lvov: Centenary of Virology, in Concepts in Virology: From Ivanovsky to the Present [Mahy BWJ (Ed.)]. Brian Taylor & Francis, Inc.; 1993. ***It was Ivanovsky…***

A. van Kammen: Beijerinck's Contribution to the Virus Concept. In 100 Years of Virology – The Birth and Growth of a Discipline. [Calisher CH, Horzinek MC (Eds.)]. Archives of Virology, Suppl. 15; 1999. ***It was Beijerinck…***

Marc H. V. van Regenmortel: The Nature and Classification of Viruses, in Topley & Wilson's Microbiology & Microbial Infections [Mahy BWJ, ter Meulen V (Eds.)]. Hodder Arnold; 2005. ***It was Loeffler and Frosch…***

An Opinion on the Priority of the Work of Ivanovsky, Beijerinck and Loeffler and Frosch by Marc van Regenmortel, 2006 – abridged:

…Although all historical accounts of the beginnings of virology refer to the work of Ivanovsky, Beijerinck, and Loeffler and Frosch (working with Koch), there is disagreement among authors about who should be credited with the discovery that viruses were a new type of infectious agent. This debate concerns the question of what is a scientific discovery.

Although Ivanovsky was clearly the first one to show that the agent causing tobacco mosaic disease passed through a bacteria-retaining filter, all his publications show that he did not grasp the significance of his observation. He believed that the filter he used might have had fine cracks and that small spores of a microbe might have passed through the filter.

Beijerinck on the other hand, realized he was dealing with something different from a microbe but he thought that the virus was an infectious liquid and not a particle.

Only Loeffler and Frosch correctly concluded that the virus causing foot-and-mouth disease was a small particle that passed through a Chamberland filter, but was stopped by a finer-grained Kitasato filter.

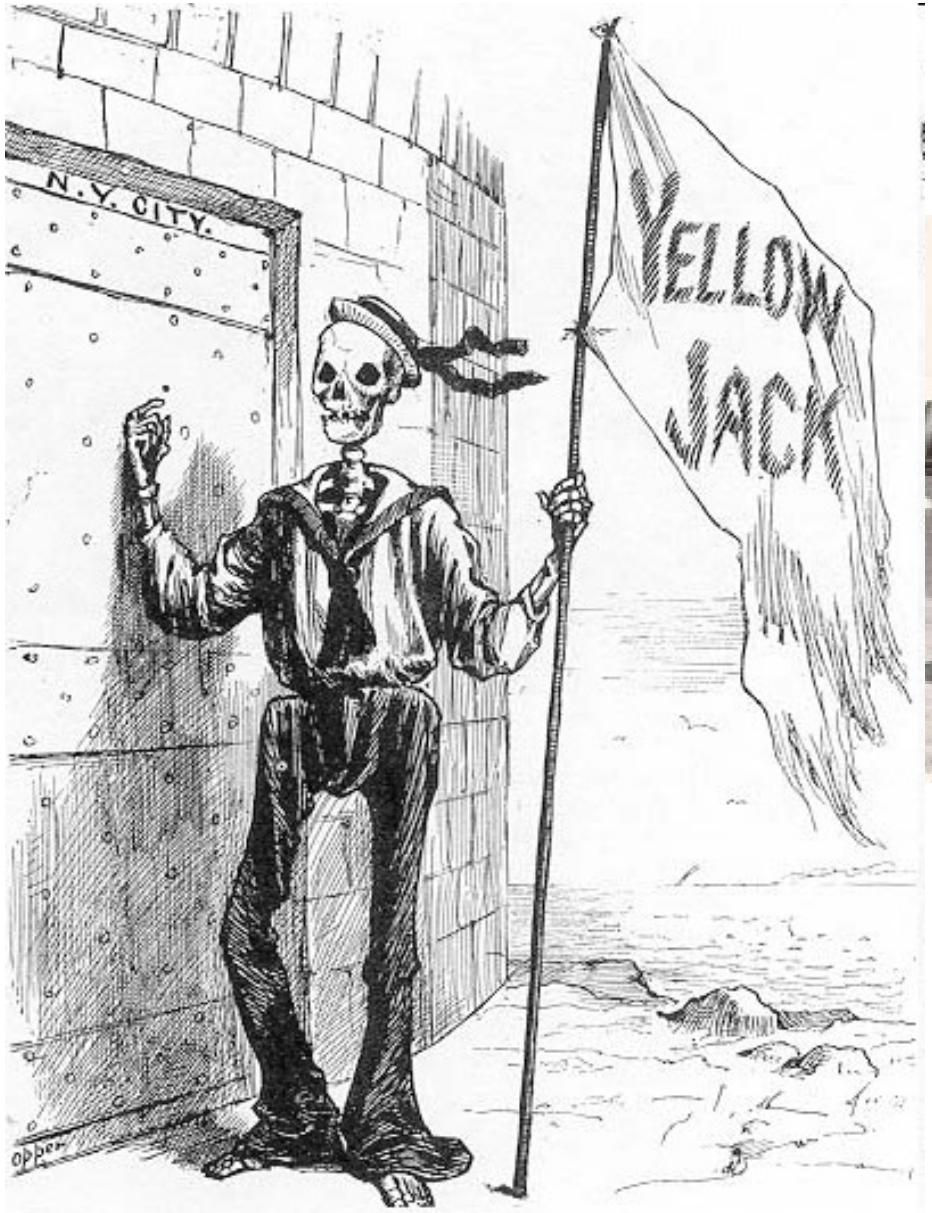
The debate about who should be considered the founder of virology may be settled only if it is accepted that, in order to make a discovery, it is not sufficient to make a novel observation (i.e., the filterability of an infectious agent), but that it is also necessary to interpret the observation correctly.

Good science does not consist only in making new observations but it requires also unbiased, imaginative thinking which enables the scientist to arrive at the correct interpretation of his experimental findings.

Loeffler and Frosch's interpretation of their filtration experiments came the closest to the modern concept of a virus and so they should be acknowledged as the founders of virology.

1898 • Friedrich Loeffler and Paul Frosch • discovery of foot-and-mouth disease virus (the first virus: see text)

Žlutá zimnice



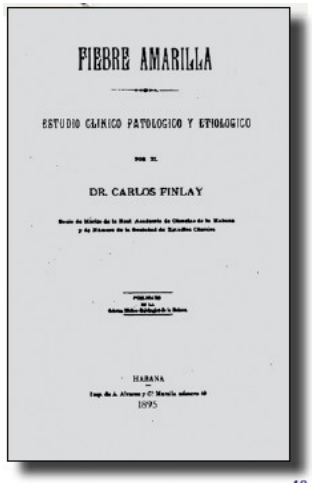
The Dead Wagon, 2nd Division Hospital, Havana, ca. 1898.

Co způsobuje žlutou zimnici a jakým způsobem je infekce přenášena

- domnělý původce *Bacillus icteroides*

THE BACILLUS ICTEROIDES AS THE CAUSE OF YELLOW FEVER

Science 15 September 1899: Vol. 10. no. 246, pp. 379 - 380

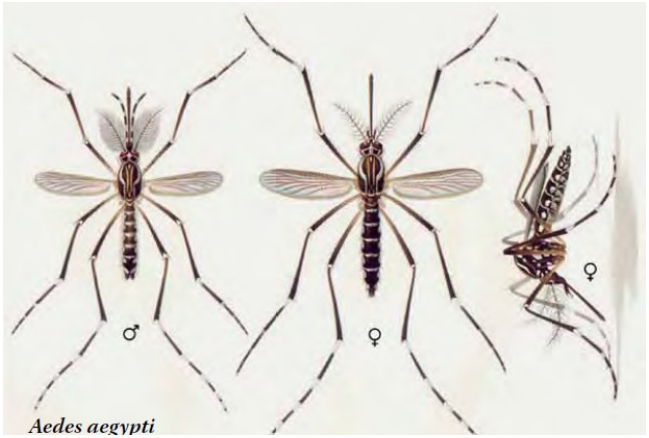

FIEBRE AMARILLA

ESTUDIO CLINICO PATOLOGICO Y ETIOLOGICO

DR. CARLOS FINLAY

HABANA

1895



Aedes aegypti

Dr. Carlos J. Finlay

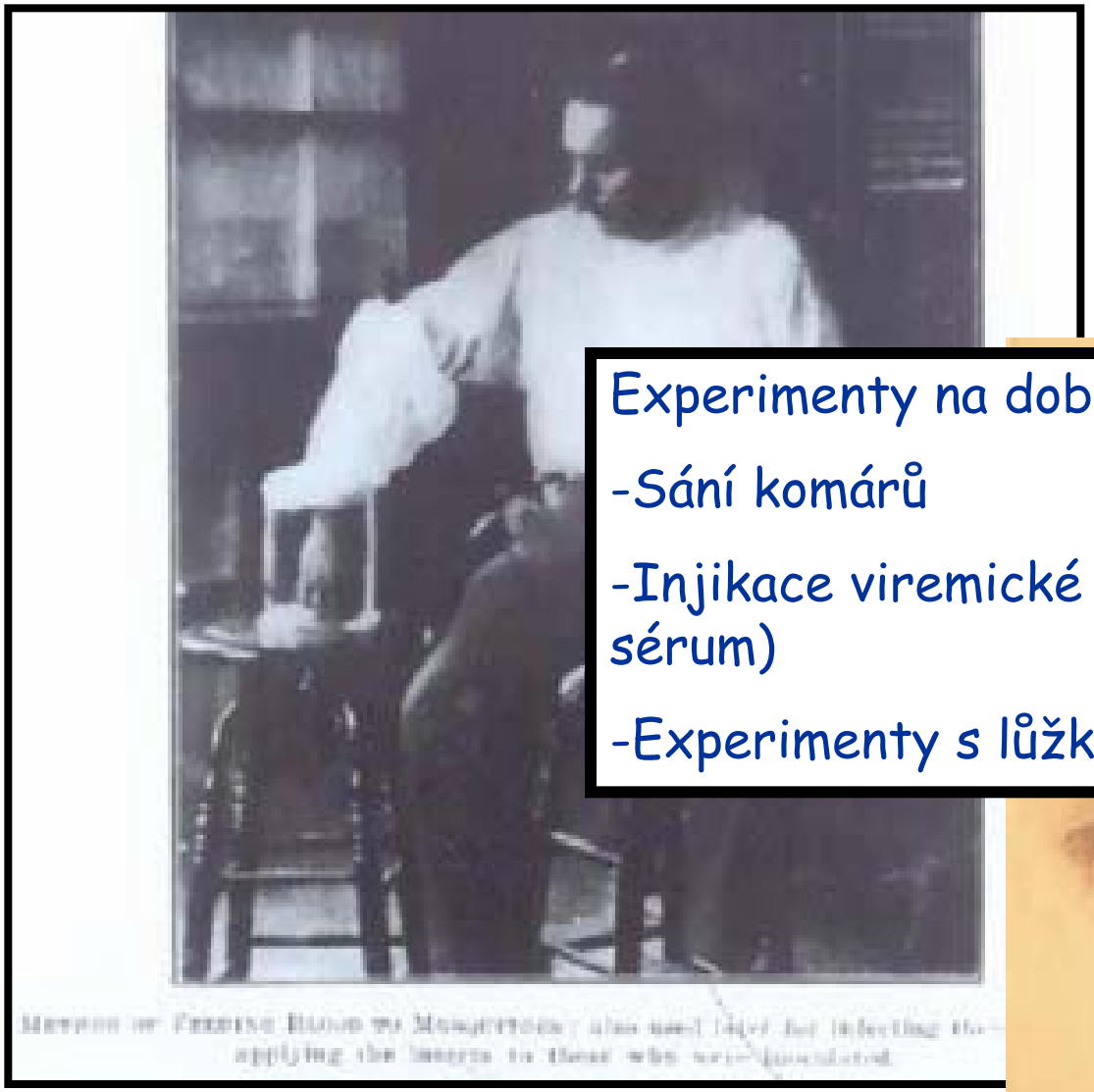
Havana in 1900

Reed on shipboard, en route to Cuba

TABLE II.

No. of case.	Day of disease.	Time of autopsy.	Source of culture.	B. icteroides.
1	Seventh	2 hours after death	Blood, liver, spleen, kidney	Negative.
2	Sixth	13 hours after death	do	Do.
3	Fourth	8 hours after death	do	Do.
4	Eighth	4 hours after death	Abdominal cavity, blood, liver, spleen, kidney, bile, duodenum.	Do.
5	Fourth	do	Blood, liver, spleen, kidney, bile, duodenum.	Do.
6	Sixth	6½ hours after death	Abdominal cavity, blood, pericardial fluid, lung, spleen, kidney, liver, bile, duodenum.	Do.
7	do	50 minutes after death	Blood, lung, liver, spleen, kidney, bile, jejunum.	Do.
8	do	½ hour after death	Blood, lung, liver, spleen, kidney, urine, small intestine.	Do.
9	Fourth	2 hours after death	Liver, spleen, small intestine	Do.
10	Fifth	7 hours after death	Liver, kidney, spleen, small intestine	Do.
11	Third	½ hour after death	Liver, kidney, spleen	Do.

[1] Cultures from the blood during life had been taken by Dr. Lazear in three other cases of yellow fever, but owing to the death of our colleague, the necessary data as to the day of the disease on which cultures had been taken can not be ascertained. These cultures were negative as regards the finding of Sanarelli's bacillus.



Mosquito feeding method

Experimenty na dobrovolnících

-Sání komárů

-Injikace viremické krve (+ filtrované sérum)

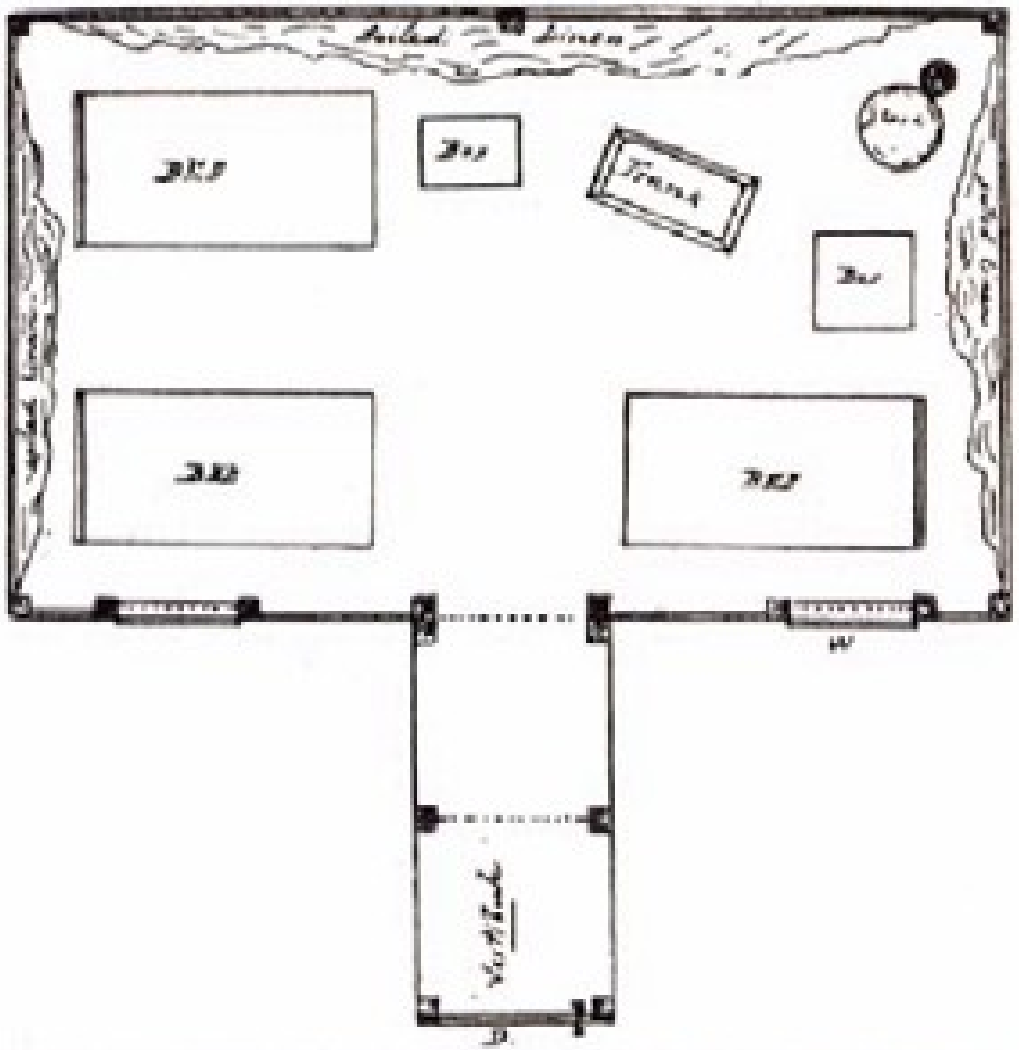
-Experimenty s lůžkovinami

Dr. Jesse Lazear

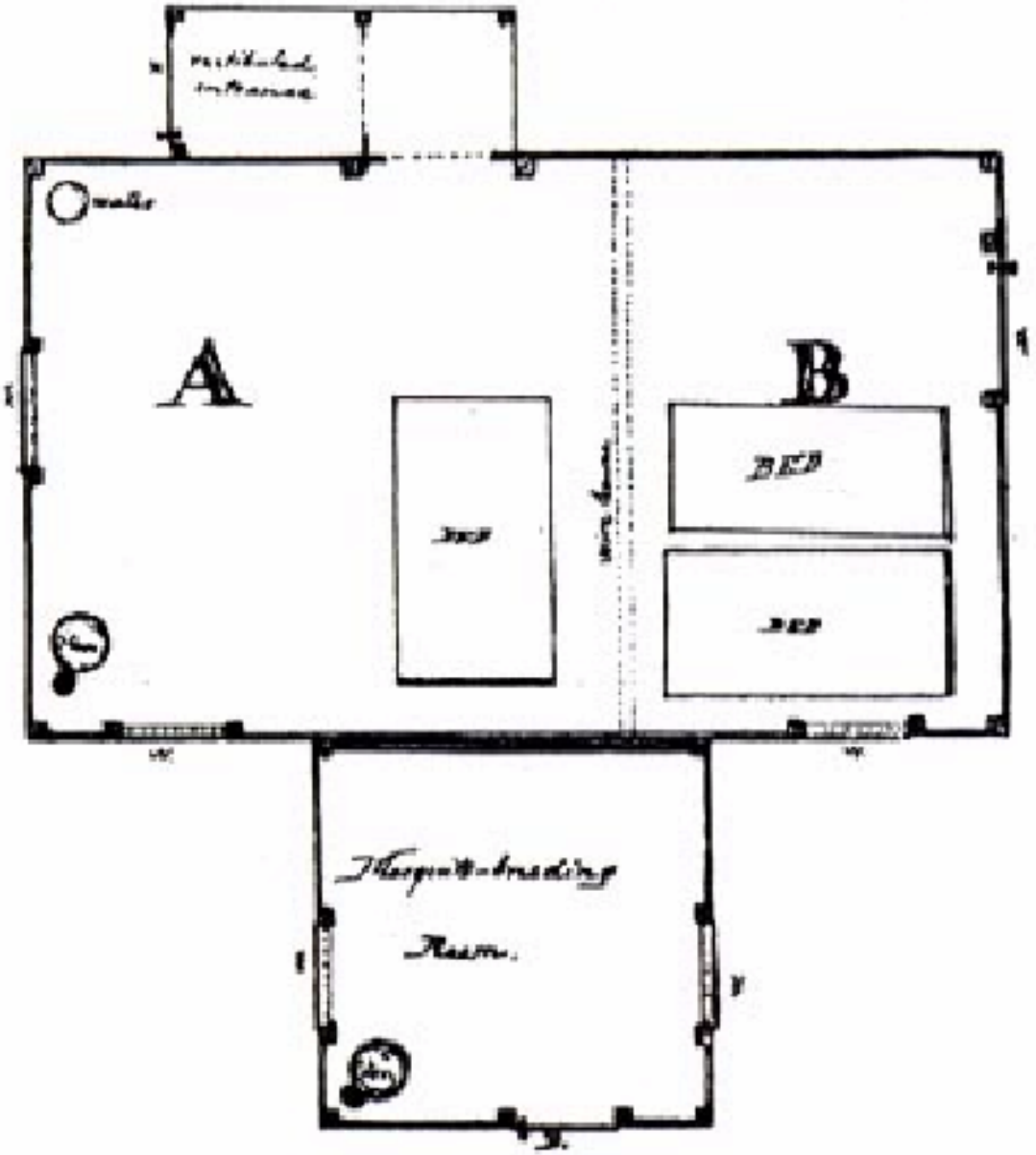
Building Number Two (Mosquitoes)

Building Number One (Fomites)

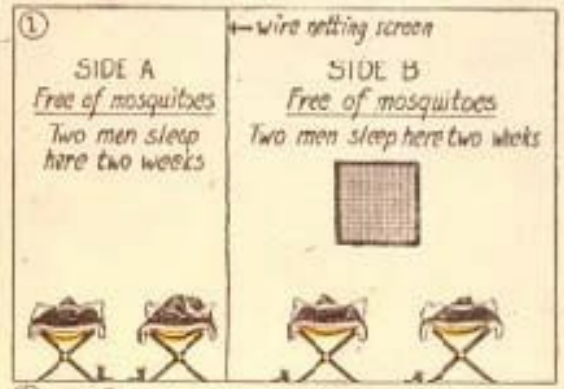
Camp Lazear



PLAN OF THE "INFECTED CLOTHING BUILDING" AT CAMP LAZEAR. Men who were susceptible to the disease slept many nights in this soiled linen room, without contracting yellow fever.

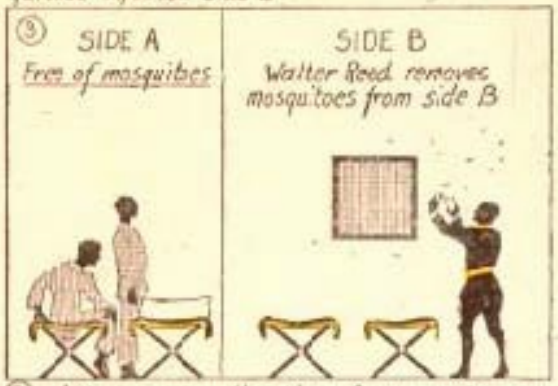


PLAN OF THE "MOSQUITO BUILDING" AT CAMP LAZEAR. The man who short time occupied the bed in room marked "A" became infected by the bites quitoes previously introduced, while those, equally susceptible, who occupied section marked "B" remained in good health. Only a wire-screen partition se the two compartments.



① All four men remain well. Therefore the building is not infected with yellow fever.

② J. Moran enters side B, is bitten and has yellow fever in four days. The men in side A remain well. Therefore the presence of contaminated mosquitoes infected side B.



③ Men sleep on both sides of wire netting as before without taking yellow fever. Therefore side B has been disinfected by removing mosquitoes

Compliments of the writers

[Reprinted from THE PHILADELPHIA MEDICAL JOURNAL, October 27, 1900.]

THE ETIOLOGY OF YELLOW FEVER.

A Preliminary Note.[1]

By WALTER REED, M.D., Surgeon, U. S. A.,

AND

JAMES CARROLL, M.D., A. AGRAMONTE, M.D., JESSE W. LAZEAR, M.D.,[2] Acting Assistant Surgeons, U. S. A.

Reed Carroll Agramonte Lazear

CONCLUSIONS.

1. The mosquito (*C. fasciatus*) serves as the intermediate host for the parasite of yellow fever.

2. Yellow fever is transmitted to the nonimmune individual by means of the bite of the mosquito that has previously fed on the blood of those sick with this disease.

3. An interval of about 12 days or more after contamination appears to be necessary before the mosquito is capable of conveying the infection.

4. The bite of the mosquito at an earlier period after contamination does not appear to confer any immunity against a subsequent attack.

5. Yellow fever can also be experimentally produced by the subcutaneous injection of blood taken from the general circulation during the first and second days of this disease.

6. An attack of yellow fever, produced by the bite of the mosquito, confers immunity against the subsequent injection of the blood of an individual suffering from the nonexperimental form of this disease.

7. The period of incubation in 13 cases of experimental yellow fever has varied from 41 hours to 5 days and 17 hours.

8. Yellow fever is not conveyed by fomites, and hence disinfection of articles of clothing, bedding, or merchandise, supposedly contaminated by contact with those sick with this disease, is unnecessary.

9. A house may be said to be infected with yellow fever only when there are present within its walls contaminated mosquitoes capable of conveying the parasite of this disease.

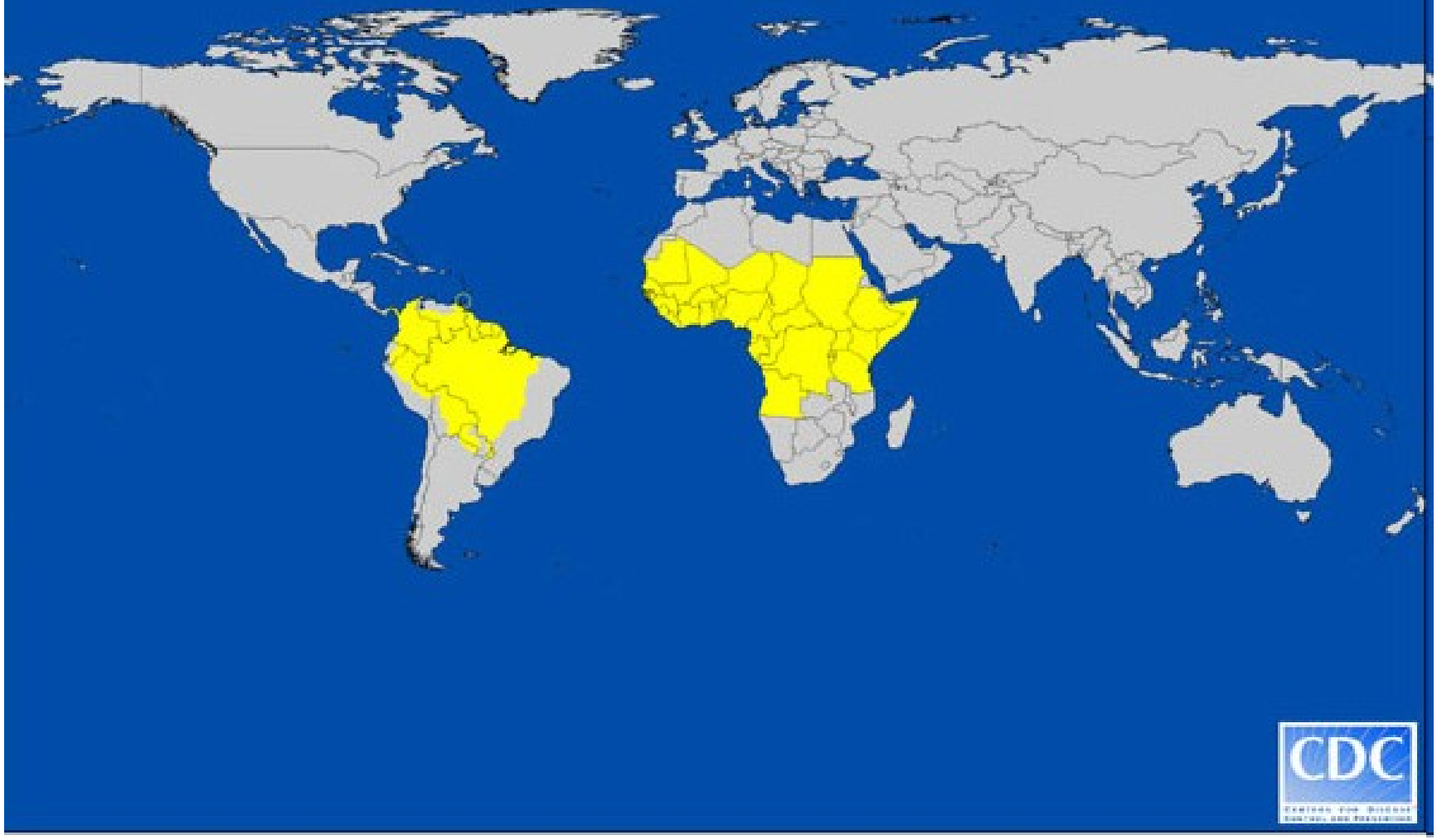
10. The spread of yellow fever can be most effectually controlled by measures directed to the destruction of mosquitoes and the protection of the sick against the bites of these insects.

11. While the mode of propagation of yellow fever has now been definitely determined, the specific cause of this disease remains to be discovered.

Reed, standing second from right, Volunteers, and others at Camp Lazear, 1901

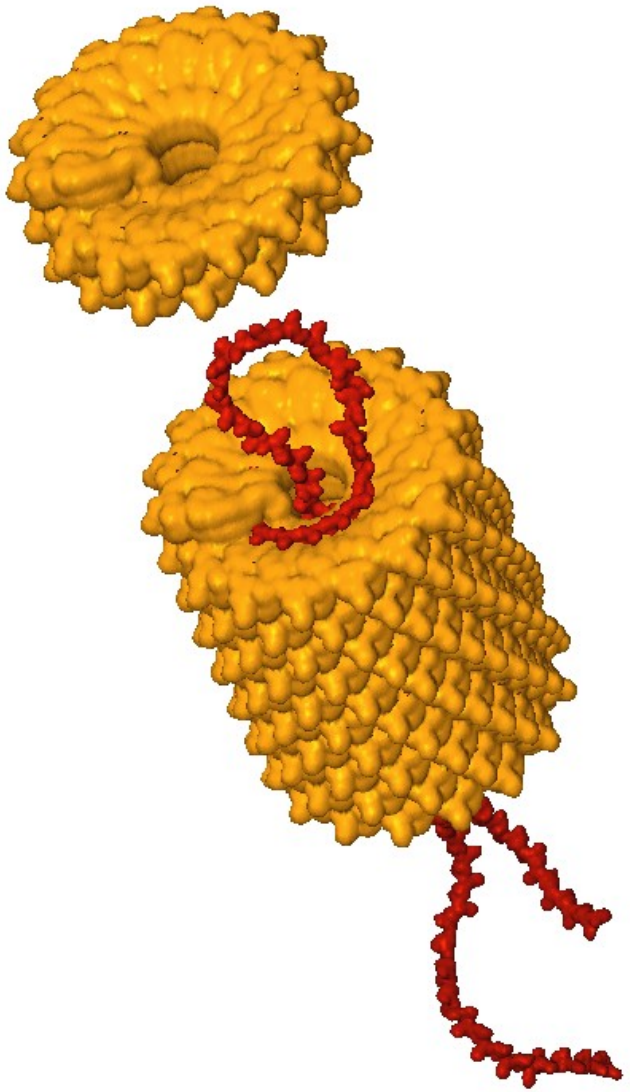
Celkem 22 experimentálních případů žluté zimnice
Kousnutí komárem – 14
Injikace krve – 6
Filtrované sérum - 2

Approximate Global Distribution of Yellow Fever, by State/Province, 2007



„Virus – to je špatná zpráva zabalená do bílkoviny“.

„Viry jsou jako Tolkienovi Nazgûlové, bytosti ani živé ani mrtvé, pohybující se v zemi nikoho, na frontě mezi životem a neživotem.“

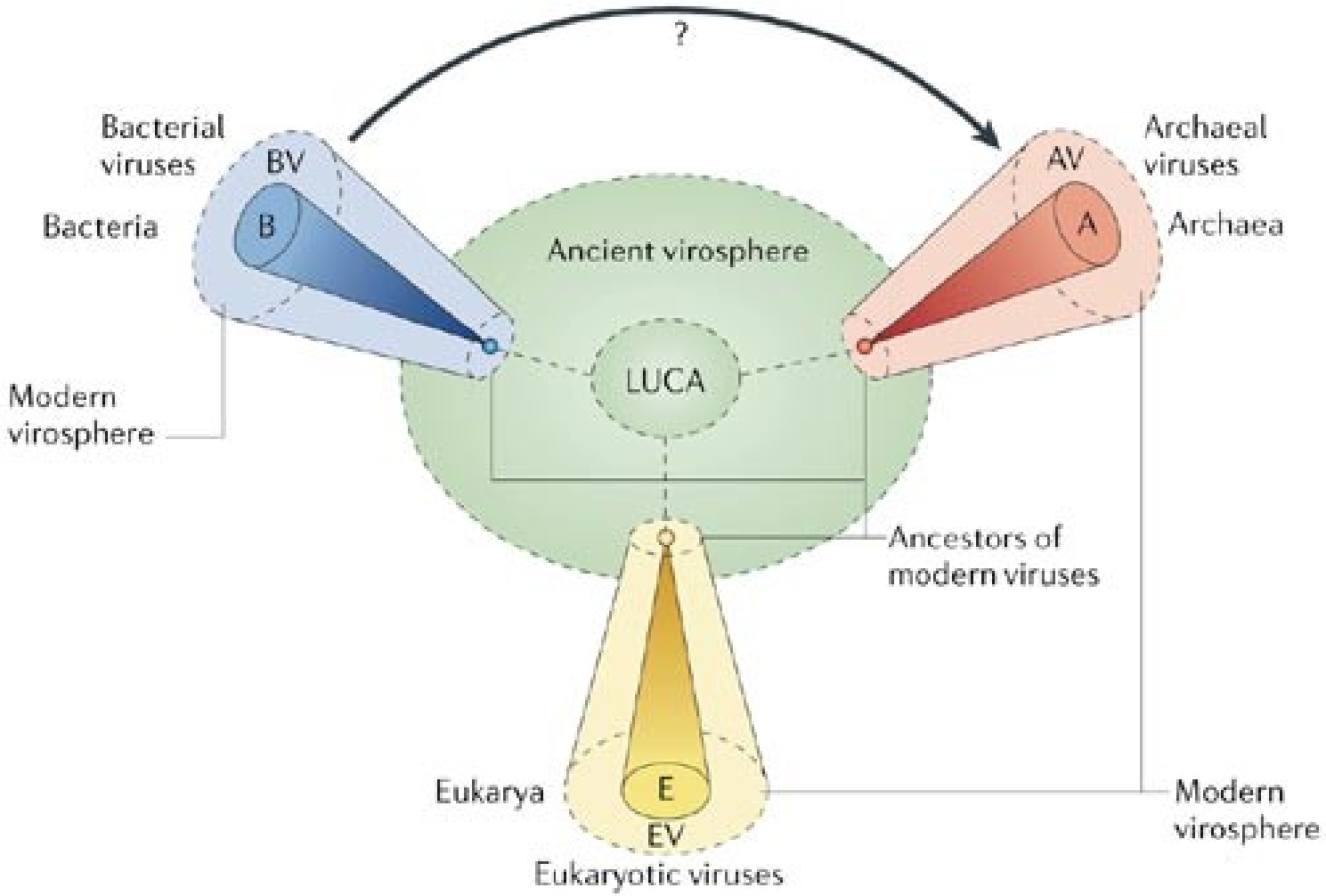


Jak viry vznikly?

Dobrá otázka, ale není na ni známa odpověď

Teorie:

1) Virus first – viry jsou přímí pokračovatelé předbuněčných živých entit
   x viry nejsou schopny množit se samy, potřebují hostitelskou buňku
2) Gene escape – viry vznikly ze „zdivočelých“ genů, které unikly z původních organizmů
   - příbuznost s plasmidy
   x kde se vzaly unikátní virové proteiny (kapsida)
3) Degenerace – viry se vyvinuly zjednodušením z vnitrobuněčných parazitů, kteří měli sami buněčnou strukturu (Rickettsie)
   x chybí přechodový článek



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	Růst na umělém médiu	Binární dělení	Mají současně DNA a RNA	Mají ribozómy	Mají kyselinu muramovou	Jsou citlivé k antibiotikům
Bakterie	Ano	Ano	Ano	Ano	Ano	Ano
Mykoplasmata	Ano	Ano	Ano	Ano	Ne	Ano
Rickettsie	Ne	Ano	Ano	Ano	Ano	Ano
Chlamydie	Ne	Ano	Ano	Ano	Ne	Ano
Viry	Ne	Ne	Ne	Ne *	Ne	Ne

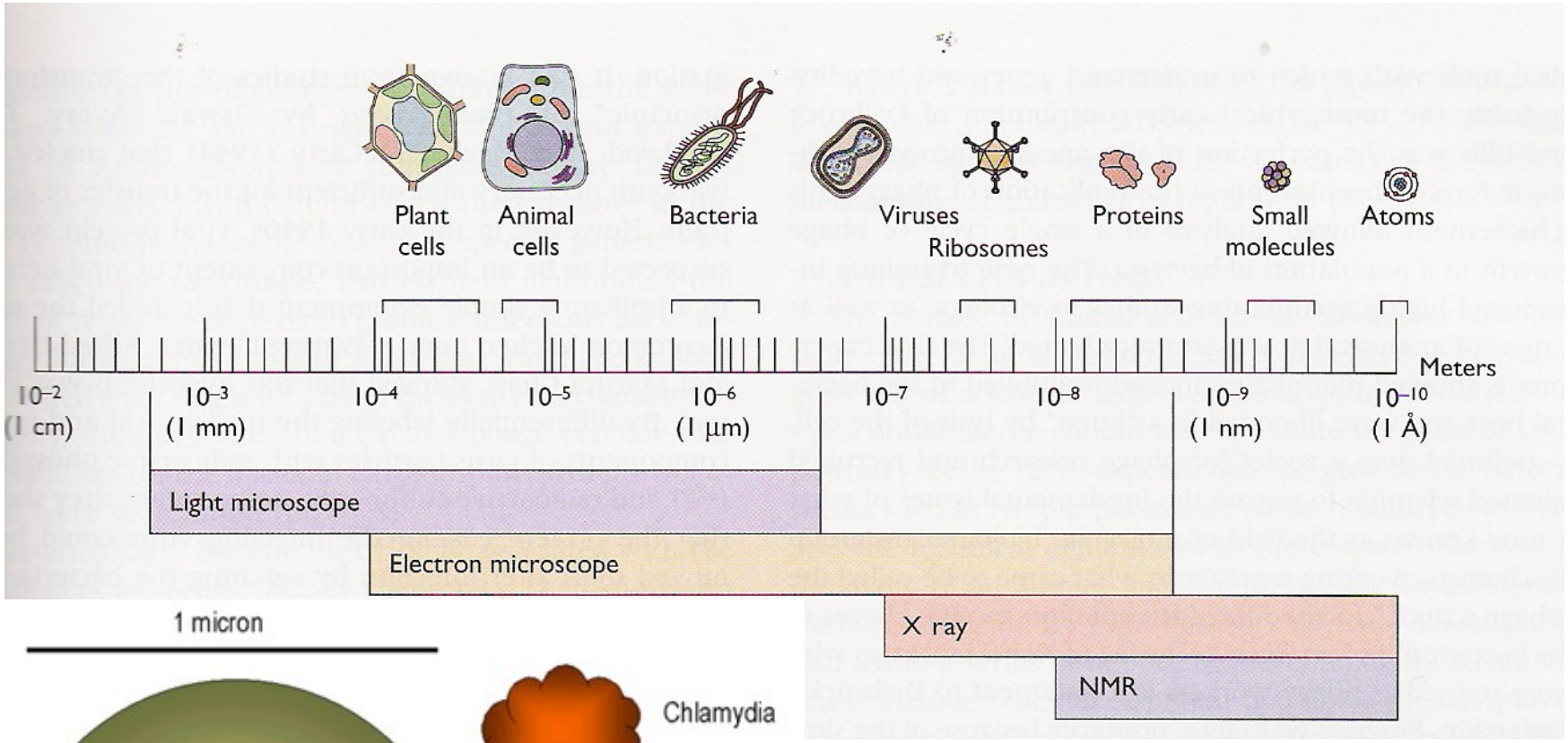
* Arenaviry mají ribozómy zabalené náhodně a nesehrávají žádnou roli při replikaci virové částice.

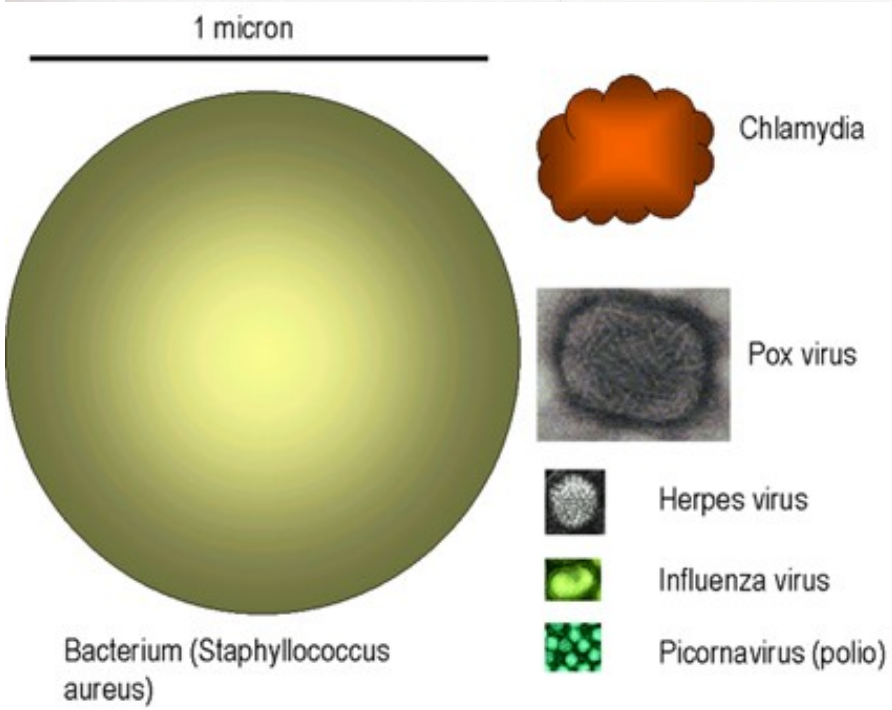
Vlastnosti virů obdobné buněčným organismům

1. **Schopnost rozmnožování**. Množení virů neprobíhá binárně jako je tomu u jiných organismů, tj. rozdělením virové částice na dvě nové, nýbrž tím způsobem, že jednotlivé složky nových virů jsou syntetizovány odděleně a posléze se z nich poskládá větší množství virových částic.
2. **Dědičné vlohy**. Nositelem dědičnosti je virový genom tvořený DNA nebo RNA a určující morfologické, strukturní a biologické vlastnosti virů.
3. **Proměnlivost**. Příkladem proměnlivosti vyvolané změnou virové genetické informace je např. vznik virových mutantů odolných vůči působení virostatik, adaptovaných na určité prostředí apod.
4. **Přizpůsobivost k vnějším podmínkám**.
5. **Schopnost vývoje**. Vývoj virů je podobně jako je tomu u všech organismů dán přirozeným výběrem. Vzhledem ke způsobu života viru a jeho obvykle vysoké rychlosti množení bývají vývojové etapy virů poměrně krátké.

Vlastnosti typické pouze pro viry

1. **Odlišný charakter genetického materiálu**. Zatímco genom všech organismů je tvořen výlučně molekulou DNA, u virů to může být RNA nebo DNA. Kromě toho může být virová nukleová kyselina jedno- i dvouřetězcová, kruhová, lineární nebo fragmentovaná.
2. **Specifický typ parazitismu**. Zatímco jiní parazité napadají organismy na buněčné nebo organismální úrovni, viry jsou parazité na úrovni genetické a biochemické, neboť v infikované buňce přejímají informační a řídící funkce.
3. **Nepřítomnost proteosyntetického aparátu**. Všechny organismy, včetně těch nejjednodušších, obsahují vlastní výbavu k syntéze bílkovin. U virů však tento aparát chybí a virové bílkoviny jsou syntetizovány pomocí aparátu hostitelské buňky.
4. **Nebuněčná forma života.** Viry postrádají buňku jako základní stavební a funkční útvar.


Plant cells
Animal cells
Bacteria
Viruses
Ribosomes
Proteins
Small molecules
Atoms
Meters
10–2 (1 cm)
10–3 (1 mm)
10–4
10–5
10–6 (1 µm)
10–7
10–8
10–9 (1 nm)
10–10 (1 Å)
Light microscope
Electron microscope
X ray
NMR


1 micron
Chlamydia
Pox virus
Herpes virus
Influenza virus
Picornavirus (polio)
Bacterium (Staphyllococcus aureus)

Struktura virů

Virion – jednotlivá částice viru

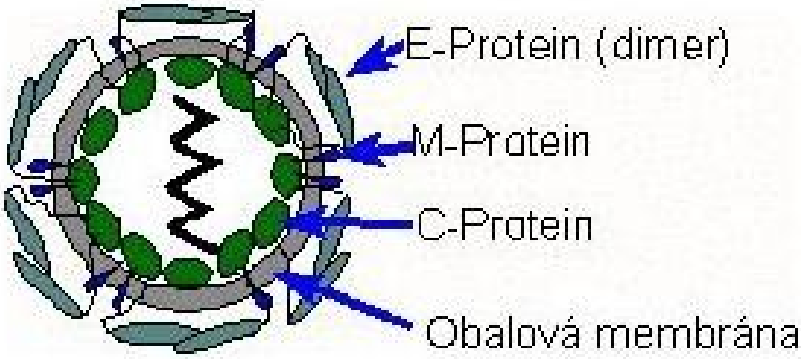
Kapsida – bílkovinný obal – hlavní stavební jednotka virionu, chrání NK, skládá se z kapsomer

Nukleokapsida – kapsida + NK

Kapsomera – morfologická jednotka, útvar viditelný v elektronovém mikroskopu

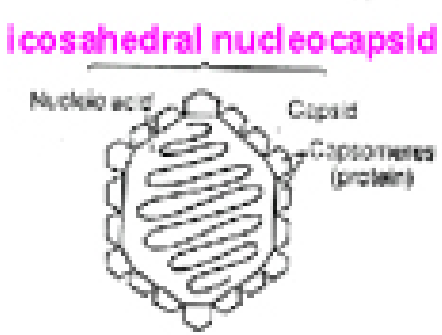
Protomera – strukturní jednotka, nejmenší stavební jednotka kapsidu, skládá se z jendoho nebo více ve většině případů identických polypeptidů

Virový obal – lipidová dvojvrstva odvozená od hostitelské buňky s glykoproteiny, které často tvoří výběžky (spikes, peplomery)

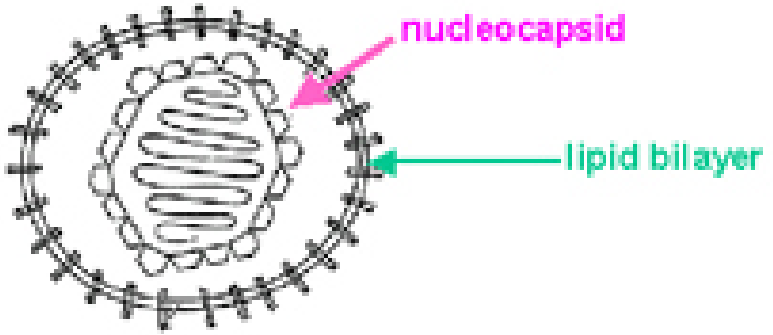


- Struktura virů vychází ze snahy zaujmout stav o co nejnižší volné energii

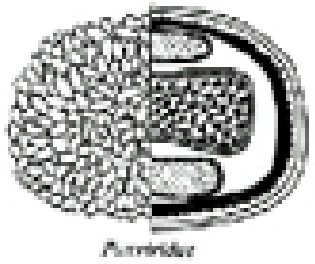
5 BASIC TYPES OF VIRAL SYMMETRY



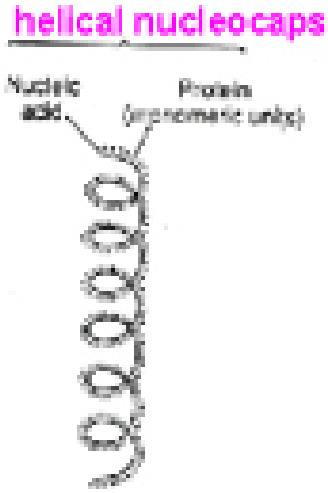
ICOSAHEDRAL



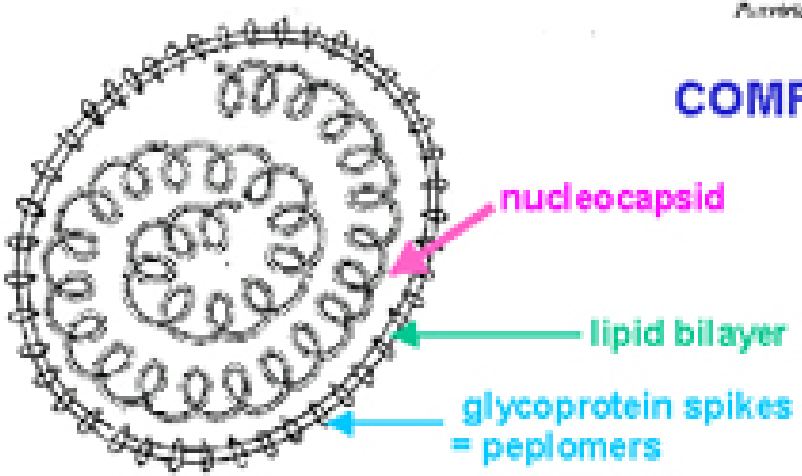
ENVELOPED ICOSAHEDRAL



COMPLEX

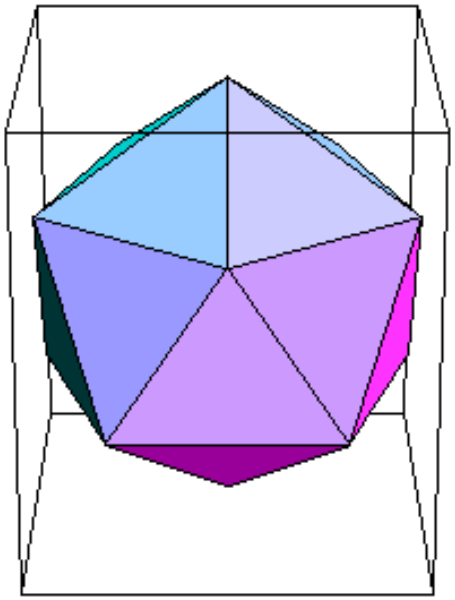


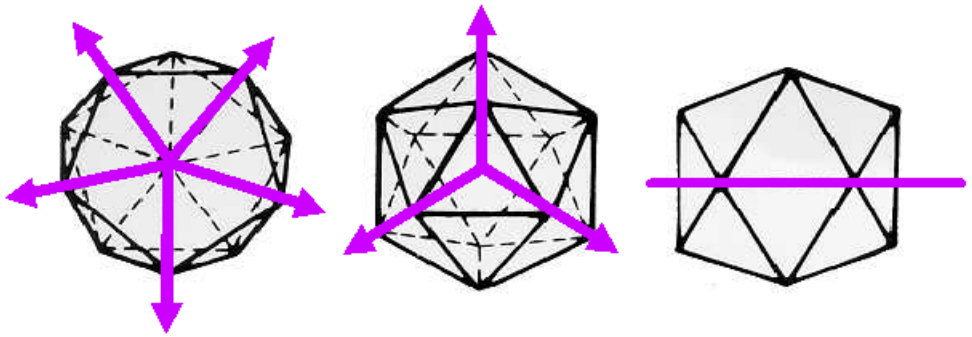
HELICAL



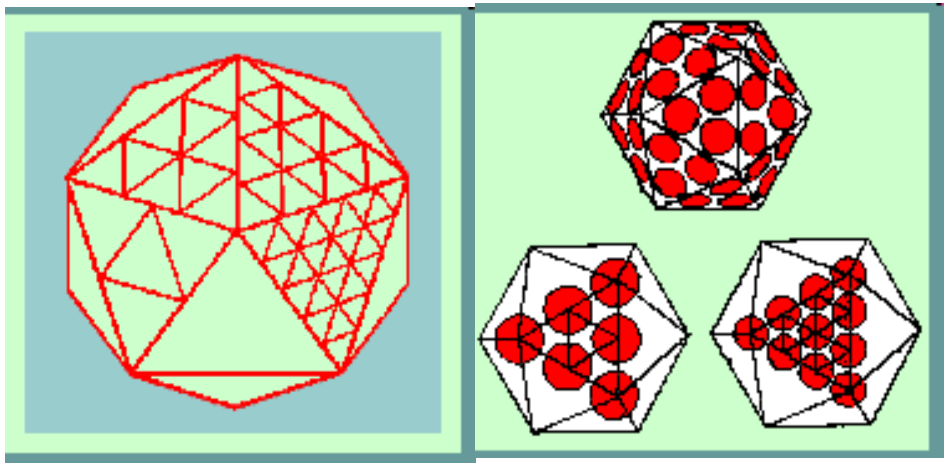
ENVELOPED HELICAL

ICOSAHEDRAL SYMMETRY

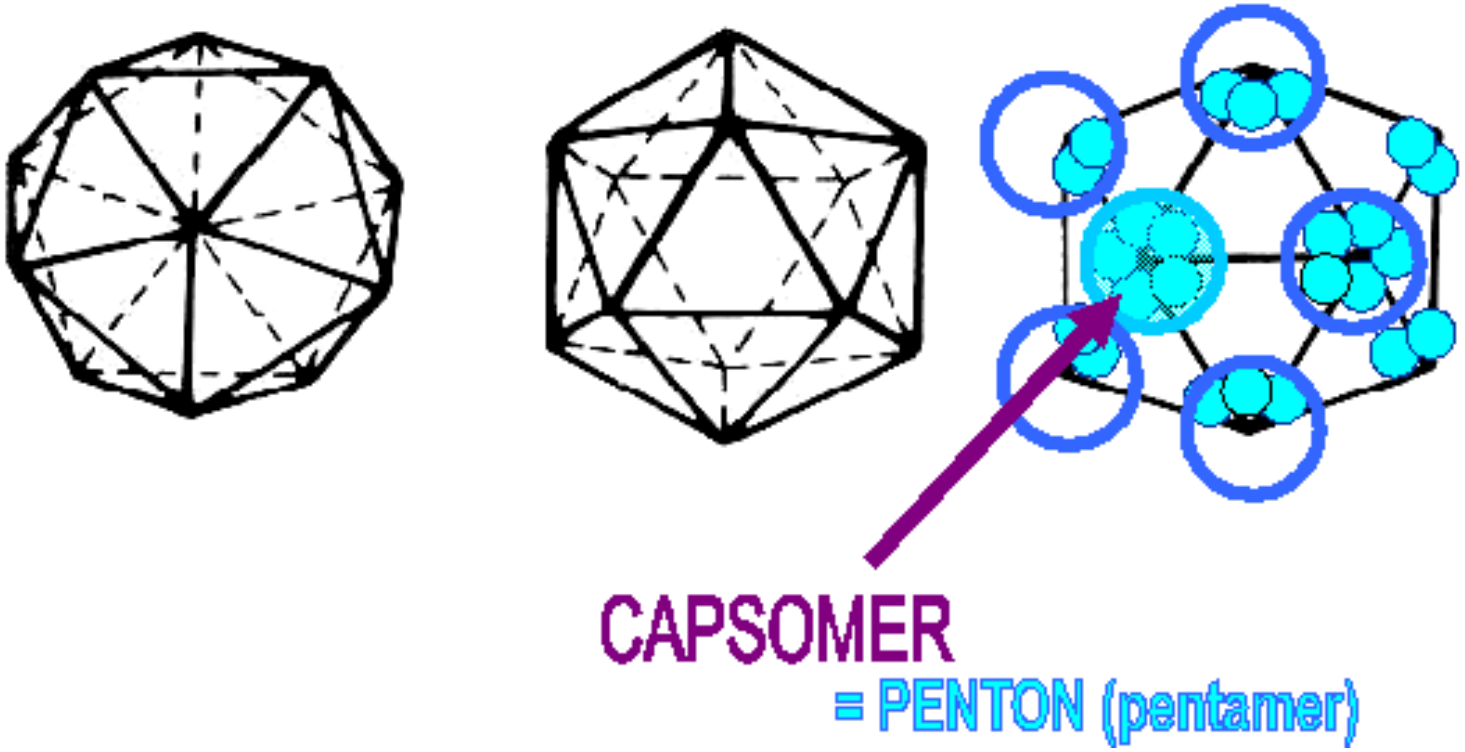




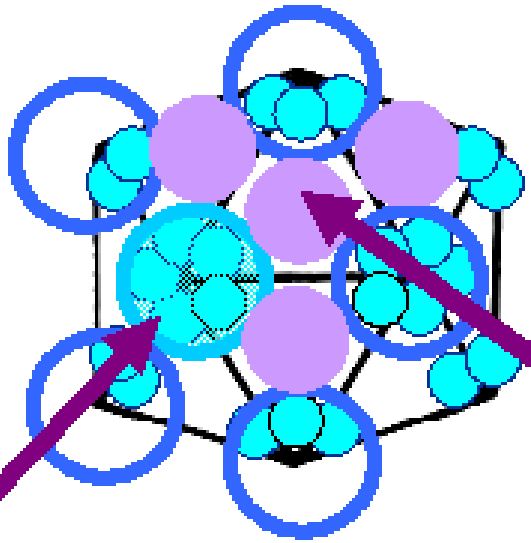
5-FOLD **3-FOLD** **2-FOLD**



ICOSAHEDRAL SYMMETRY



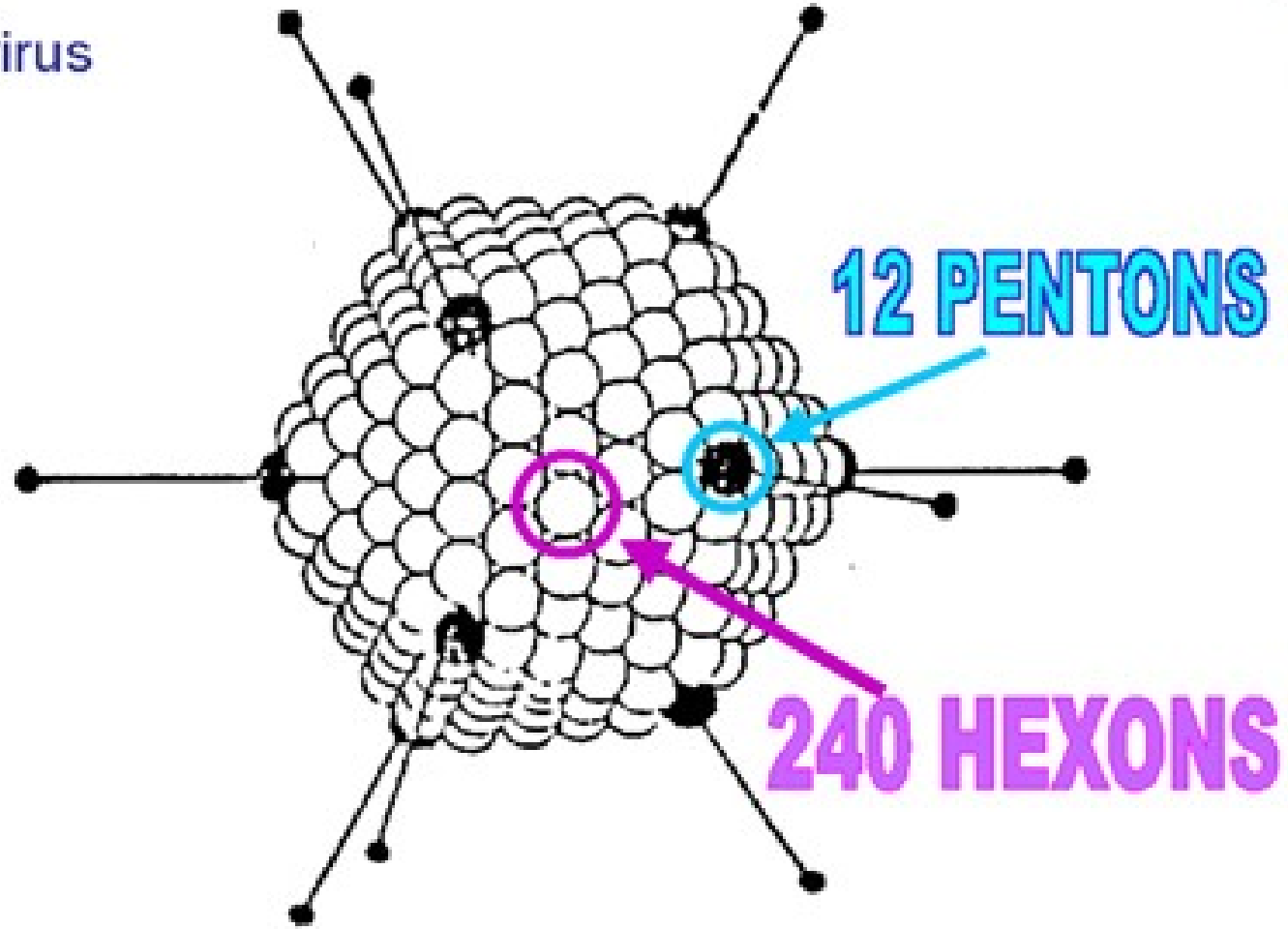
ICOSAHEDRAL SYMMETRY

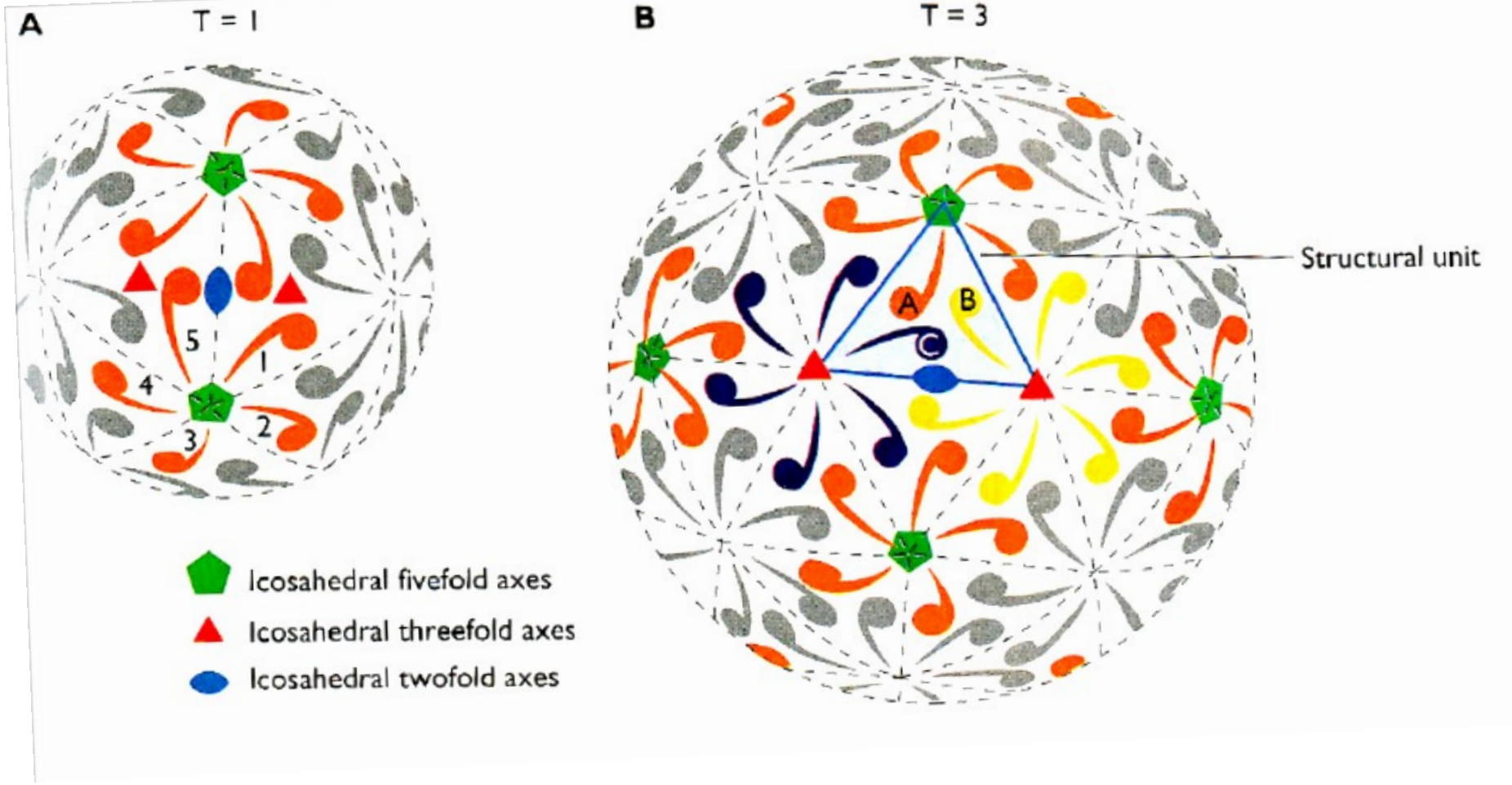


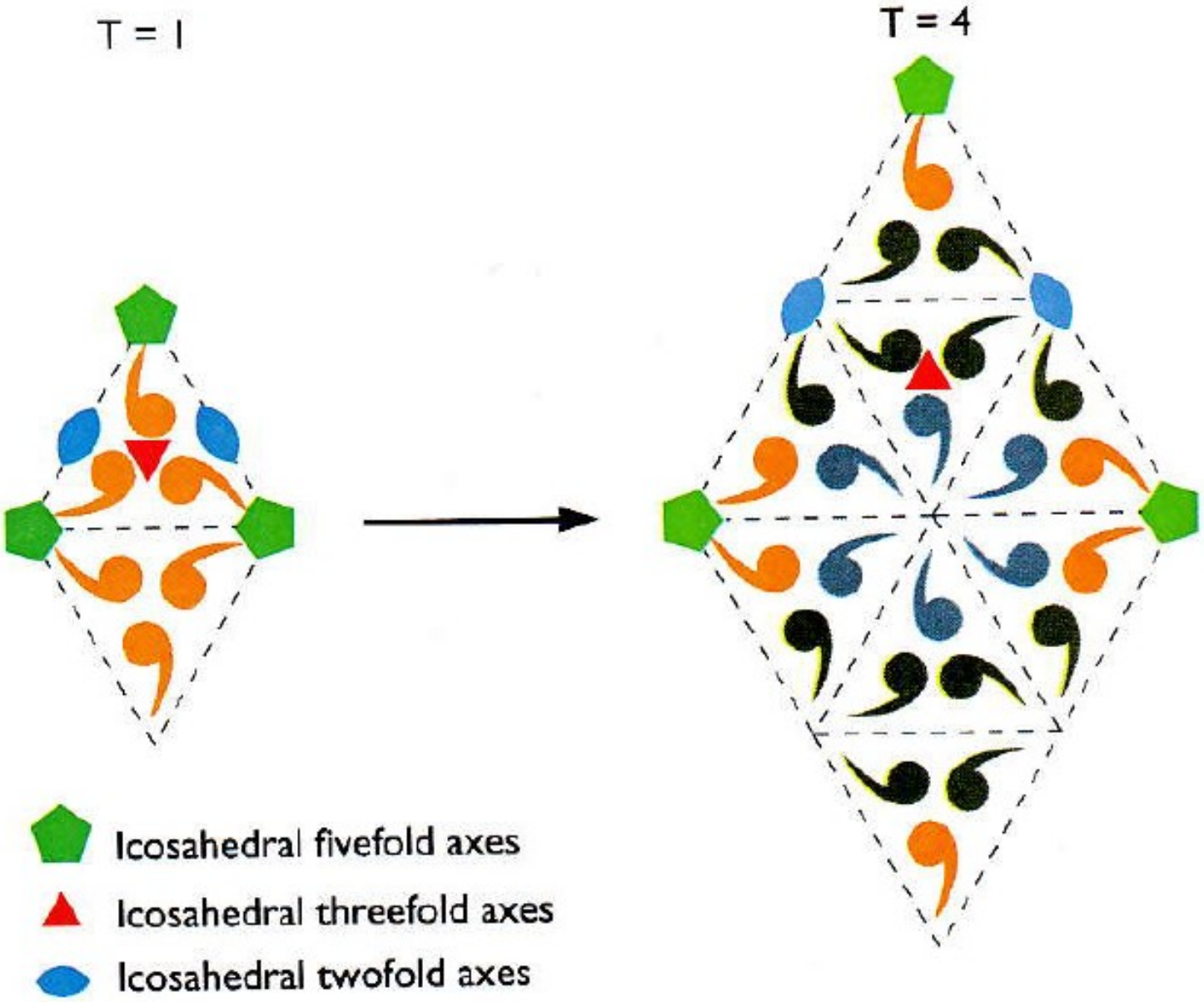
CAPSOMER
= PENTON

CAPSOMER
= HEXON

Adenovirus

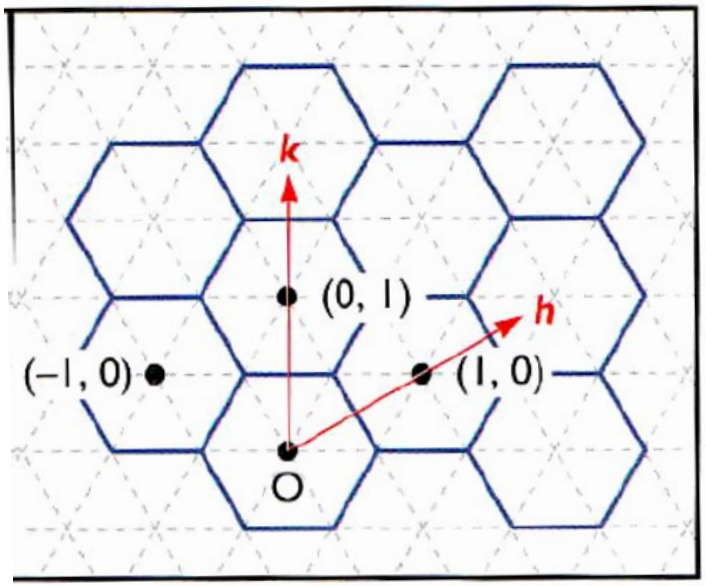


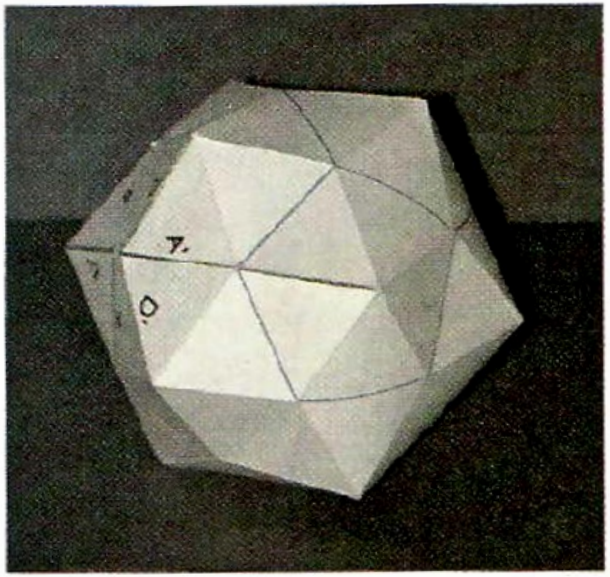

A
T = 1
5
1
4
2
3
Icosahedral fivefold axes
Icosahedral threefold axes
Icosahedral twofold axes
B
T = 3
A
B
C
Structural unit

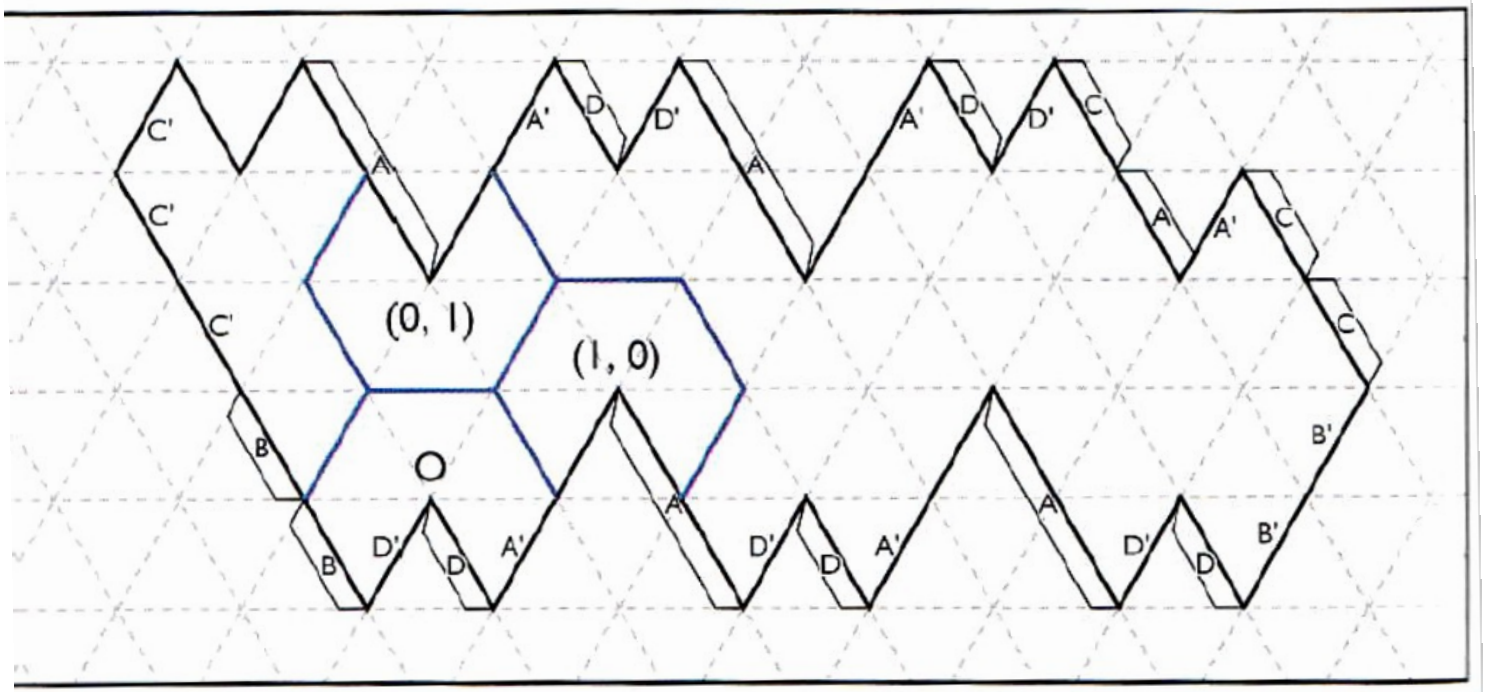

T = 1
T = 4
Icosahedral fivefold axes
Icosahedral threefold axes
Icosahedral twofold axes

A

$T = 1$

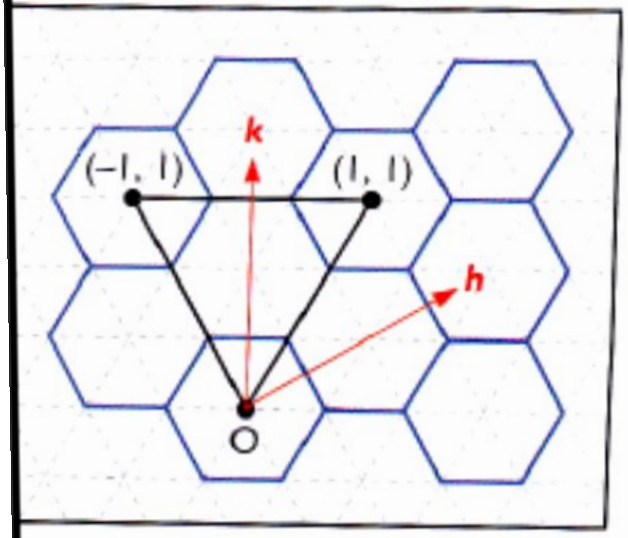

k
h
(0, 1)
(−1, 0)
(1, 0)
O

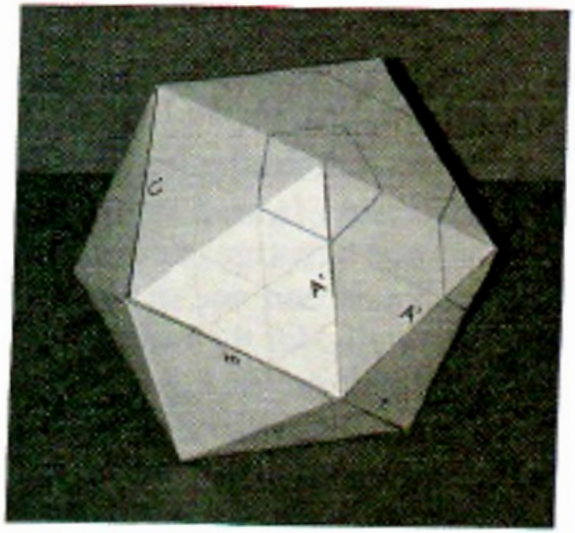


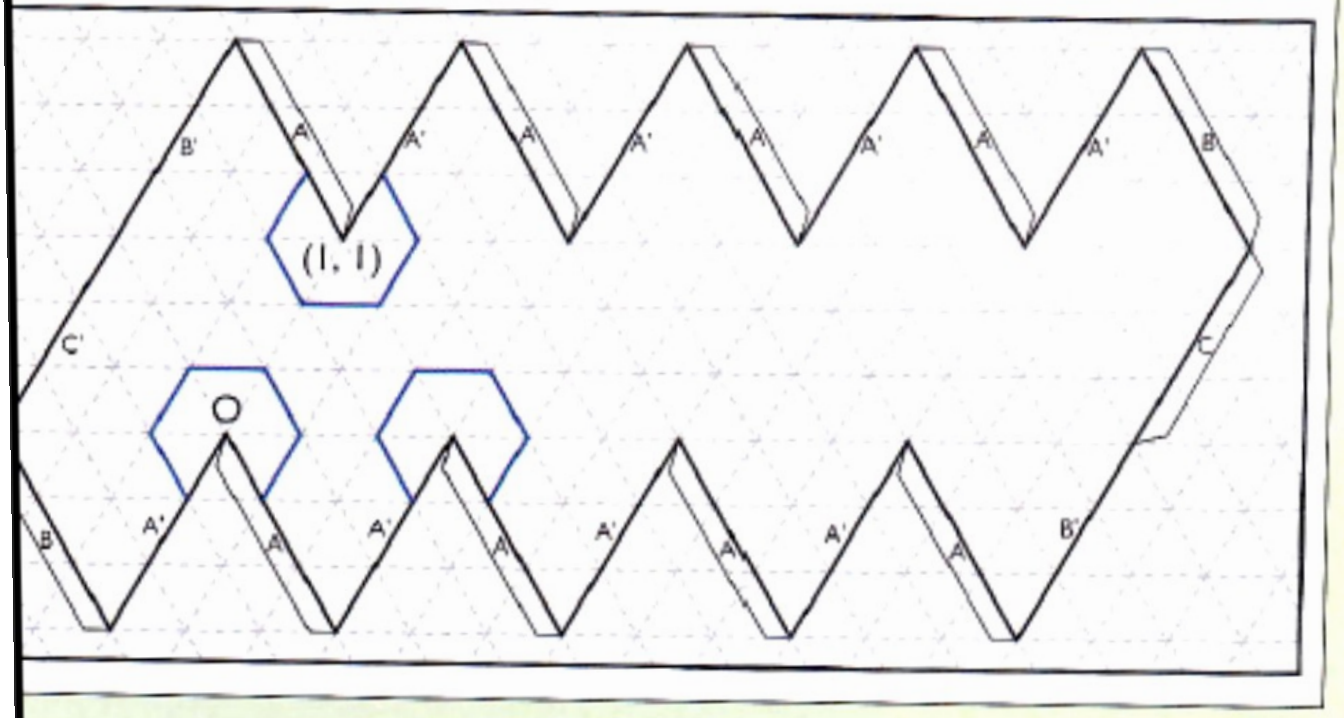

C'
C'
C'
A'
A'
D
D'
A
A'
D
D'
A
A'
D
D'
C
A
A'
C
C
B'
B'
B'
(0, 1)
(1, 0)
O
B
B
D'
D
A'
A
D'
D
A'
A
D'
D
A'

B

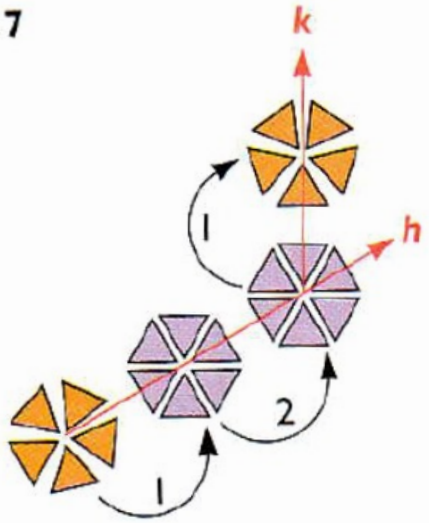
$T = 3$





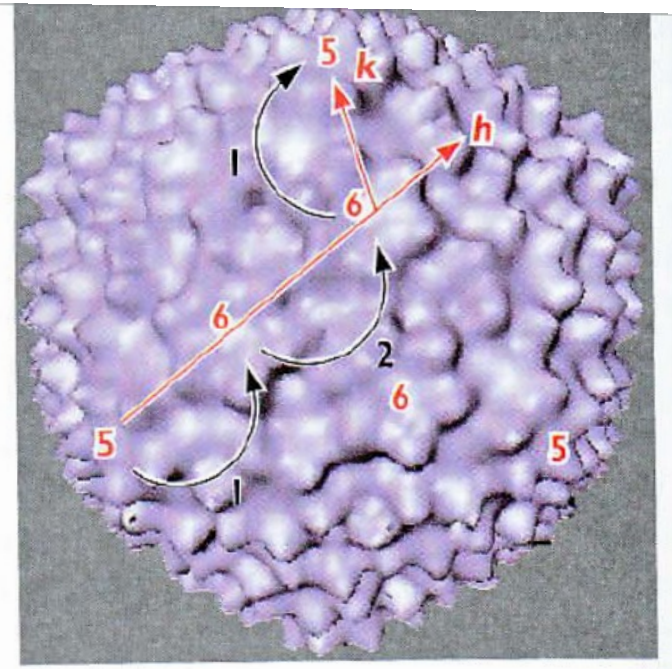


D **P22, $T = 7$**

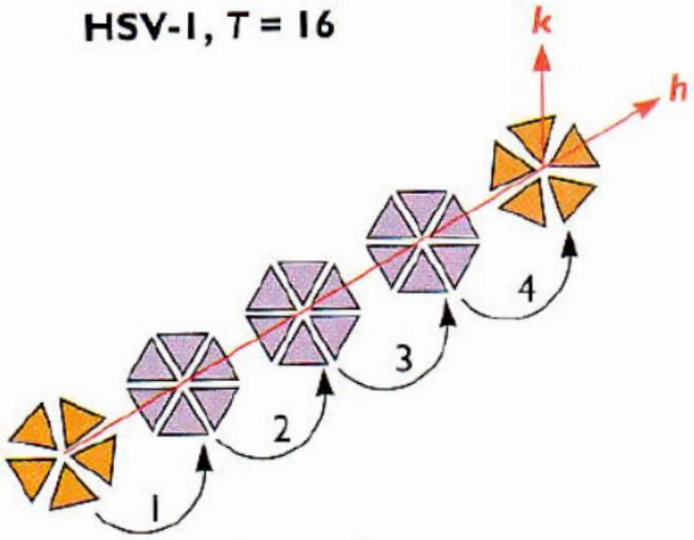


$h = 2, k = 1$

$\therefore T = (2)^2 + (2)(1) + (1)^2 = 7$

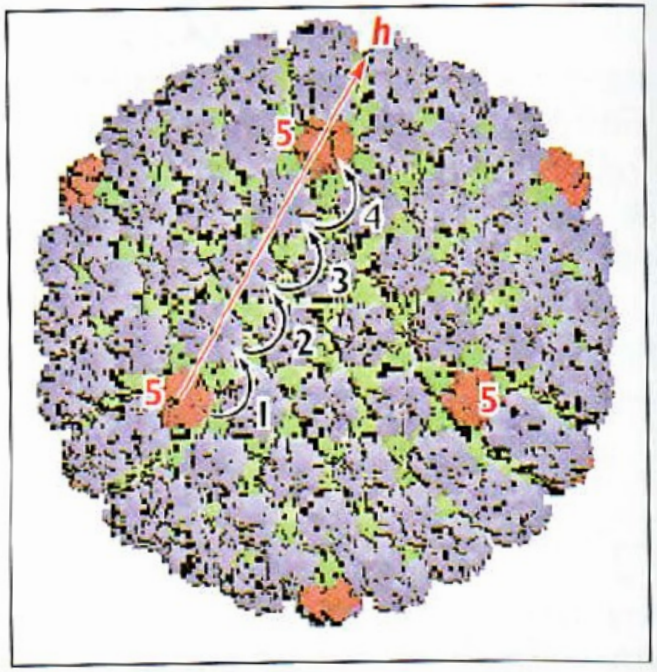


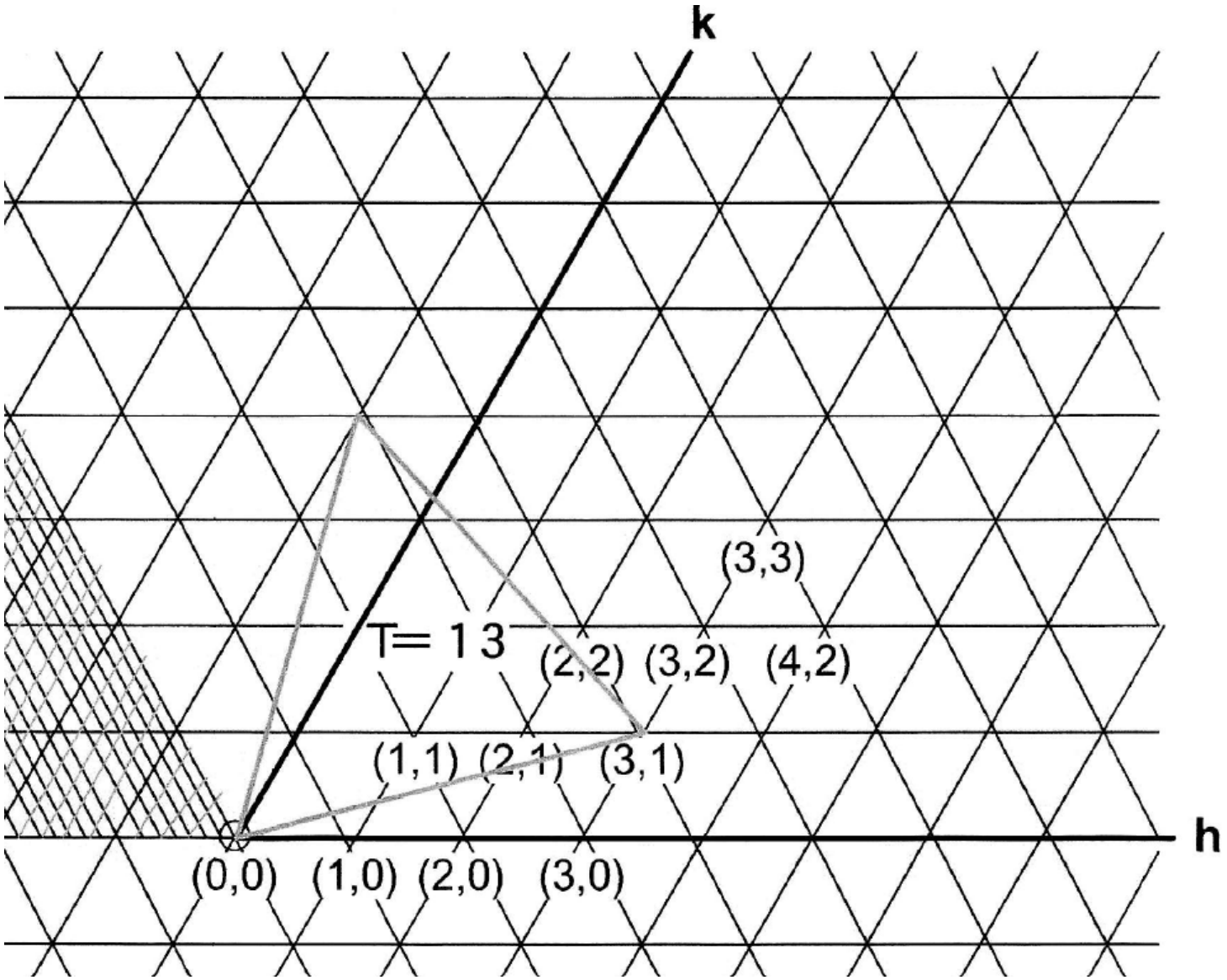
HSV-1, $T = 16$

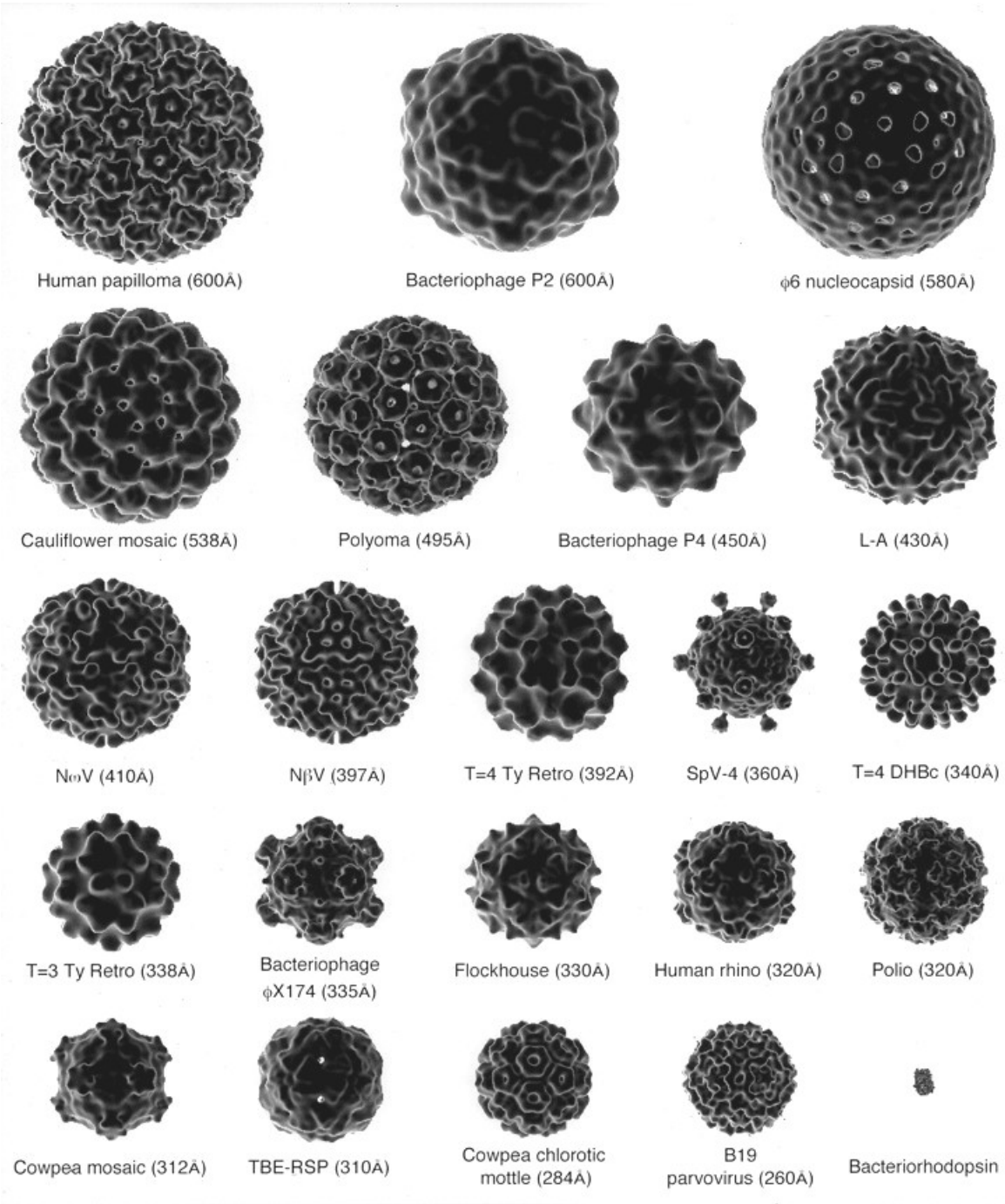


$h = 4, k = 0$

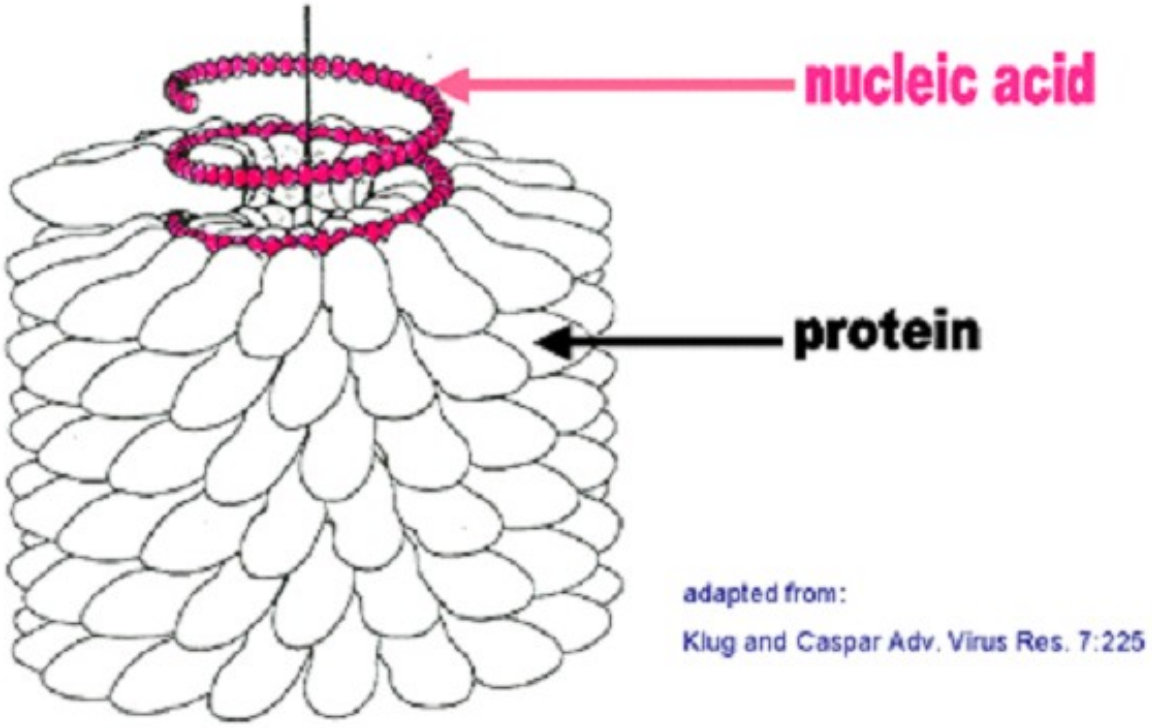
$\therefore T = (4)^2 + (4)(0) + (0)^2 = 16$

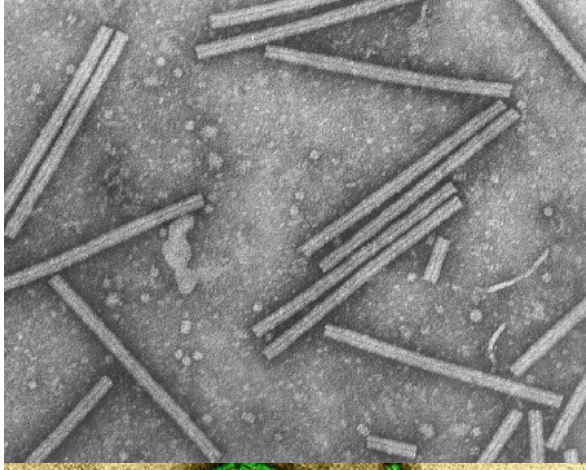


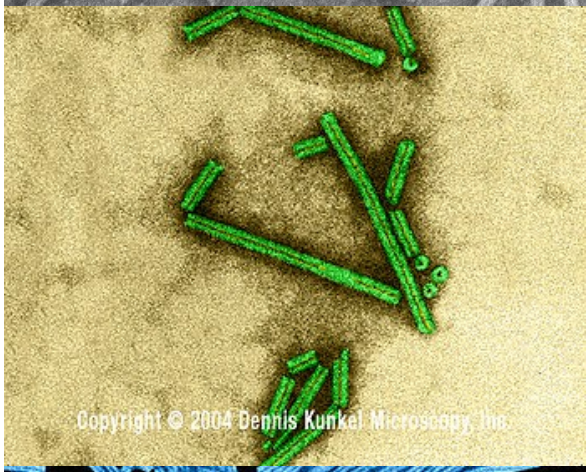

k
h
(3,3)
T= 1 3
(2,2)
(3,2)
(4,2)
(1,1)
(2,1)
(3,1)
(0,0)
(1,0)
(2,0)
(3,0)


Human papilloma (600Å)
Bacteriophage P2 (600Å)
ϕ6 nucleocapsid (580Å)
Cauliflower mosaic (538Å)
Polyoma (495Å)
Bacteriophage P4 (450Å)
L-A (430Å)
NωV (410Å)
NβV (397Å)
T=4 Ty Retro (392Å)
SpV-4 (360Å)
T=4 DHBc (340Å)
T=3 Ty Retro (338Å)
Bacteriophage ϕX174 (335Å)
Flockhouse (330Å)
Human rhino (320Å)
Polio (320Å)
Cowpea mosaic (312Å)
TBE-RSP (310Å)
Cowpea chlorotic mottle (284Å)
B19 parvovirus (260Å)
Bacteriorhodopsin

TOBACCO MOSAIC VIRUS



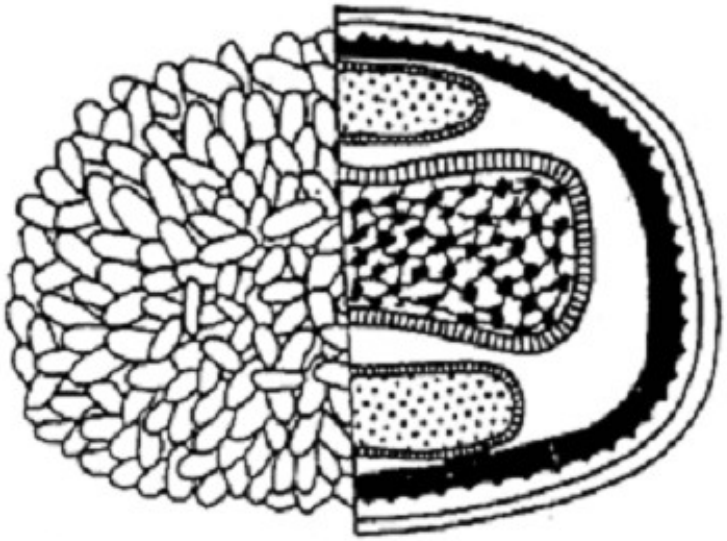




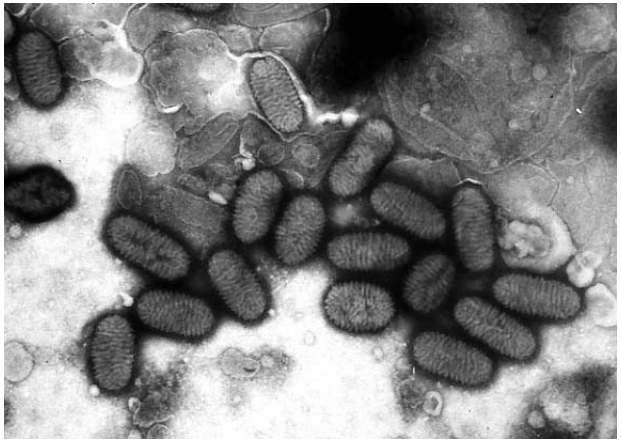
Průměr 18 nm, délka 300 nm, délku určuje délka NK

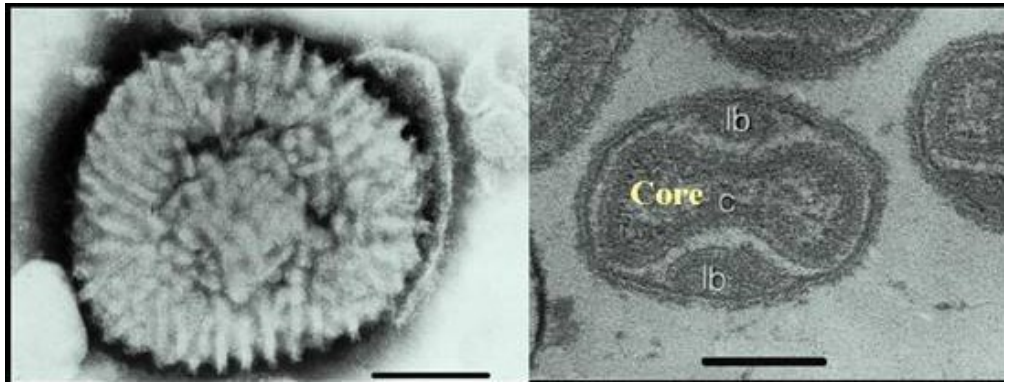
Monomery mají tendenci polymerizovat i v nepřítomnosti NK

COMPLEX SYMMETRY

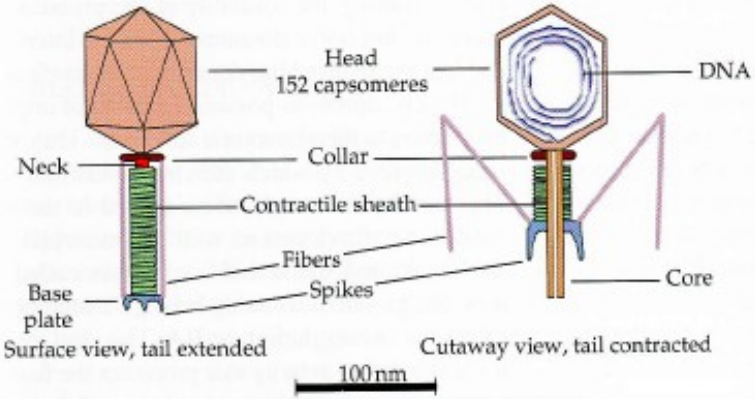


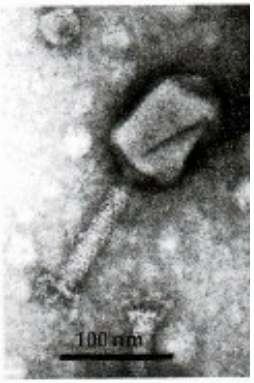
POXVIRUS FAMILY





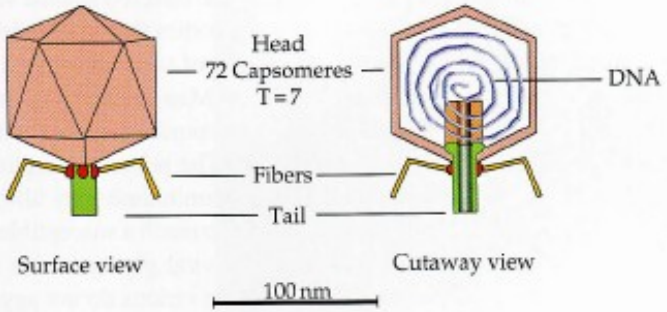
A. Enterobacteria phage T2

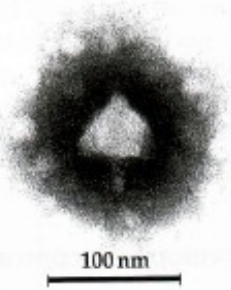




Electron micrograph

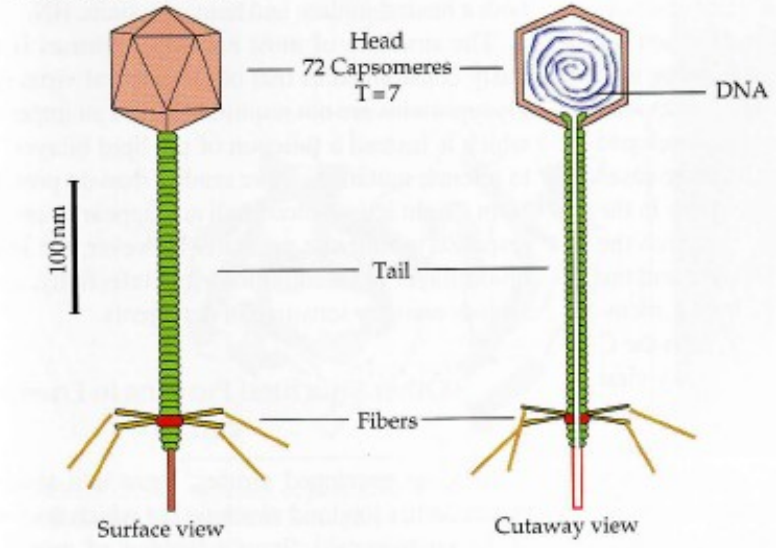
B. Enterobacteria phage T7

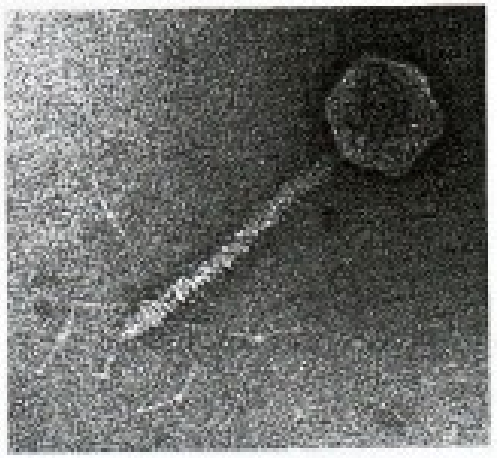




Electron micrograph

C. Lambda-like Phage

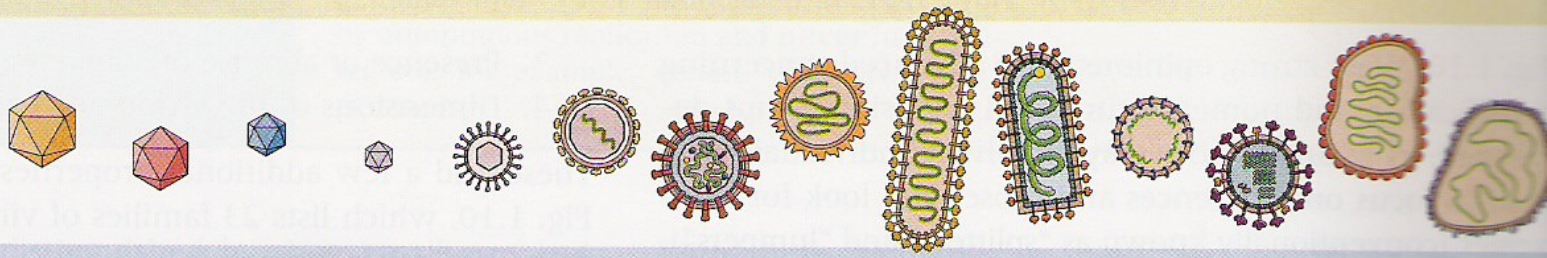




Electron micrograph

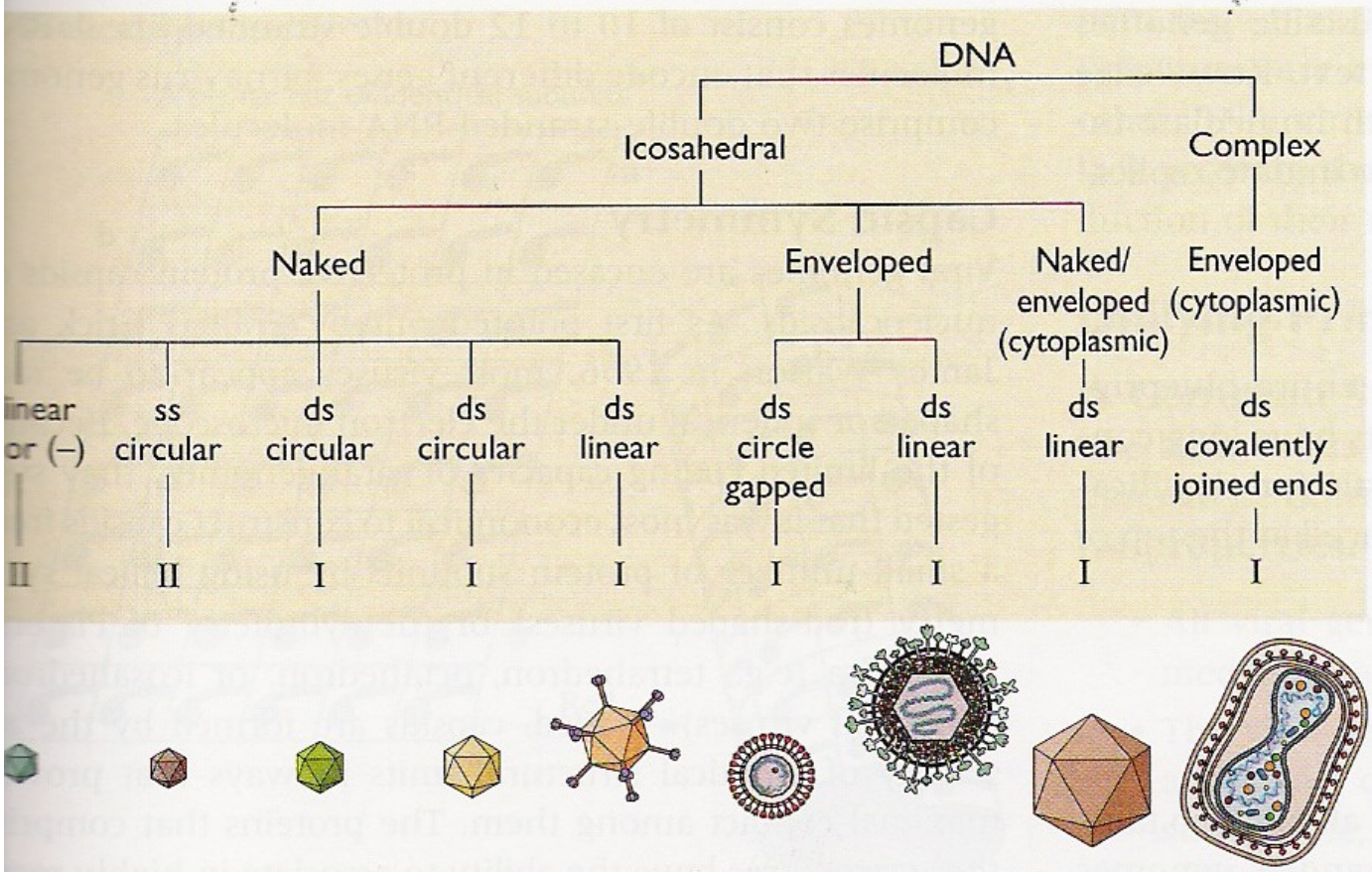
Classification criteria

Nucleic acid	RNA												
Symmetry of capsid	Icosahedral							Helical					
Naked or enveloped	Naked				Enveloped			Enveloped					
Genome architecture	ds 10–18 segments	ds 2 segments	(+) ss	(+) ss	(+) ss	(+) ss	(+) ss 2 copies	(+) ss	(–) ss	(–) ss	(–) ss 3 segments	(–) ss 8 segments	(–) ss	(–) ss 2 segments
Baltimore class	III	III	IV	IV	IV	IV	VI	IV	V	V	V	V	V	V



Properties

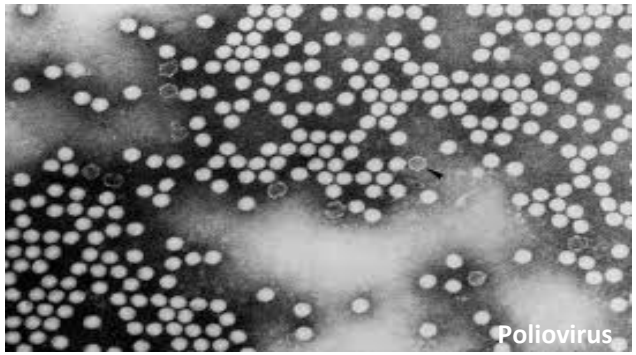
	Reo	Birna	Calici	Picorna	Flavi	Toga	Retro	Corona	Filo	Rhabdo	Bunya	Ortho-myxo	Para-myxo	Arena
Virion polymerase	(+)	(+)	(–)	(–)	(–)	(–)	(+)	(–)	(+)	(+)	(+)	(+)	(+)	(+)
Virion diameter (nm)	60–80	60	35–40	28–30	40–50	60–70	80–130	80–160	80 × 790–14,000	70–85 × 130–380	90–120	90–120	150–300	50–300
Genome size (total in kb)	22–27	7	8	7.2–8.4	10	12	3.5–9	16–21	12.7	13–16	13.5–21	13.6	16–20	10–14

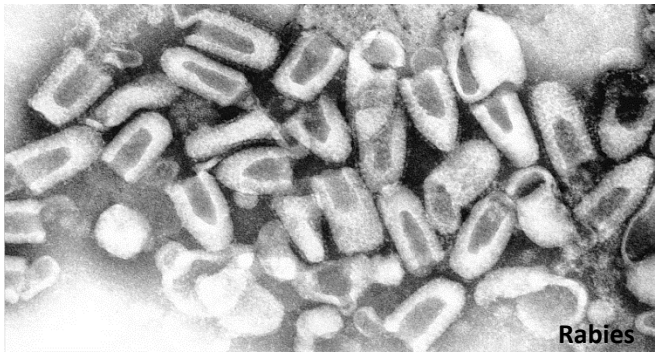

DNA
Icosahedral
Complex
Naked
Enveloped
Naked/ enveloped (cytoplasmic)
Enveloped (cytoplasmic)
linear or (–)
ss circular
ds circular
ds circular
ds linear
ds circle gapped
ds linear
ds linear
ds covalently joined ends
II
II
I
I
I
I
I
I
I
arvo
Circo
Polyoma
Papilloma
Adeno
Hepadna
Herpes
Irido
Pox
(–)
(–)
(–)
(–)
(–)
(+)
(–)
(–)
(+)
3–26
12–26
40
55
70–90
42
150–200
125–300
170–200 × 300–450
5
1.8–2.3
5
7–8
36–38
3.2
120–200
150–350
130–280

Dělení podle hostitele

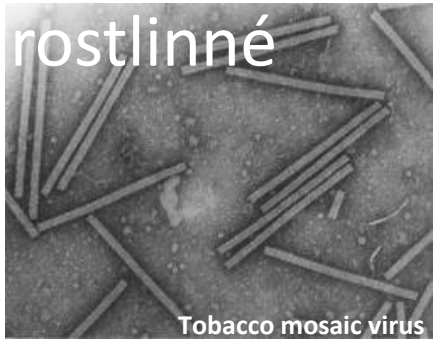
živočišné

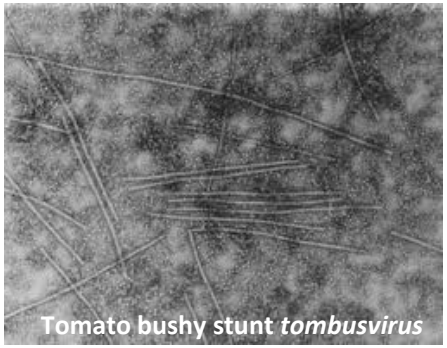

Human immunodeficience virus

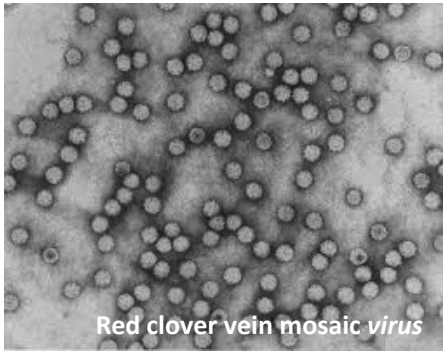

Poliovirus

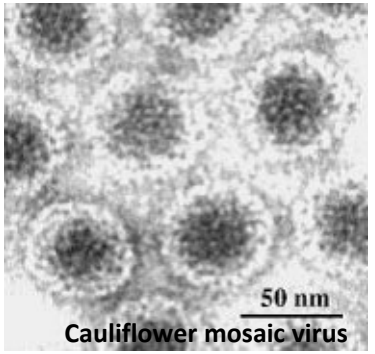

Rabies

rostlinné


Tobacco mosaic virus

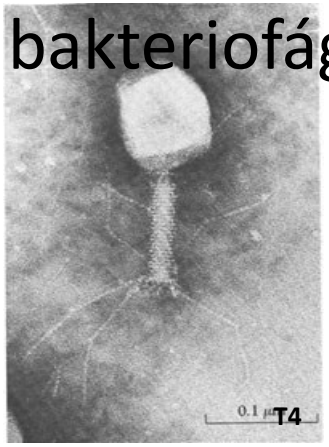

Tomato bushy stunt *tombusvirus*


Red clover vein mosaic *virus*

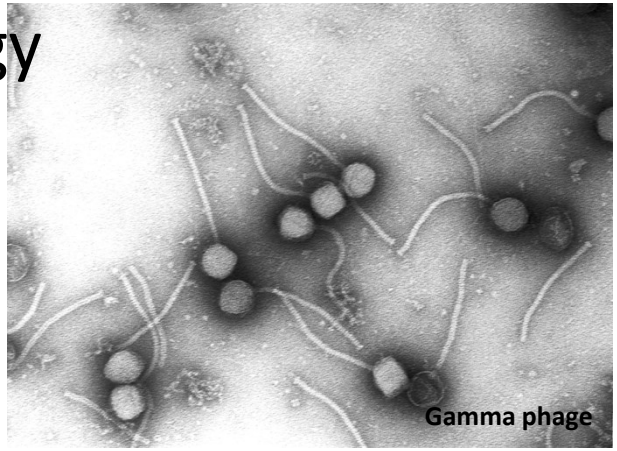


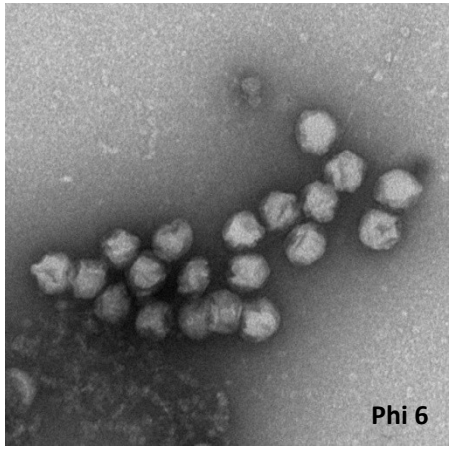
Cauliflower mosaic virus

bakteriofágy



T4

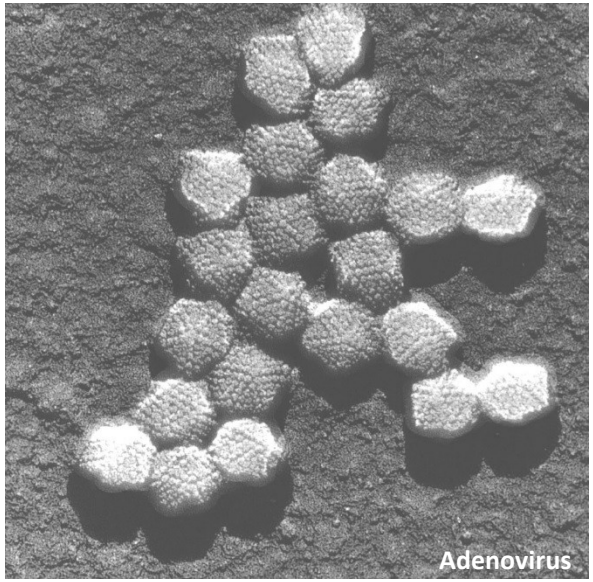

Gamma phage


Phi 6

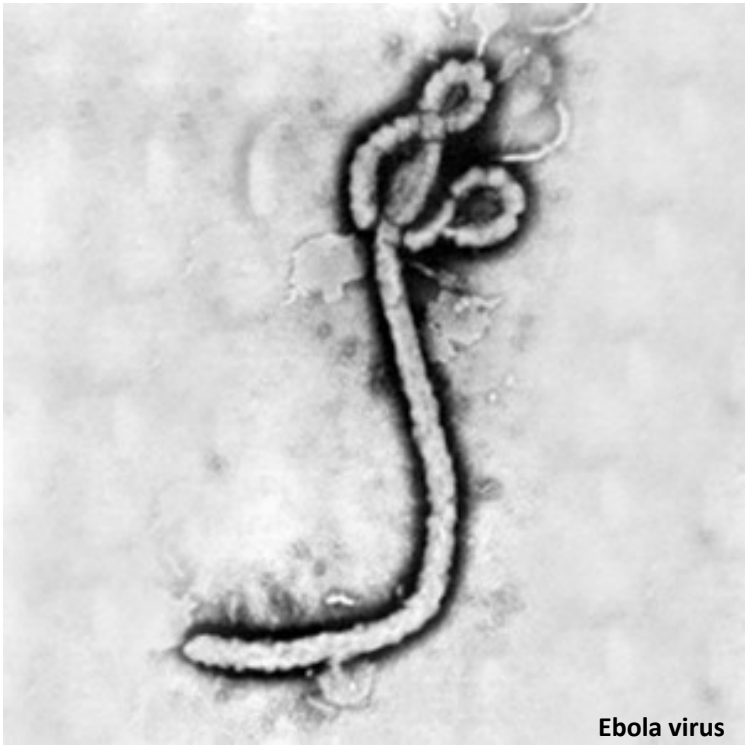

MS2

Podle morfologie kapsidy

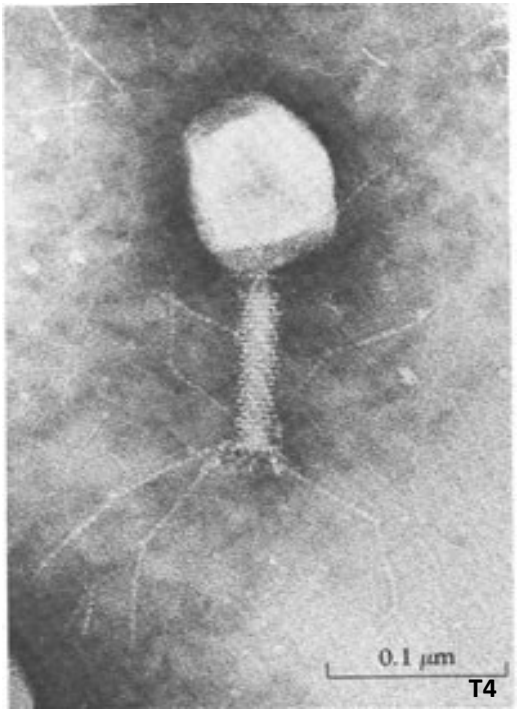
ikosahedrální



helikální

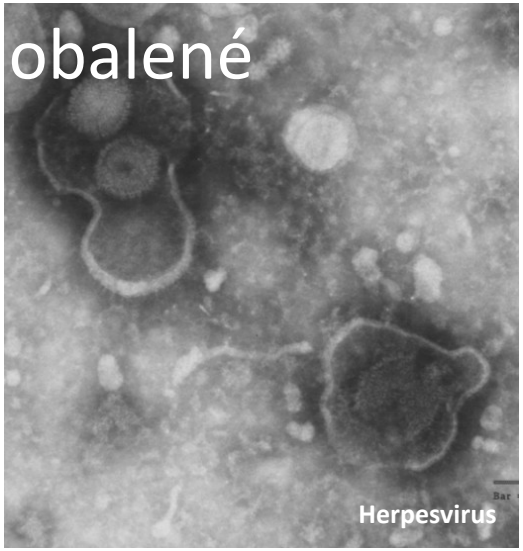


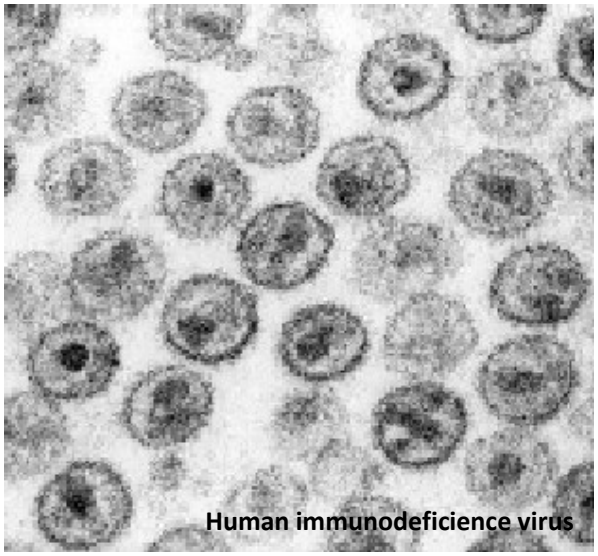
komplexní

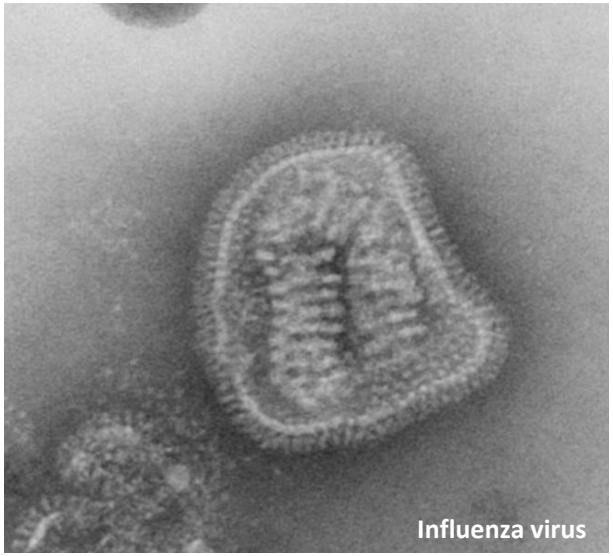


Podle obalu

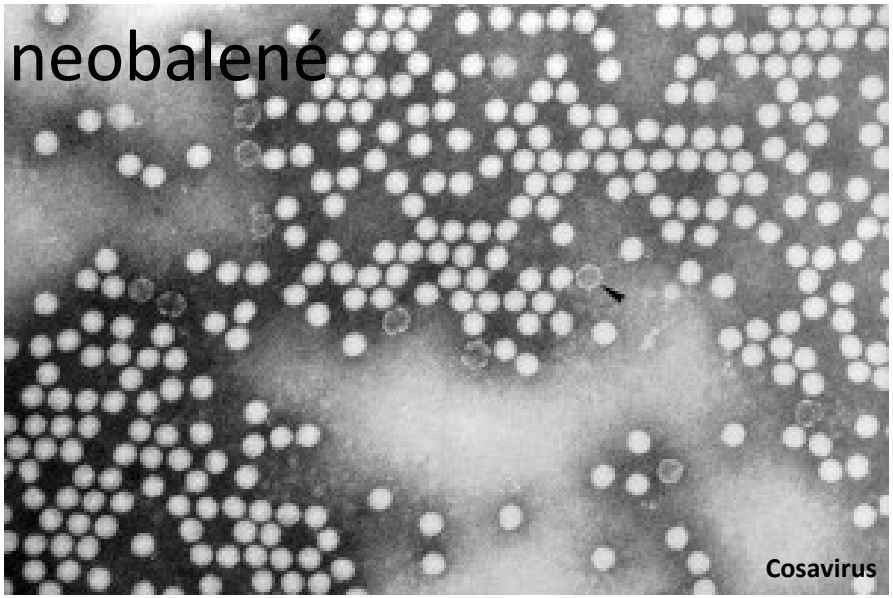
obalené

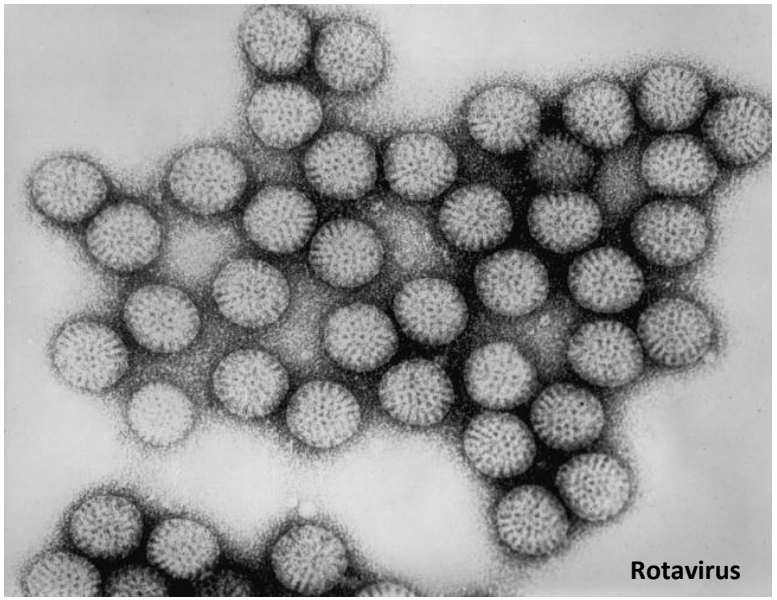






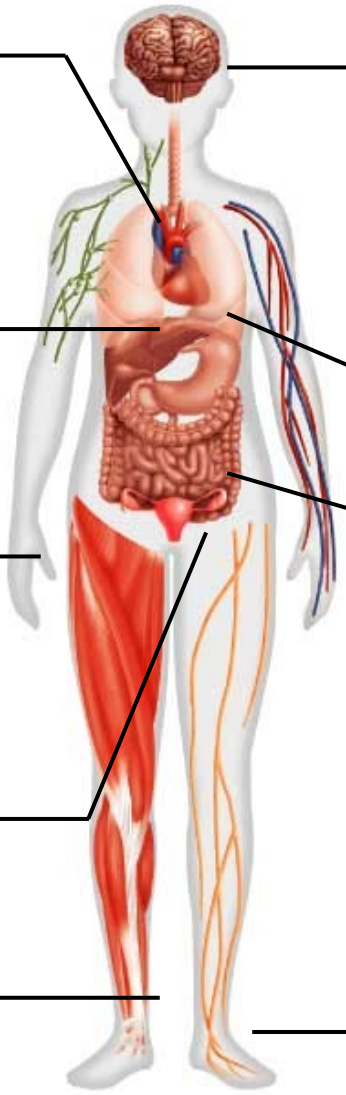
neobalené





Podle afinity (tropismu)

buněčná specifita x orgánová a tkáňová specifita



Lymphatic system
- Epstein-Barr
- HIV
- paramyxovirus (e.g., measles)

Trachea and lungs
- parainfluenza
- RSV
- influenza
- adenovirus

Skin
- rubella
- variola
- papillomavirus
- herpes 1
- molluscum contagiosum

Reproductive system
- herpes 2
- papillomavirus

Skeletal muscles
- coxsackie virus

Brain and CNS
- encephalitis
- rabies
- polio virus
- herpes zoster
- yellow fever
- Ebola
- dengue
- West Nile virus

Heart
- coxsackie virus

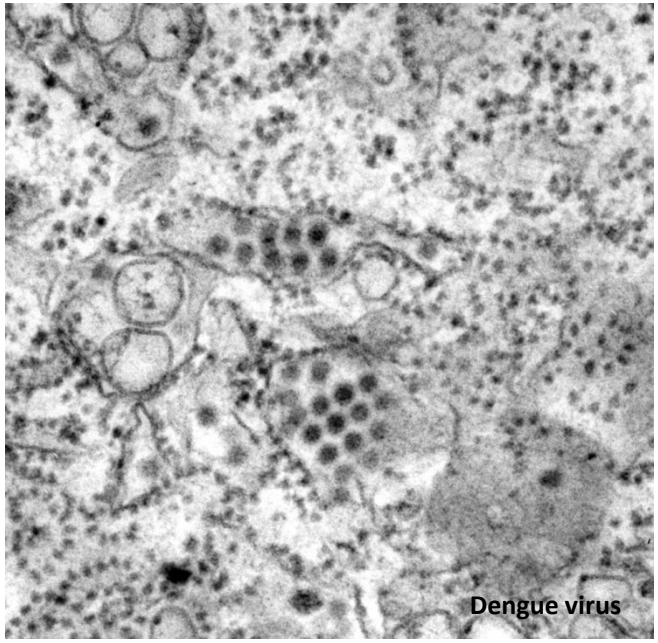
Gastrointestinal tract and liver
- hepatitis A, B, C, D, E
- rotavirus, • poliovirus

Blood vessels and blood cells
- erythrovirus
- Ebola virus
- Hantavirus

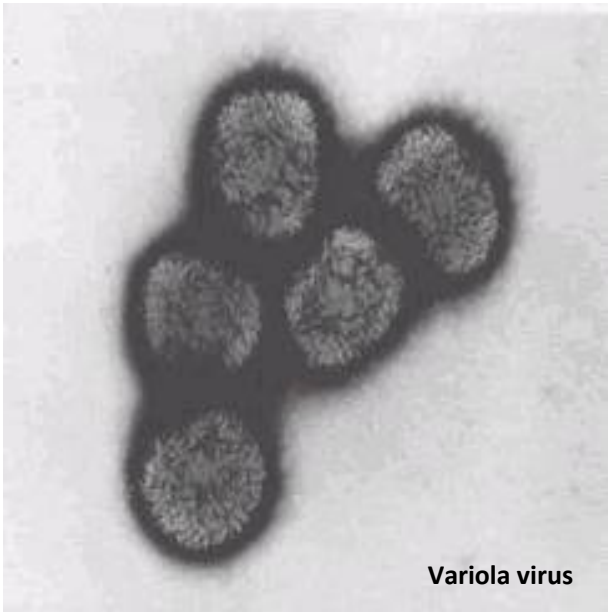
Peripheral nerves
- rabies

Podle nukleové kyseliny

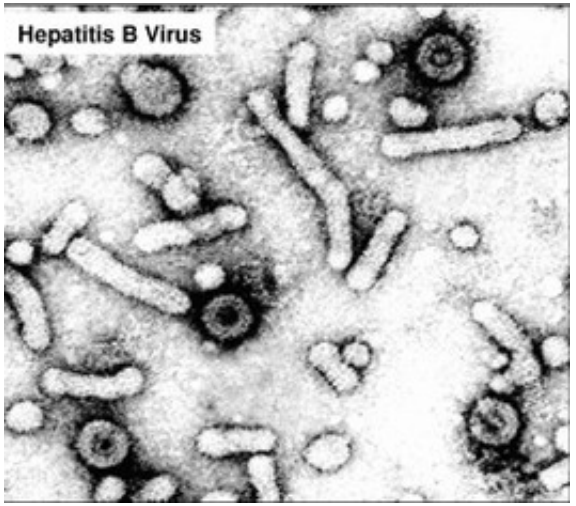
RNA



DNA



RNA ←→ DNA



Baltimore classification

- kombinuje podobu nukleové kyseliny a replikační strategie
- 7 tříd

David Baltimore

I: **dsDNA viruses** (e.g. Adenoviruses, Herpesviruses, Poxviruses)

II: **ssDNA viruses** (+ strand or "sense") DNA (e.g. Parvoviruses)

III: **dsRNA viruses** (e.g. Reoviruses)

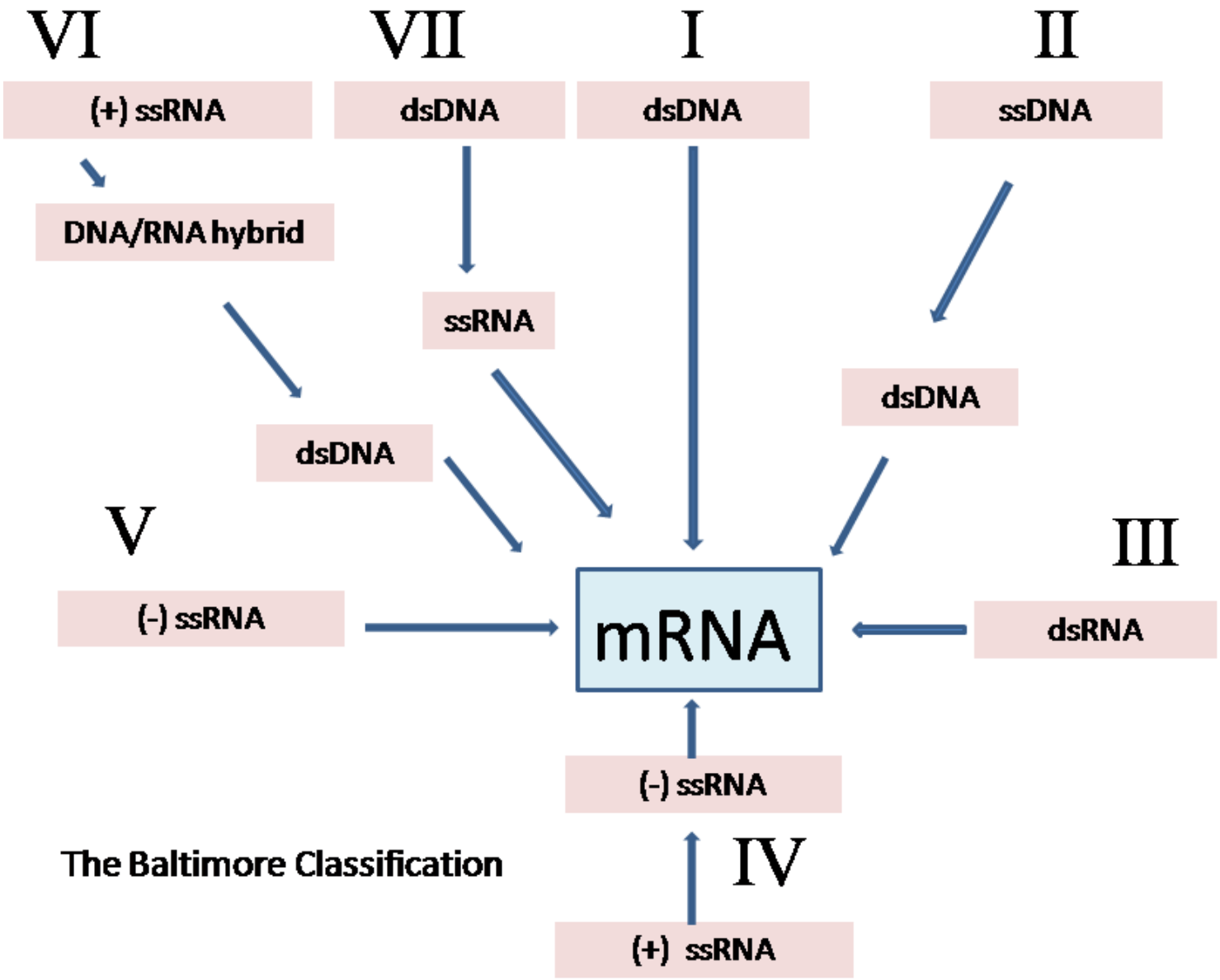
IV: **(+)ssRNA viruses** (+ strand or sense) RNA (e.g. Picornaviruses, Togaviruses)

V: **(–)ssRNA viruses** (− strand or antisense) RNA (e.g. Orthomyxoviruses, Rhabdoviruses)

VI: **ssRNA-RT viruses** (+ strand or sense) RNA with DNA intermediate in life-cycle (e.g. Retroviruses)

VII: **dsDNA-RT viruses** (e.g. Hepadnaviruses)

Baltimore classification



ICTV

= Internacional Comitee for Taxonomy of Viruses

- klasifikace se snaží sledovat evoluci virů

← klasifikace není kompletní (chybí mnoho taxonů, některé taxony nejsou zařazeny)

- sleduje klasickou taxonomickou nomenklaturu:

řád	order	*-virales*	*Herpesvirales*
čeleď	family	*-viridae*	*Herpesviridae*
podčeleď	subfamily	*-virinae*	*Alphaherpesvirinae*
rod	genus	*-virus*	*Simplexvirus*
druh	species	virus	Human herpesvirus 1
kmen/izolát	strain/isolate	???	HHV1 strain F

ICTV taxonomie

AKTUÁLNÍ STAV TAXONOMIE (k 22. 2. 2014)
-7 řádů, 96 čeledí, 22 podčeledí, 420 řádů, 2618 druhů

- Caudovirales – bakteriofágy s komplexní morfologií kapsidy
- Herpesvirales – velké živočišné oblané viry
- Ligamenvirales – dsDNA viry archaebakterií
- Mononegavirales - -ssRNA viry s nefragmentovaným genomem
- Nidovirales – živočišné +ssRNA viry
- Picornavirales – rostlinné i živočišné +ssRNA viry
- Tymovirales – obalené +ssRNA viry

dohromady 25 čeledí

71 čeledí není zařazeno k žádnému řádu !!!

ICTV nomenklatura

Virus = fyzická entita

= virový izolát, virový kmen

- teoreticky zařaditelné do všech výše zmíněných skupin (mnoho virů zůstává nezařazeno nebo se zařazení mění)

Virový druh = teoretická skupina virů

- každý druh viru musí být reprezentován alespoň jedním izolátem

Vytvoření, pojmenování i přejmenování každého taxonu musí schválit ICTV

Virový izolát není taxonomická jednotka a objevitel si ji může pojmenovat dle libosti

Pojmenování virových druhů

všechny mají anglická jména, nejsou latinská jména, někdy i česky

- podle nemoci, kterou způsobují
  - influenza virus (virus chřipky)
  - poliovirus (virus dětské obrny)
  - yellow fever virus (virus žluté zimnice)
- podle místa, kde se poprvé objevily
  - Ebola virus (Ebola je řeka v Kongu)
  - West Nile virus (poprvé izolován v provincii Západní Nil, Uganda)
  - Norwalk virus (poprvé izolován v Norwalku, Ohio, USA)
- podle hostitele
  - *Thosea asigna* virus, *Drosophila* C virus
- podle orgánu který postihuje
  - infectious bursal disease virus

Pojmenování virových izolátů

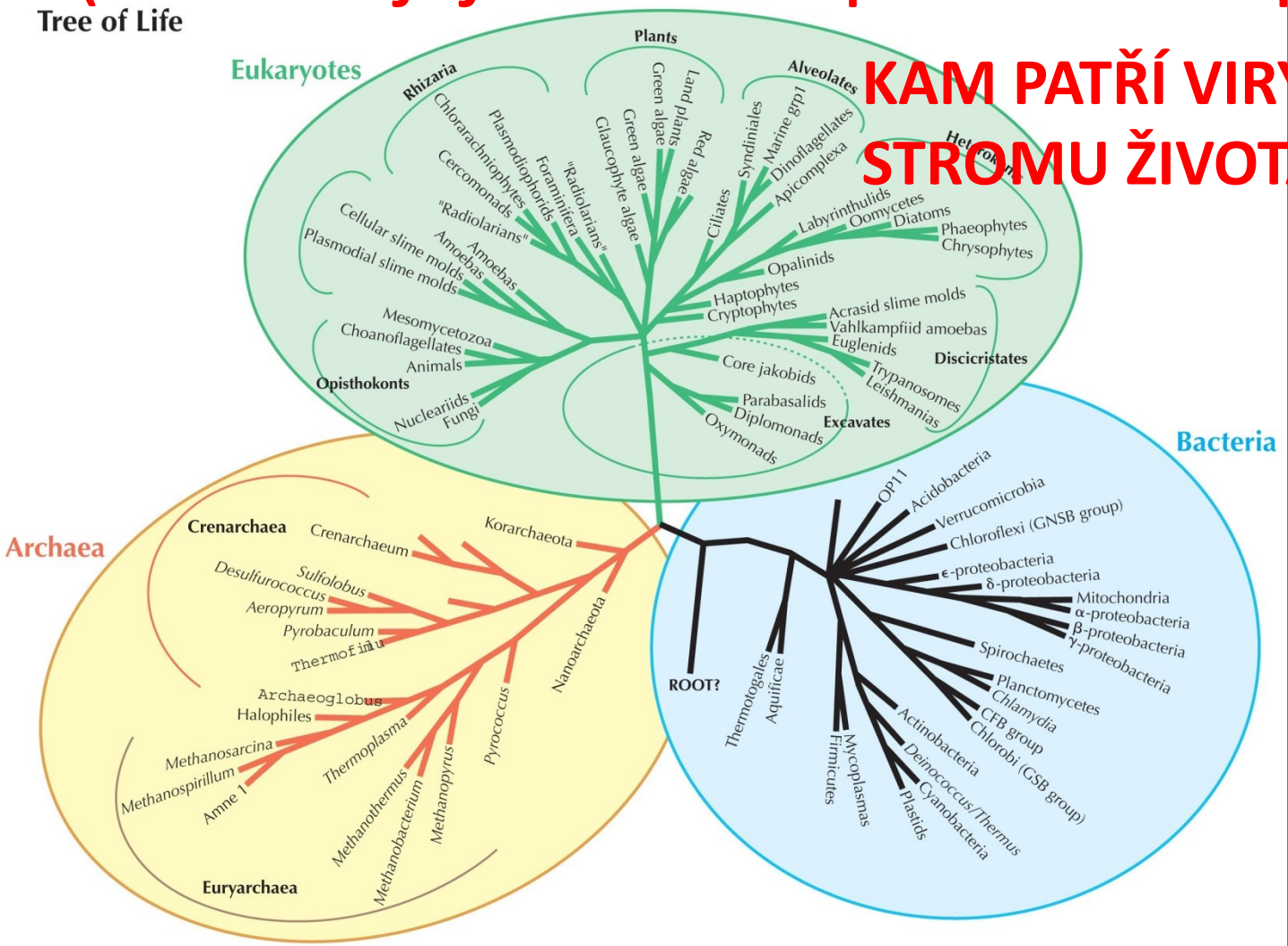
podle libovůle objevitele

- podle pacienta, ze kterého byl virus vyizolován
  - TBEV virus strain Hypr (kmen viru klíšťové encefalitidy izolovaný *post mortem* z mozku chlapce, který se jmenoval Hypr)

- podle místa, kde byl nalezen
  - TBEV strain Neudoerfel (kmen viru klíšťové encefalitidy objevený poblíž obce Neudoerfl, poblíž Wiener Neustadt)

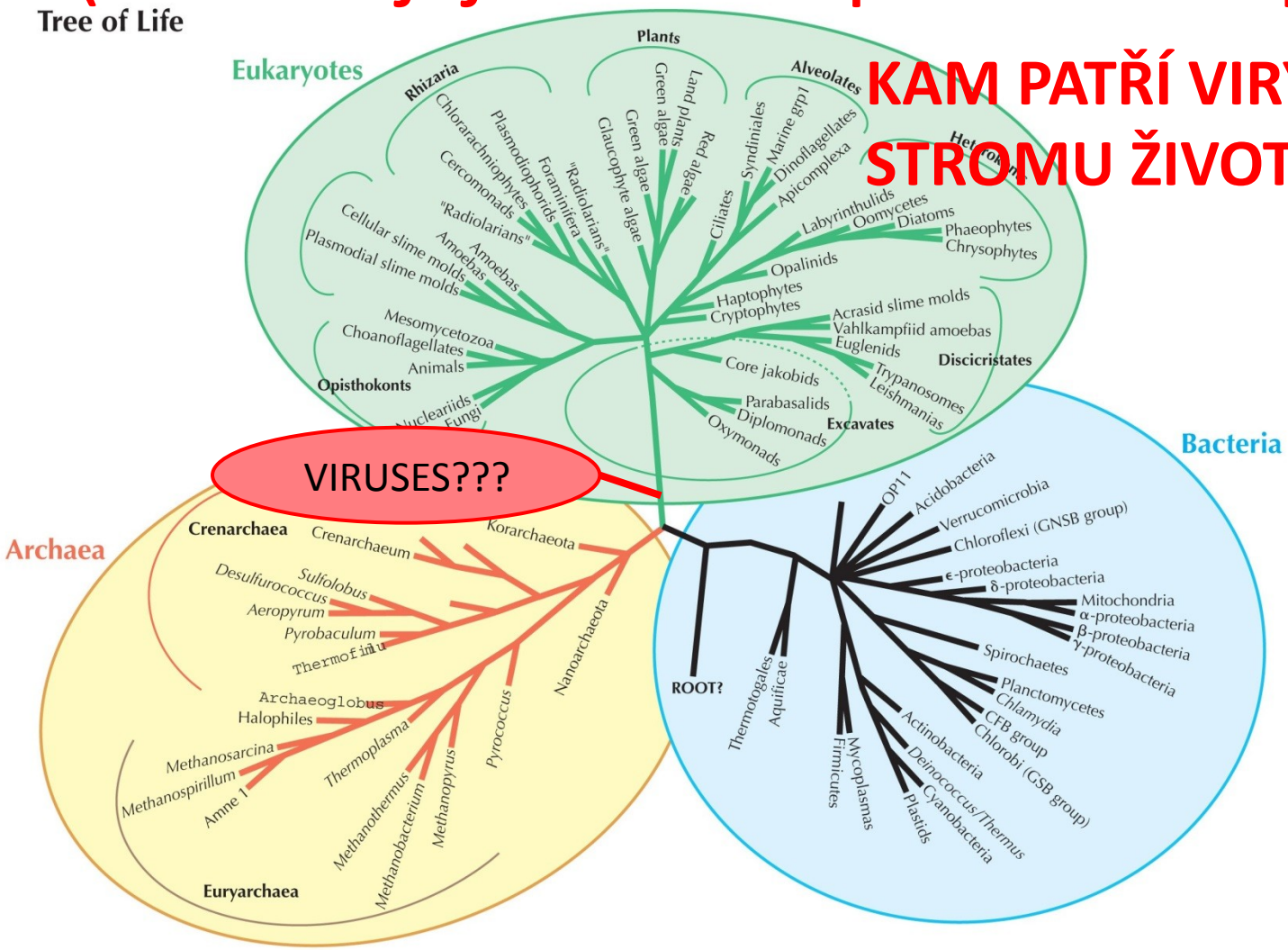
- podle nějakého systematického klíče, který je znám jen objeviteli
  - TBEV strain J49 (označení vzorku, ve kterém byl virus vyizolován –
  - zkumavka J49???)

Jsou viry monofyletická skupina? (tzn. Mají jednoho společného předka?)



KAM PATŘÍ VIRY NA STROMU ŽIVOTA???!!!

Jsou viry monofyletická skupina? (tzn. Mají jednoho společného předka?)



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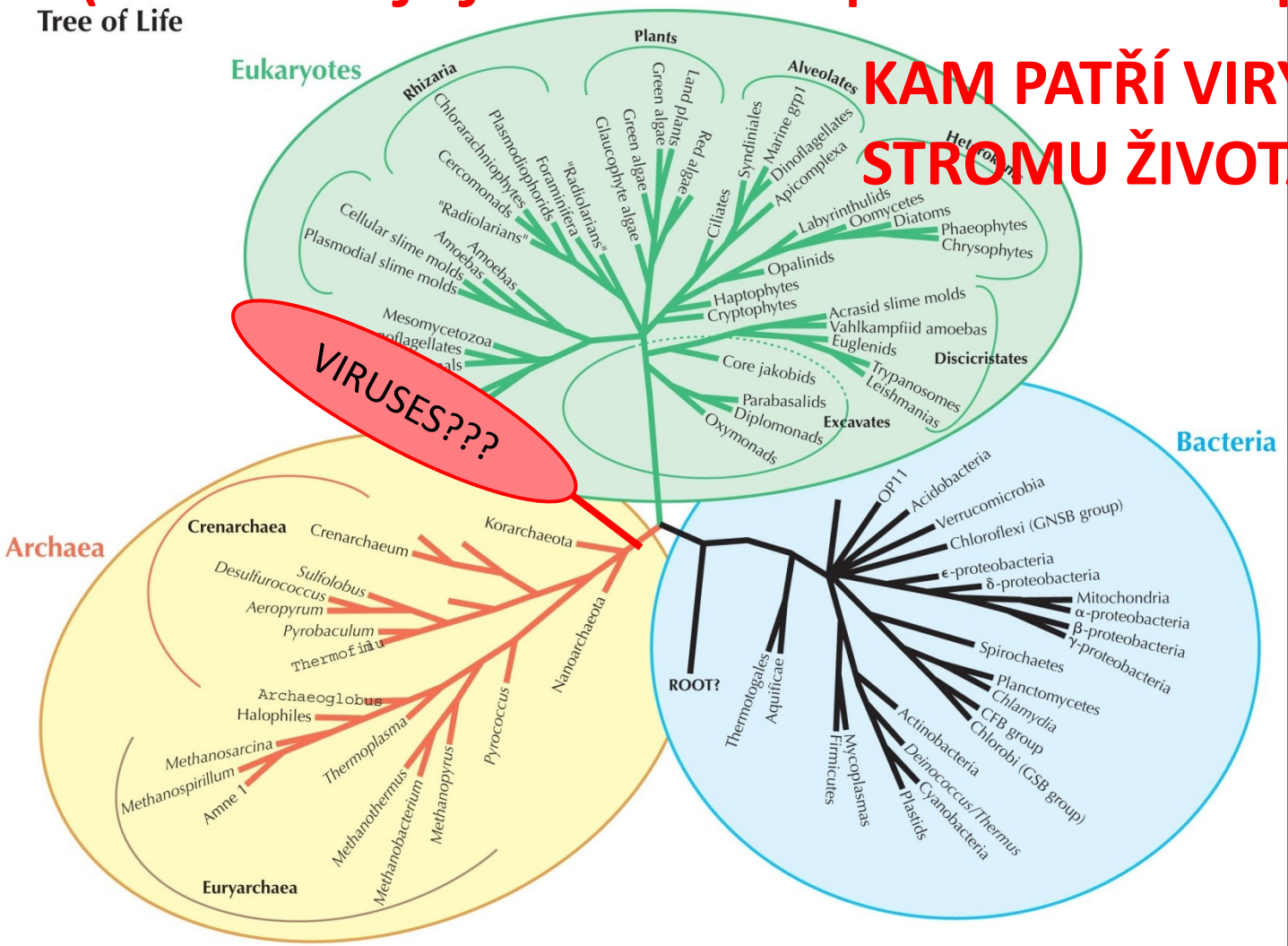

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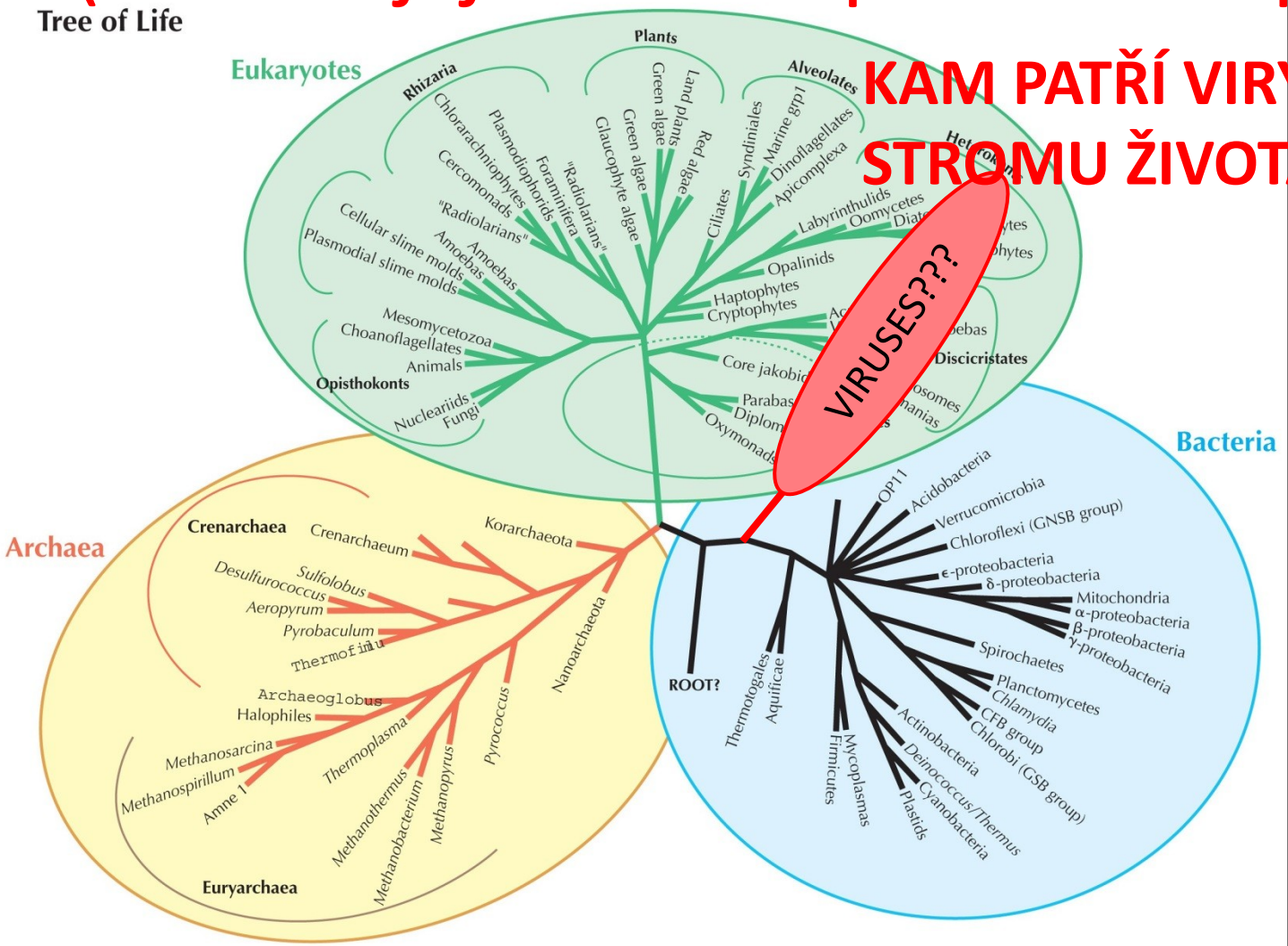

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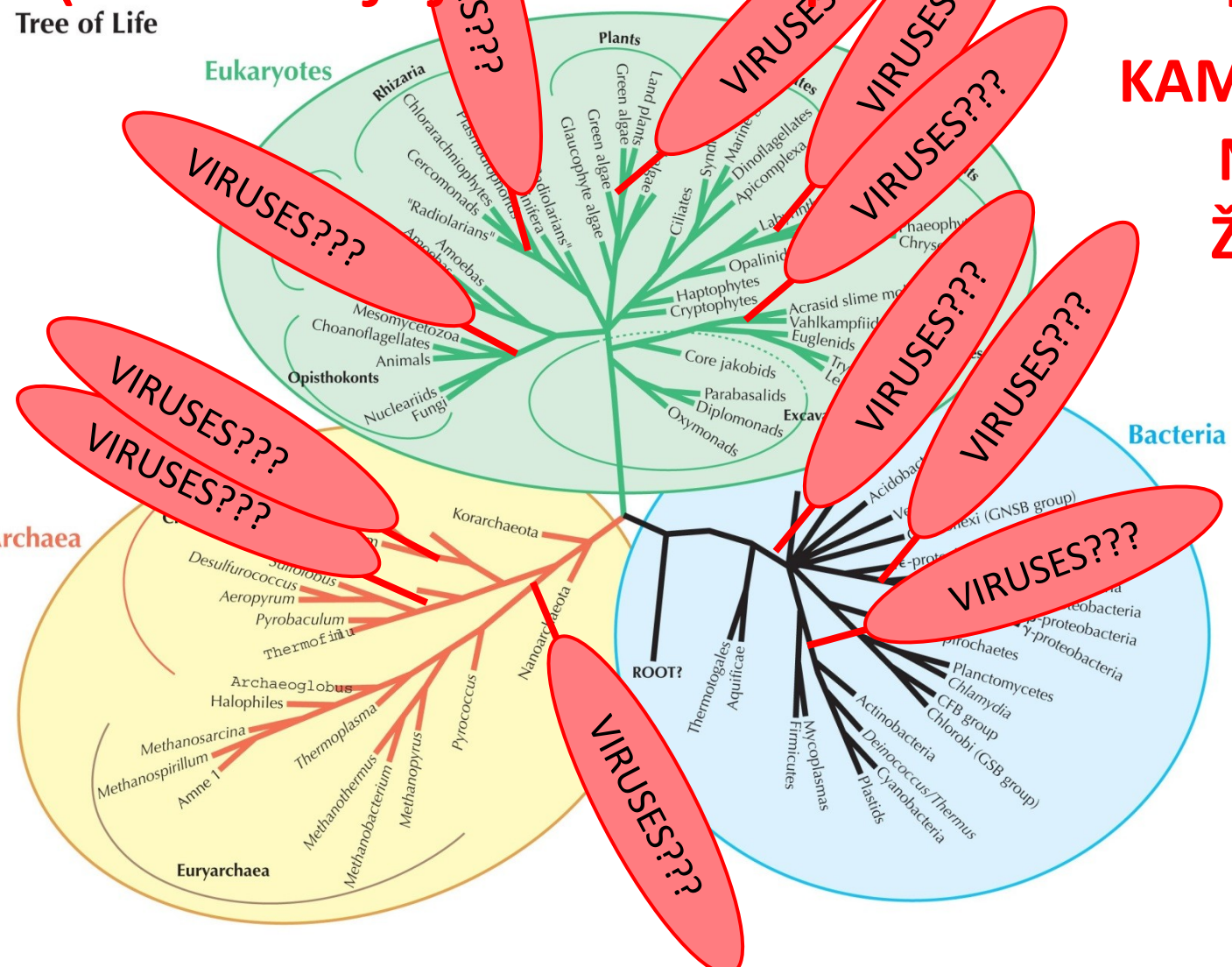
(tzn. Mají jednoho společného předka?)



KAM PATŘÍ VIRY NA STROMU ŽIVOTA???!!!

Jsou viry monofyletická skupina? (tzn. Mají jednoho společného předka?)

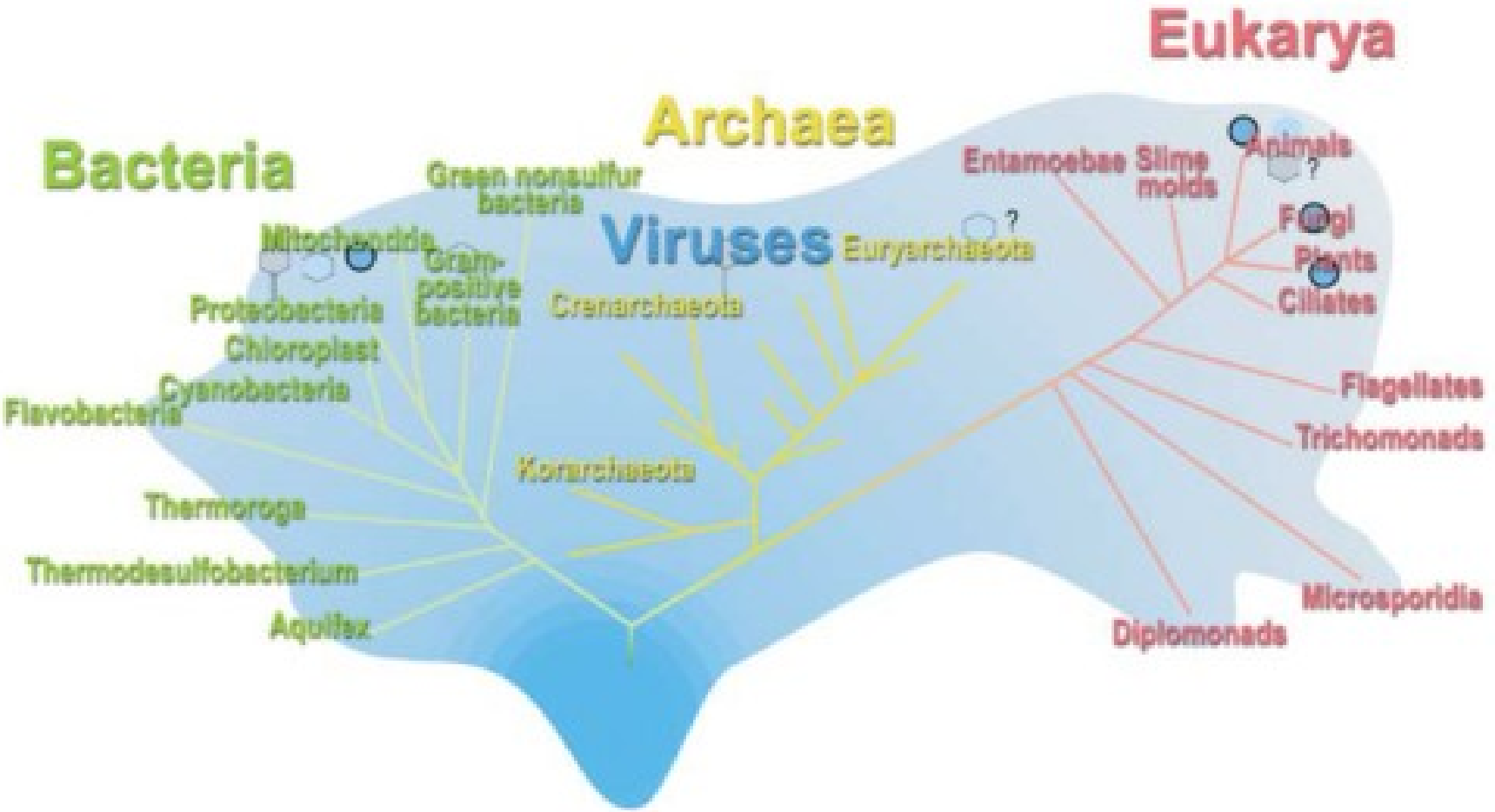
MONOFYLETICKÁ nebo POLYFYLETICKÁ skupina???

neví se přesně

spíše POLYFYLETICKÁ SKUPINA

Kam viry patří na stromu života?

Dennis H. Bamford – TEORIE VIROVÉHO OCEÁNU



Bamford, Burnett, Stuart (2002): Evolution of Viral Structure, Theoretical Population Biology

Jiří Černý

viral RdRPs
viral RdRPs
3avx (Qβ)
0,9999
0,9943
root???
0,9885
3v81 (HIV1)
viral RdDPs
1rw3 (Mo-MuLV)
0,9914
0,9997
1
0,5259
DdDPs
Family A
single
subunit
DdRPs
1tk0 (T7, DdDP)
2pi4 (T7, DdRP)
2hhu (B. stearothermophilus,
DdDP - Klenow fragment)
0,8655
0,5072
0,9838
0,9596
1jx4 (S.
sulfataricus)
0,6920
1q8i (E. coli, DdDP
- DNA pol II)
2vwj (T. gorgonarius)
1t94 (H. sappiens,
DNA pol κ)
1zet (H. sappiens,
DNA pol ι)
2r8j (S. cerevisiae,
DNA pol η)
DdDPs
Family Y
2pyj (Φ29)
DdDPs
Family B

Evoluce virů

- rychlá (error prone polymerázy, silný evoluční tlak)

- chybí univerzální marker gene (kapsida, polymerázy)
  ← jeden gen je sdílen maximálně v několika čeledích
  ➔ rekonstrukce evoluce nad úroveň čeledí je velmi obtížná

- používá se porovnání mnoha znaků (replikační strategie, architektura kapsidy…)
  ➔ velmi náchylné k artefaktům způsobeným rekombinací

➔ EVOLUCE VIRŮ JE CHÁPÁNA SPÍŠ JAKO REKOMBINAČNÍ SÍŤ NEŽ JAKO FYLOGENETICKÝ STOM

Evoluce virů

- 10 000 000 virů na 1 ml oceánské vody
  = 1 000 000 000 000 000 000 000 000 000 000 (0)
  = 10^{30}- 10^{31} virů v oceánech

  →viry oceánů představují cca 200Mt uhlíku to je stejně jako 75 milionům velryb (počítá li se, že C je 10% jejich váhy)

  → pokud by se naskládaly bakterofágy vedle sebe hlava-ocas vytvoří dráhu cca 100 x větší než představuje vzdálenost napříč naší galaxií

Evoluce proteinů

- 22 aminokyselin (včetně selenocysteinu a pyrrolysinu)
- 1000 AA dlouhý protein má 1000^{22} variant = 10^{24}

←10^{30}- 10^{31} virů v oceánech

→VIRY JSOU HRAVĚ SCHOPNÉ PROZKOUŠET CELÝ PROTEIN UNIVERSE

Koevoluce virů a hostitelů

- teorie červené královny

  viry (a ostatní patogeny) jsou hybnou silou evoluce a hlavním důvodem vzniku a udržení sexuálního rozmnožování

- viry stojí za mnoha důležitými evolučními událostmi
  - vznik života???
  - přechod od RNA světa k DNA světu
  - vznik buněčného jádra
  - vznik a evoluce savců (sycytin)

- endogenní proviry jsou skvělý marker pro studium evoluce hostitelů

Quasispecies

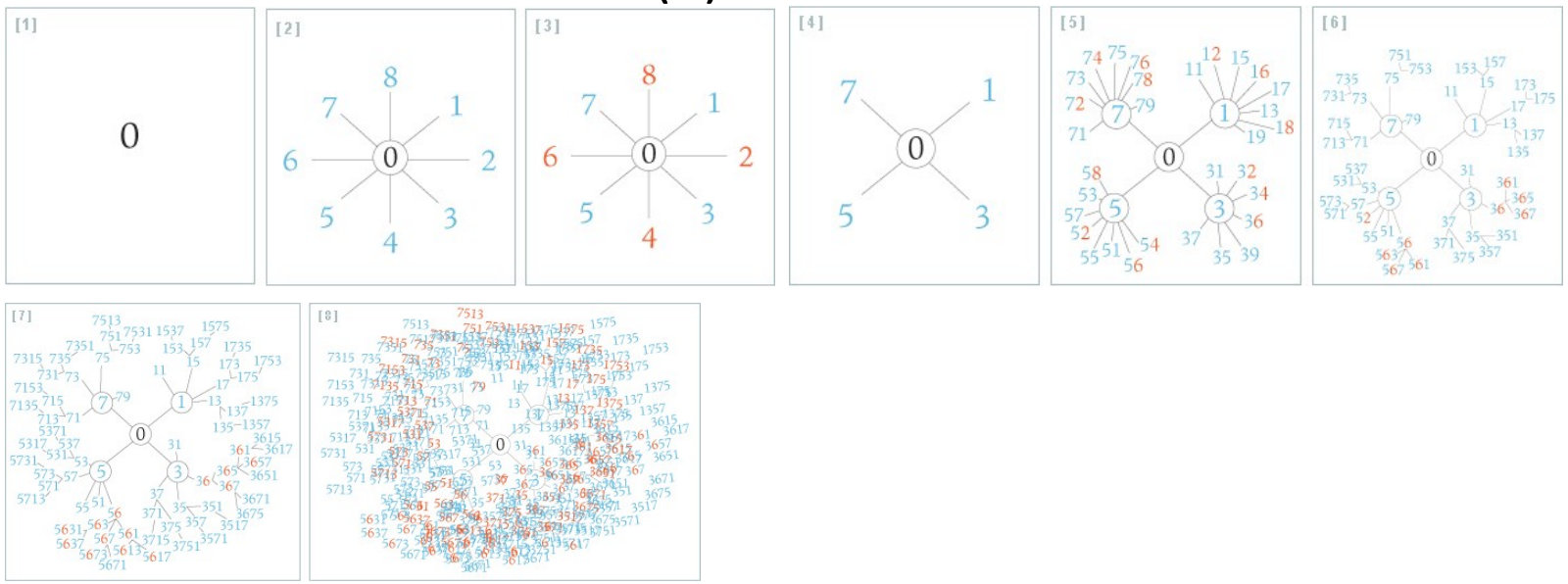
- pokud dochází k replikaci nukleové kyseliny polymerázou s velkou pravděpodobností vzniku mutace není populace soubor jednoho klonu ale směsí mnoha různých variant

- RNA viry mají velmi vysokou pravděpodobnost vzniku mutací
  → existují quasispecies

- DNA viry mají nízkou pravděpodobnost vzniku mutací
  → neexistují quasispecies

Quasispecies - example

- two dimensional map, where any point on the map represents a different viral variant
- one single original genome – 0 (1)
- 8 nucleotide positions on this genome can vary, after a single round of replication, all eight mutants are generated (2)
- some mutants are nonviable (in red) (3)
- after 4 rounds (7)
- there are 4 nucleotides!!! (8)



Quasispecies - důležitost

- únik před hostitelským imunitním systémem

- rychlé získání rezistence proti antivirotikům

Hybná síla evoluce virů

1) Funkčnost virových proteinů (pozitivní mutace)

→evoluce vybírá funčnější varianty

	1 generace	3 generace	10 generací
virus A	50	125 000	10^{17}
virus B	100	1 000 000	10^{20}

2) Imunitní systém hostitele (neutrální mutace)

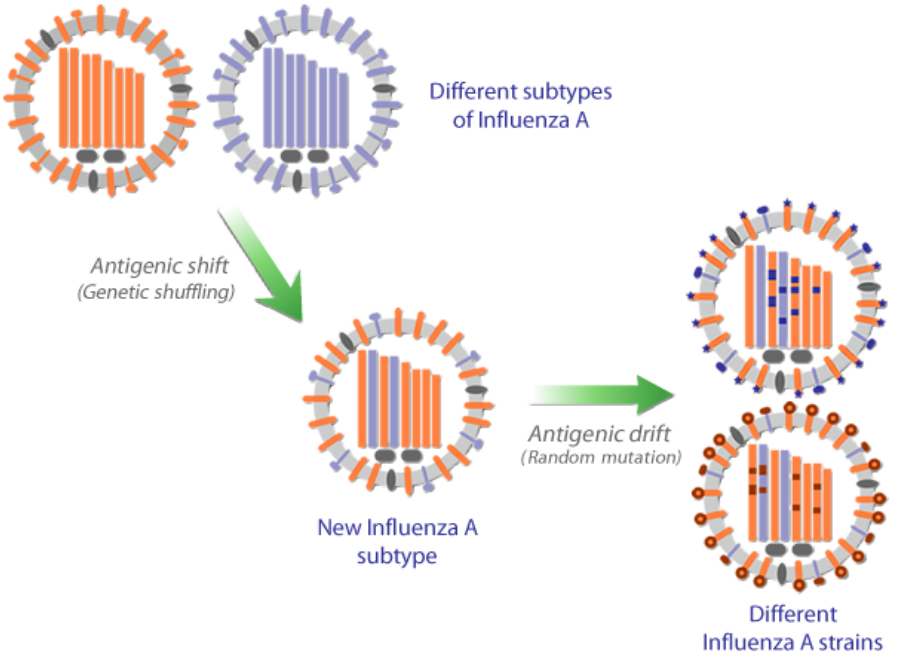
- silná protilátková odpověď proti známým antigenům

→ nové antigeny mají výhodu opožděné a slabší reakce imunitního systému (může dokonce převážit i mírně sníženou funkčnost mutantů)

drift x shift

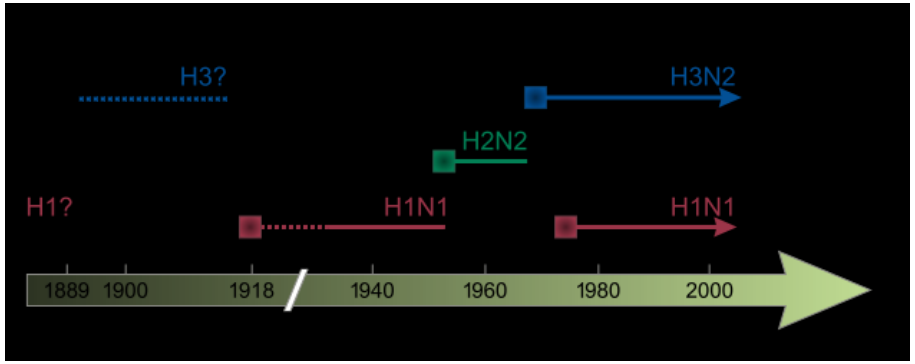
Drift = mutace

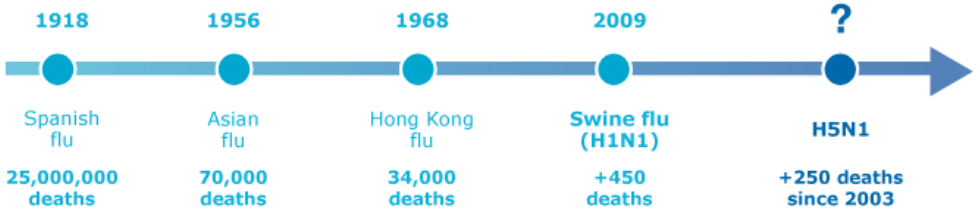
- postupné hromadění mutací
- pomalé antigenní změny
- všechny viry



Shift = rekombinace

- rekombinace mezi virovými různými kmeny
- rychlá a velmi výrazná změna v genotypu viru
- viry s fragmentovaným genomem (chřipa → pandemie)





vymetení x koexistence

vymetení

- nový kmen viru v populaci kompletně nahradí starý kmen
- u vysoce infekčních virů (chřipka typ A)

koexistence

- v jednom časovém úseky koexistuje více kmenů (odděleny místně, neodděleny)
- u méně infekčních virů, u virů s širokým rezervoárem hostitelů

Hostitelská bariéra

Viry s úzkým okruhem hostitelů

(virus černých neštovic, cytomegalovirus, HCV, HIV)

- často specifické jen pro jeden druh
- dlouhá koevoluce s hostitelem (ne HIV)
- většinou způsobují perzistentní infekci a akutní infekce nebývají většinou příliš vážné (neplatí pro černé neštovice a jen částečně pro HCV)
- teoreticky jsou zlikvidovat (viz. černé neštovice)

Viry se širokým okruhem hostitelů

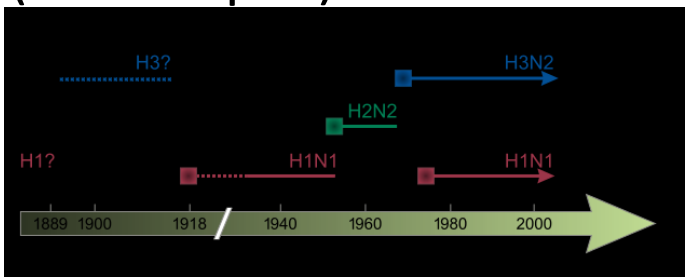
(chřipka, West Nike virus, …)

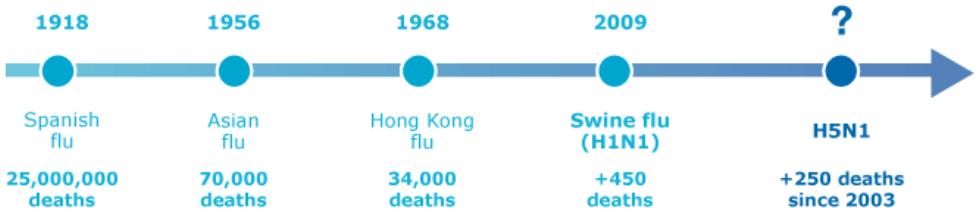
- často způsobují závažné akutní infekce
- nebezpečné zejména při přechodu mezi hostitely

Překonání hostitelské bariéry

- nutné mnoho změn v genomu
  → rozpoznání jiných receptorů, interakce s rozdílnými buněčnými proteiny

1) překonání hostitelské bariéry
2) virus se ocitá imunologicky naivní populaci
3) virus vyvolá silnou epidemii
4) populace se viru přizpůsobí (citlivý jedinci zemřou, zbytek získá protilátky)
5) Virus se populací šíří pomalu a nevyvolává velké epidemie (viz. chřipka)

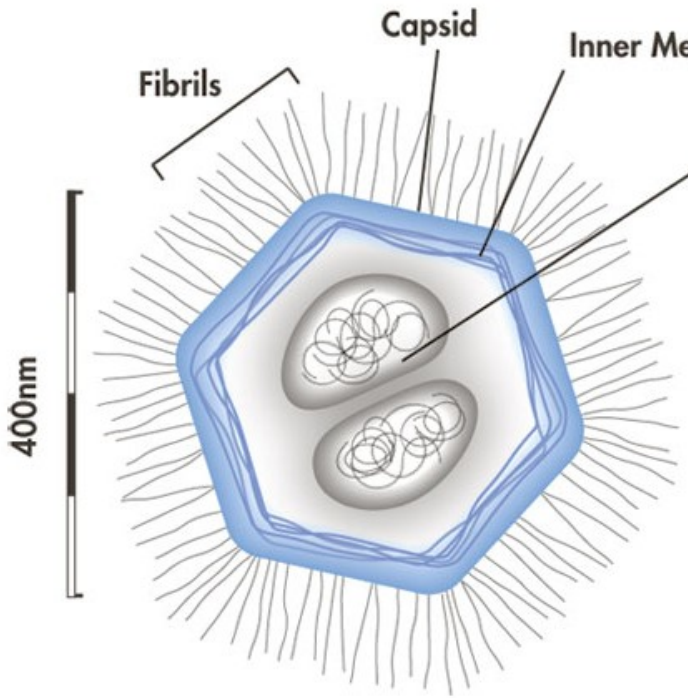




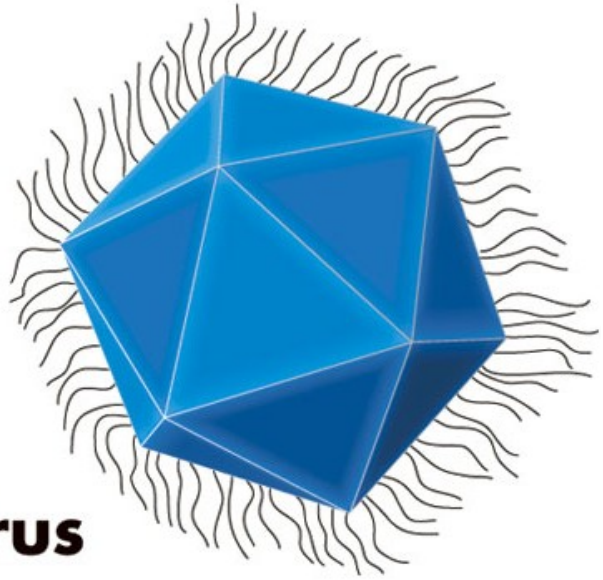
2003 • Bernard La Scola, Didier Raoult, others • discovery of mimivirus, at the time the largest virus known

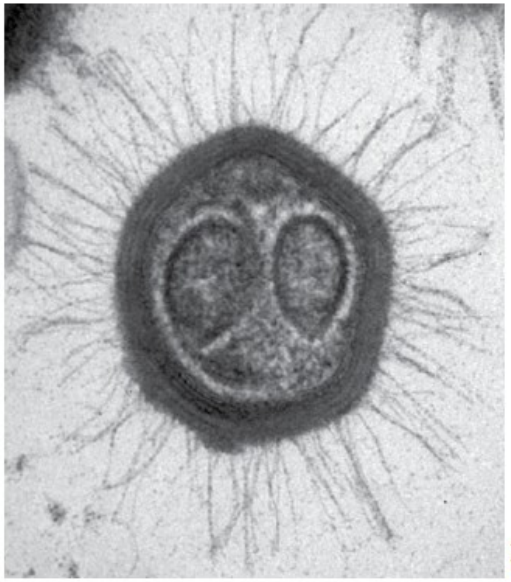
Bernard B. La Scola

Didier Raoult



- dsDNA virus
- 90% coding capacity
- 10% Junk DNA
- 1.2 million base pairs
- ~911 protein coding genes
- additional genes (inc. aminoactyl tRNA synthetases; sugar, lipid, and amino acid metabolism)

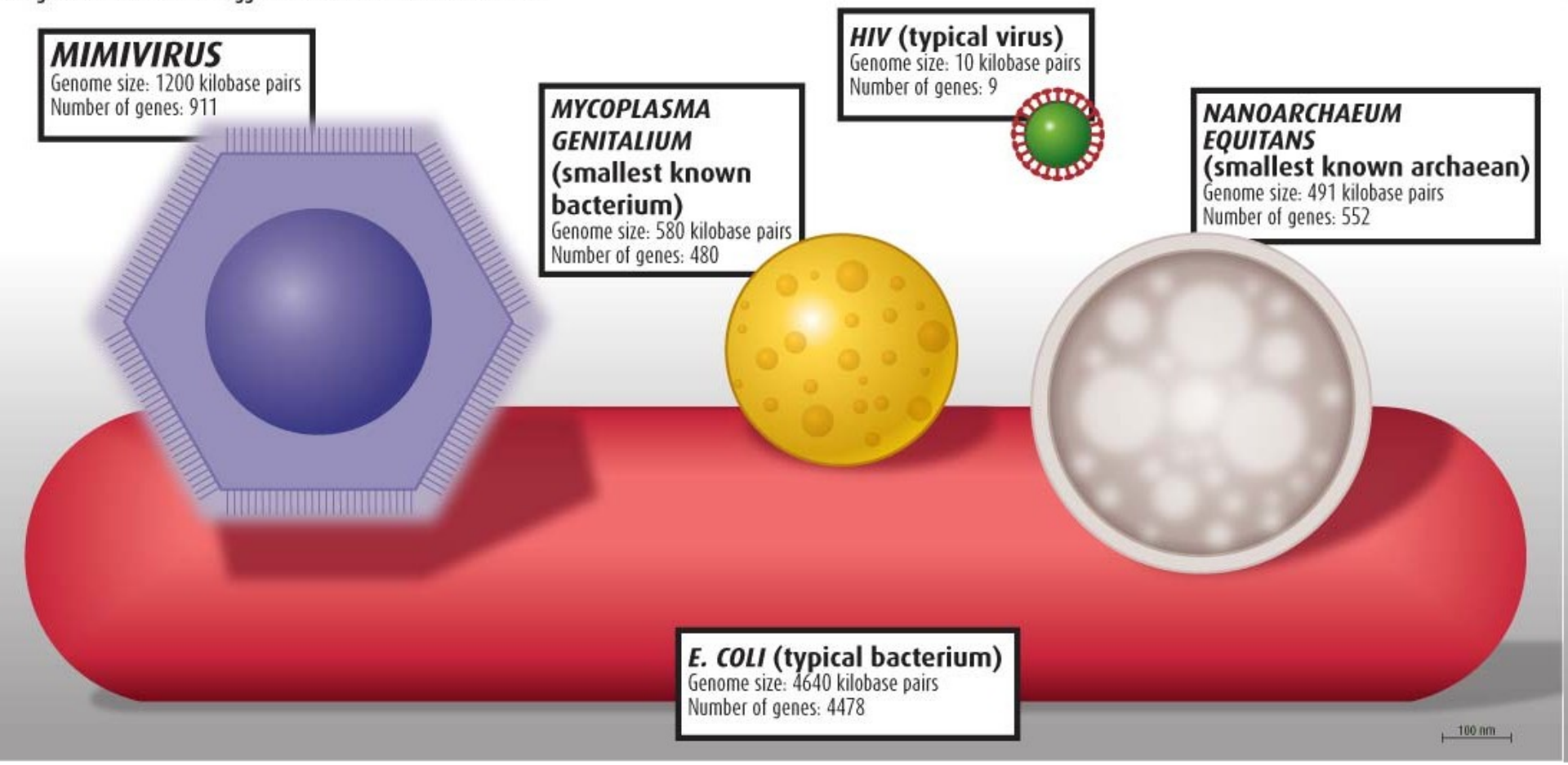


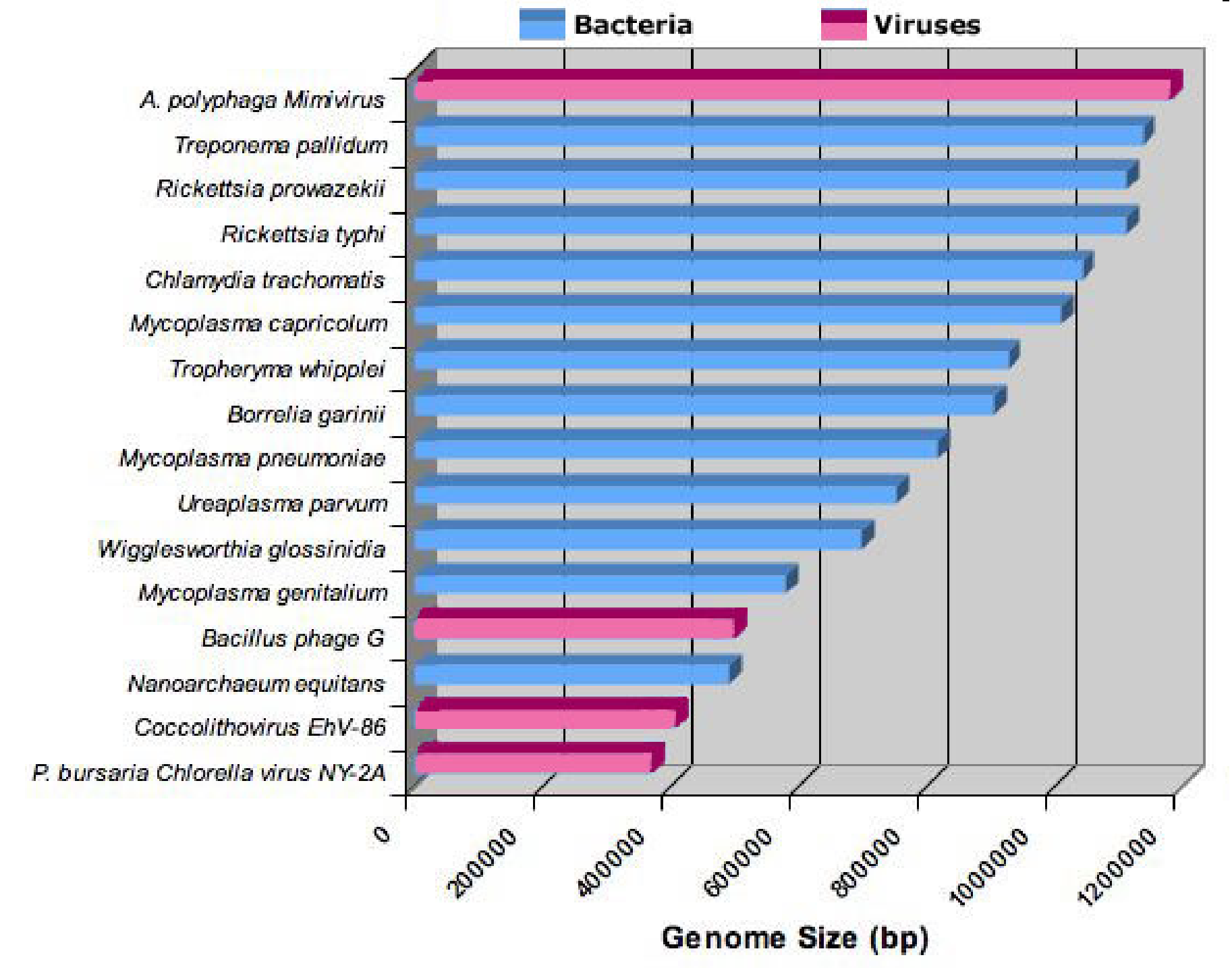


acanthamoeba polyphaga mimivirus

MONSTER MICROBE

The giant virus Mimivirus is bigger than some bacteria and archaeans

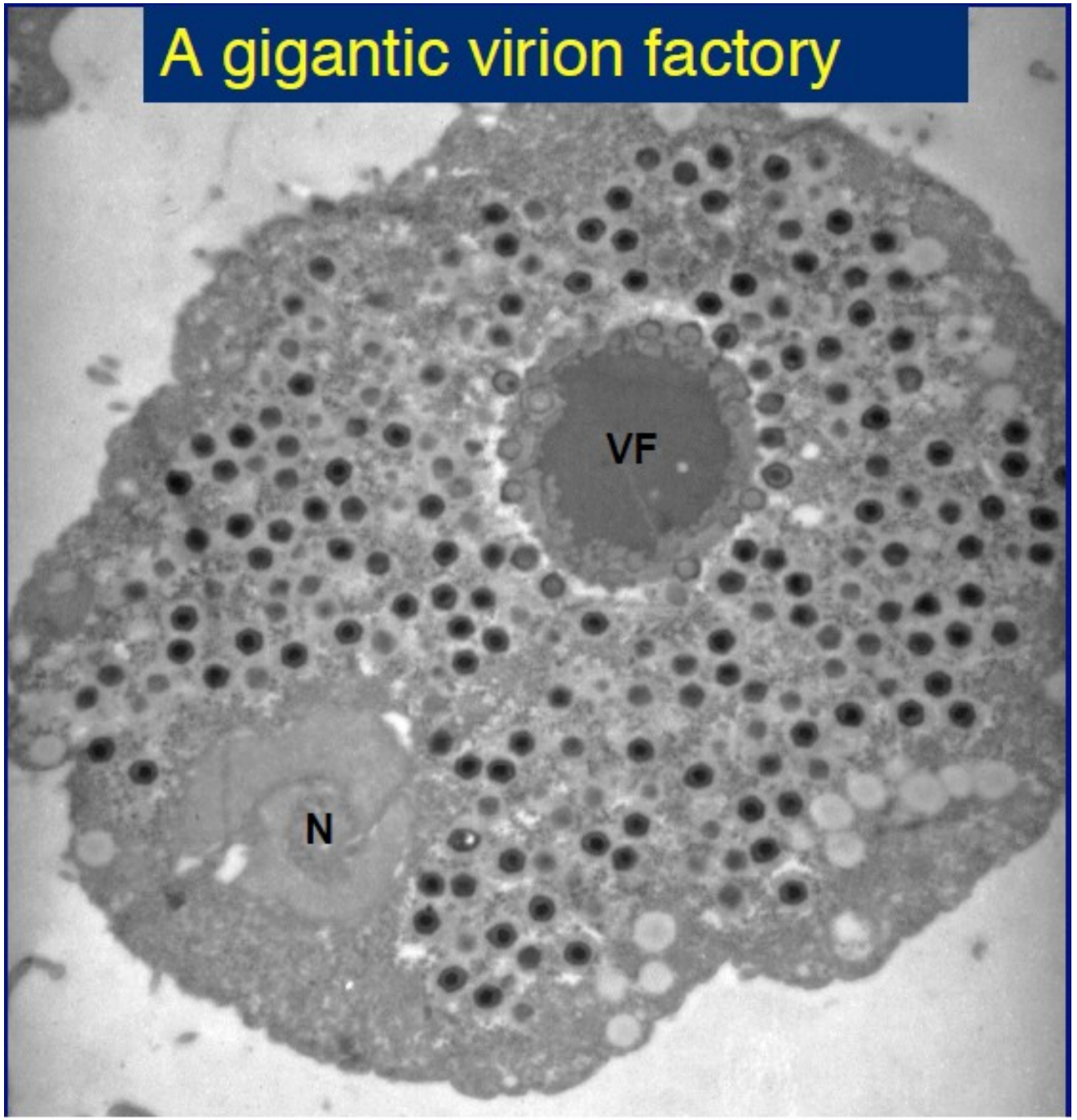



Bacteria
Viruses
A. polyphaga Mimivirus
Treponema pallidum
Rickettsia prowazekii
Rickettsia typhi
Chlamydia trachomatis
Mycoplasma capricolum
Tropheryma whipplei
Borrelia garinii
Mycoplasma pneumoniae
Ureaplasma parvum
Wigglesworthia glossinidia
Mycoplasma genitalium
Bacillus phage G
Nanoarchaeum equitans
Coccolithovirus EhV-86
P. bursaria Chlorella virus NY-2A
0
200000
400000
600000
800000
1000000
1200000
Genome Size (bp)

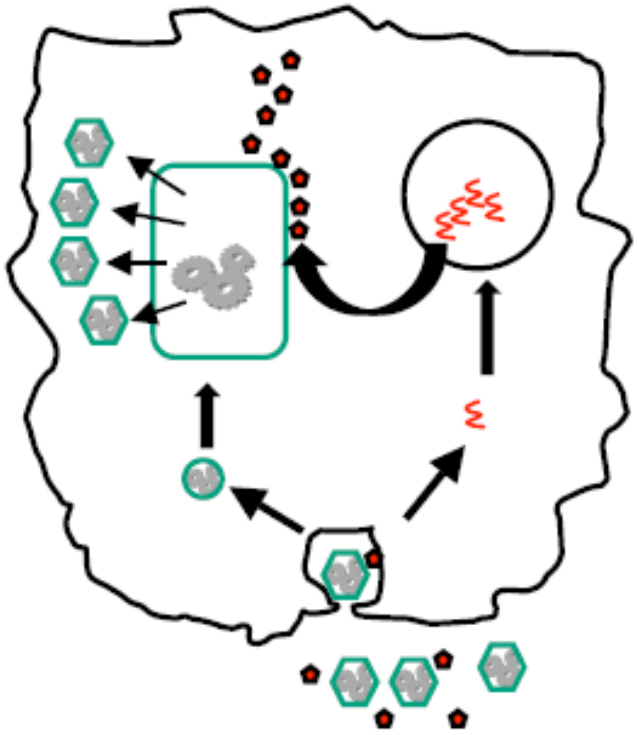
The next big surprise: remnants of a translation apparatus

ORF #	Function	Comment
R663	**Arginyl-tRNA synthetase**	Translation
L124	**Tyrosyl-tRNA synthetase**	Translation
L164	**Cysteinyl-tRNA synthetase**	Translation
R639	**Methyonyl tRNA synthetase**	Translation
R726	**Peptide chain release factor eRF1**	Translation END
R624	GTP binding elongation factor eF-Tu	Translation ELONG
R464	Translation initiation factor SUI1	Translation START
L496	Translation initiation factor 4E (mRNA cap binding)	Translation START
R405	tRNA (Uracil-5-)-methyltransferase	tRNA modification

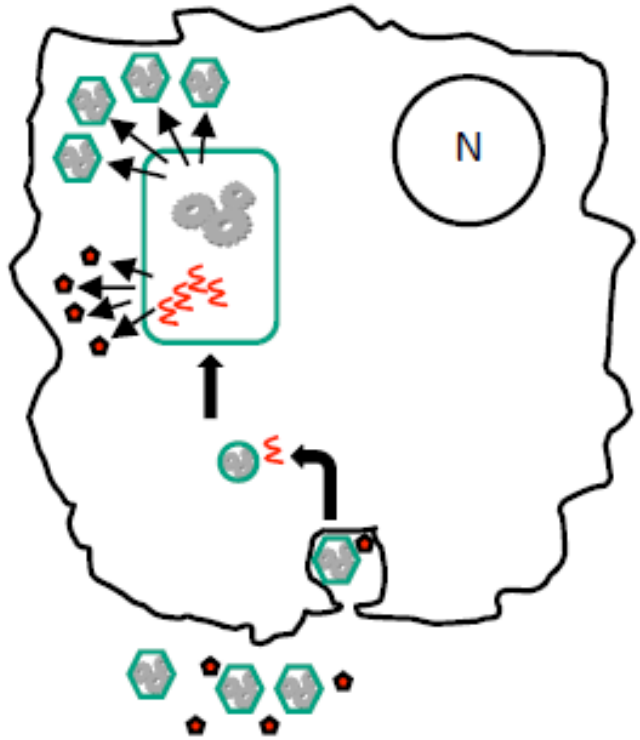
+ 6 tRNA: 3 Leu, Trp, His, Cys


A gigantic virion factory
VF
N

Satellite *versus* « virophage »



Traditional satellite virus



« Virophage »

LETTERS

The virophage as a unique parasite of the giant mimivirus

Bernard La Scola[1]*, Christelle Desnues[1]*, Isabelle Pagnier[1], Catherine Robert[1], Lina Barrassi[1], Ghislain Fournous[1], Michèle Merchat[2], Marie Suzan-Monti[1], Patrick Forterre[3,4], Eugene Koonin[5] & Didier Raoult[1]

Viruses are obligate parasites of Eukarya, Archaea and Bacteria. *Acanthamoeba polyphaga* mimivirus (APMV) is the largest known virus; it grows only in amoeba and is visible under the optical microscope. Mimivirus possesses a 1,185-kilobase double-stranded linear chromosome whose coding capacity is greater than that of numerous bacteria and archaea[1–3]. Here we describe an icosahedral small virus, Sputnik, 50 nm in size, found associated with a new strain of APMV. Sputnik cannot multiply in *Acanthamoeba castellanii* but grows rapidly, after an eclipse phase, in the giant virus factory found in amoebae co-infected with APMV[4]. Sputnik growth is deleterious to APMV and results in the production of abortive forms and abnormal capsid assembly of the host virus. The Sputnik genome is an 18,343-kilobase circular double-stranded DNA and contains genes that are linked to viruses infecting each of the three domains of life Eukarya, Archaea and Bacteria. Of the 21 predicted protein-coding genes, eight encode proteins with detectable homologues, including three proteins apparently derived from APMV, a homologue of an archaeal virus integrase, a predicted primase–helicase, a packaging ATPase with homologues in bacteriophages and eukaryotic viruses, a distant homologue of bacterial insertion sequence transposase DNA-binding subunit, and a Zn-ribbon protein. The closest homologues of the last four of these proteins were detected in the Global Ocean Survey environmental data set[5], suggesting that Sputnik represents a currently unknown family of viruses. Considering its functional analogy with bacteriophages, we classify this virus as a virophage. The virophage could be a vehicle mediating lateral gene transfer between giant viruses.

The original strain of APMV, mimivirus, was obtained from a cooling tower in Bradford, UK. Its size challenged the definition of a virus[6] and led to the idea that giant viruses might be an uncharacterized but important part of the biosphere. We isolated a new strain of APMV, by inoculating *A. polyphaga* with water from a cooling tower, in Paris. We denoted this new strain mamavirus because it seemed to be even larger than mimivirus[7] when observed by transmission electron microscopy. The main features of mamavirus closely resembled those described for mimivirus, including the formation of a giant viral factory and the typical particle morphology with a multilayered membrane covered with fibrils[6]. We also observed unknown icosahedral small viral particles, 50 nm in size, in virus factories and in the cytoplasm of the infected cells (Fig. 1). Considering the association of this newly detected virus with mamavirus, we named it Sputnik.

Sputnik did not multiply when inoculated into *A. castellanii* (Supplementary Information and Supplementary Table 4). However, this virus did grow, as demonstrated by transmission electron microscopy and polymerase chain reaction, in *A. castellanii* co-infected with mimivirus or mamavirus (Supplementary Information

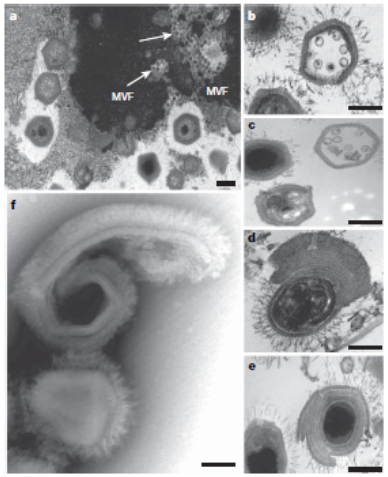


Figure 1 | Different morphological aspects of mamavirus and Sputnik. **a**–**e**, Observations by transmission electron microscopy; **f**, observation by negative staining electron microscopy. **a**, Mamavirus virus factory (MVF) with mamavirus particles at different stages of maturation. Clumps of Sputnik particles (arrows) are observed within MVF. **b**, In some cases, Sputnik is observed within mamavirus capsids. **c**, Defective particles are produced. **d**–**f**, Co-infection with mamavirus and Sputnik results in abnormal morphology of mamavirus particles, such as membrane accumulation at one side (**d**), membrane accumulation around the particles (**e**), or open particles (**f**). Scale bars, 200 nm.

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*These authors contributed equally to this work.

„Virus je kapsid-kódující organismus, který se skládá z proteinů a nukleových kyselin, je schopen samovolného složení svého nukleokapsidu a využívá organismy kódující ribozómy pro dovršení svého replikačního cyklu.“

BADVIBRATIONS
Navy report admits sonar's impact on whales.
www.nature.com/news

PUNCHSTOCK

'Virophage' suggests viruses are alive

The discovery of a giant virus that falls ill through infection by another virus[1] is fuelling the debate about whether viruses are alive.

"There's no doubt this is a living organism," says Jean-Michel Claverie, a virologist at the CNRS UPR laboratories in Marseilles, part of France's basic-research agency. "The fact that it can get sick makes it more alive."

Giant viruses have been captivating virologists since 2003, when a team led by Claverie and Didier Raoult at CNRS UMR, also in Marseilles, reported the discovery of the first monster[2]. The virus had been isolated more than a decade earlier in amoebae from a cooling tower in Bradford, UK, but was initially mistaken for a bacterium because of its size, and was relegated to the freezer.

Closer inspection showed the microbe to be a huge virus with, as later work revealed, a genome harbouring more than 900 protein-coding genes[3] — at least three times more than that of the biggest previously known viruses and bigger than that of some bacteria. It was named *Acanthamoeba polyphaga* mimivirus (for mimicking microbe), and is thought to be part of a much larger family. "It was the cause of great excitement in virology," says Eugene Koonin at the National Center for Biotechnology Information in Bethesda, Maryland. "It crossed the imaginary boundary between viruses and cellular organisms."

Now Raoult, Koonin and their colleagues report the isolation of a new strain of giant virus from a cooling tower in Paris, which they have named mamavirus because it seemed slightly larger than mimivirus. Their electron microscopy studies also revealed a second, small virus closely associated with mamavirus that has earned the name Sputnik, after the first man-made satellite.

With just 21 genes, Sputnik is tiny compared with its mama — but insidious. When the giant mamavirus infects an amoeba, it uses its large array of genes to build a 'viral factory', a hub where new viral particles are made. Sputnik infects this viral factory and seems to hijack its machinery in order to replicate. The team found that cells co-infected with Sputnik produce fewer and often deformed mamavirus particles, making the virus less infective. This suggests that Sputnik is effectively a viral parasite that sickens its host — seemingly the first such example.

The team suggests that Sputnik is a 'virophage', much like the bacteriophage viruses that infect and sicken bacteria. "It infects this factory like a phage infects a bacterium," Koonin says. "It's doing what every parasite can — exploiting its host for its own replication."

Sputnik's genome reveals further insight into its biology. Although 13 of its genes show little similarity to any other known genes, three are closely related to mimivirus and mamavirus genes, perhaps cannibalized by the tiny virus as it packaged up particles sometime in its history. This suggests that the satellite virus could perform horizontal gene transfer between viruses — paralleling the way that bacteriophages ferry genes between bacteria.

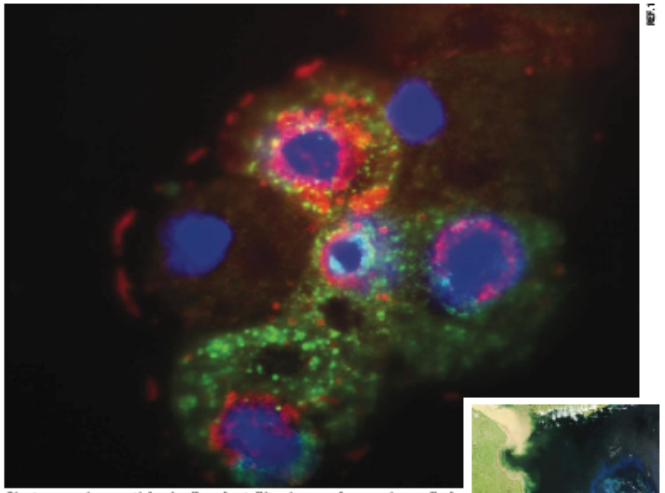
> "There's no doubt that this is a living organism. The fact that it can get sick makes it more alive."

The findings may have global implications, according to some virologists. A metagenomic study of ocean water[4] has revealed an abundance of genetic sequences closely related to giant viruses, leading to a suspicion that they are a common parasite of plankton. These viruses had been missed for many years, Claverie says, because the filters used to remove bacteria screened out giant viruses as well. Raoult's team also found genes related to Sputnik's in an ocean-sampling data set, so this could be the first of a new, common family of viruses. "It suggests there are other representatives of this viral family out there in the environment," Koonin says.

By regulating the growth and death of plankton, giant viruses — and satellite viruses such as Sputnik — could be having major effects on ocean nutrient cycles and climate. "These viruses could be major players in global systems," says Curtis Suttle, an expert in marine viruses at the University of British Columbia in Vancouver.

"I think ultimately we will find a huge number of novel viruses in the ocean and other places," Suttle says — 70% of viral genes identified in ocean surveys have never been seen before. "It emphasizes how little is known about these organisms — and I use that term deliberately." ■

Helen Pearson



REF. 1

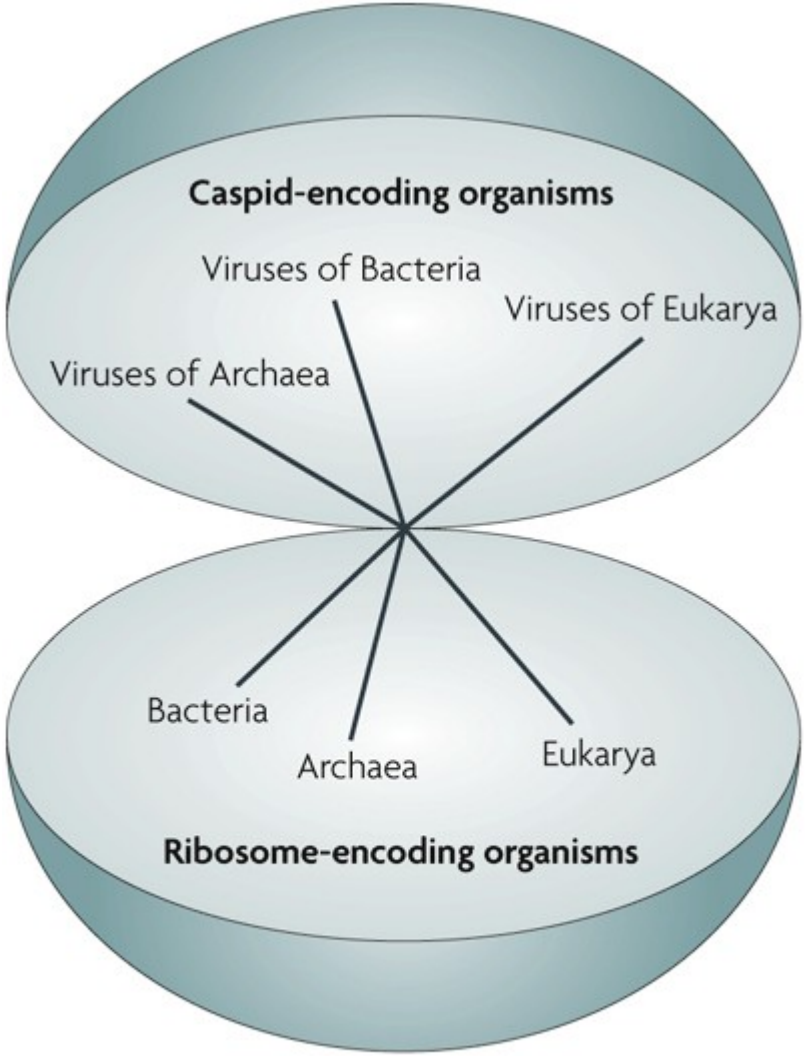
T. SCHMALTZ/NASA

Giant mamavirus particles (red) and satellite viruses of mamavirus called Sputnik (green). Virophages may be common in plankton blooms (inset).

1. La Scola, B. *et al. Nature* doi:10.1038/nature07218 (2008).
2. La Scola, B. *et al. Science* **299**, 2033 (2003).
3. Raoult, D. *et al. Science* **306**, 1344–1350 (2004).
4. Monier, A., Claverie, J.-M. & Ogata, H. *Genome Biol.* **9**, R106 (2008).

Status of Lwoff's criteria (2013)

- 1. ~~***Viruses contain a single type of nucleic acid***~~: **dismissed**.
- 2. ***Viruses are reproduced from their nucleic-acid only***: **dismissed**. The core of large DNA virus particles is an elaborate assembly of functional systems in continuity with the growing virion factory.
- 3. ***Viruses lack an energy producing system in contrast to cells***: **dismissed**. Highly reduced parasitic cells also might entirely rely on their host for ATP.
- 4. ***There is no binary fission in viruses***: this simple mechanistic criterion remains the best discriminant property, but is not useful at the genomic level because some parasitic cells do not exhibit the corresponding genes.



Nature Reviews | Microbiology

The Great Billion-year War between Ribosome- and Capsid-encoding Organisms (Cells and Viruses) as the Major Source of Evolutionary Novelties

Patrick Forterre[a,b] and David Prangishvili[a]

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