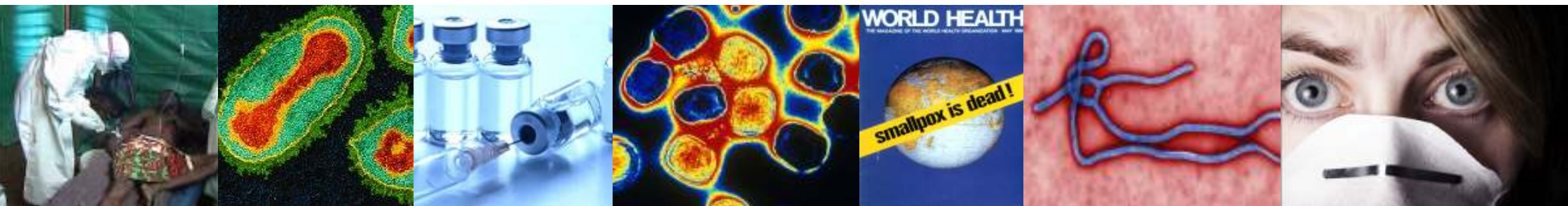


# Morfologia, Ultraestrutura e Taxonomia de Vírus

**Helena Lage Ferreira**

Disciplina ZMV0368 – Microbiologia Fundamental

Engenharia de Alimentos - Diurno



# Morfologia, Ultraestrutura e Taxonomia de Vírus

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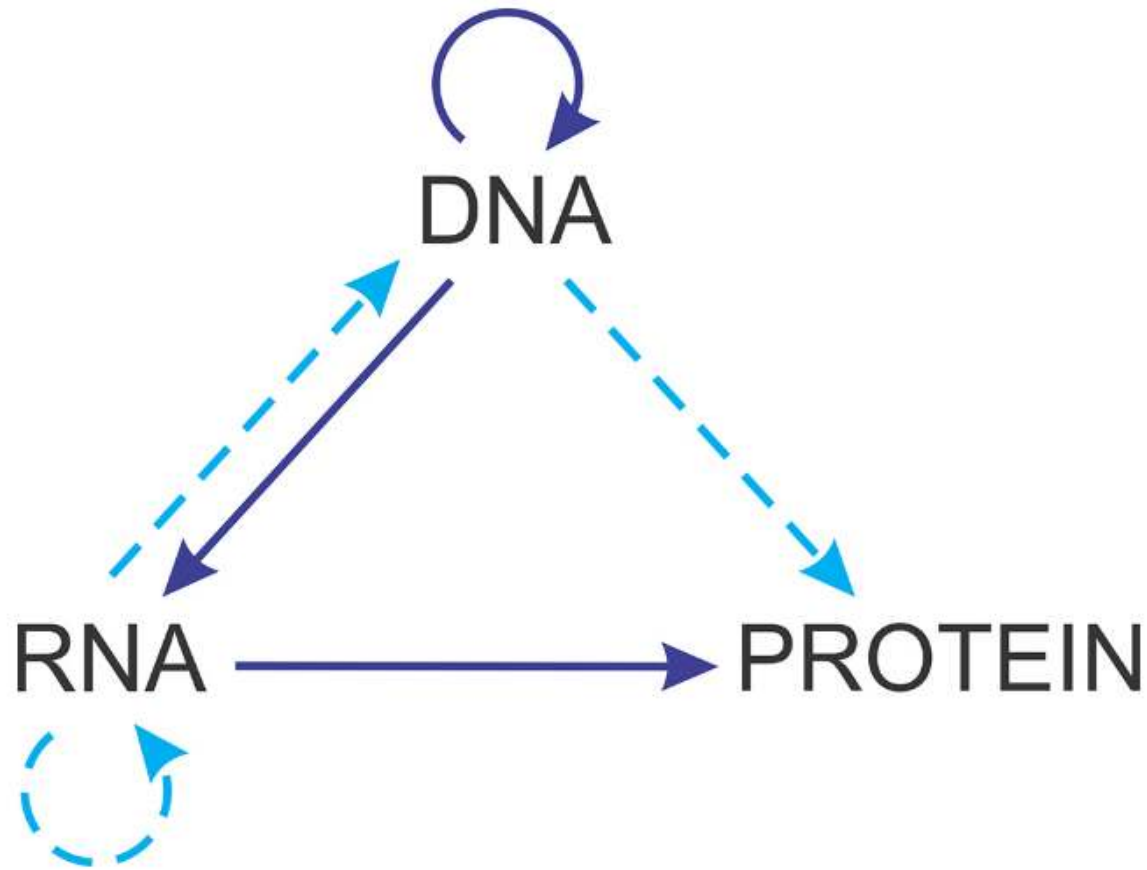
Disciplina ZMV0368 – Microbiologia Fundamental

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Cortesia slides: Flávio G. da Fonseca. ICB, CT-Vacinas, UFMG e Ricardo Moro – FZEA, USP



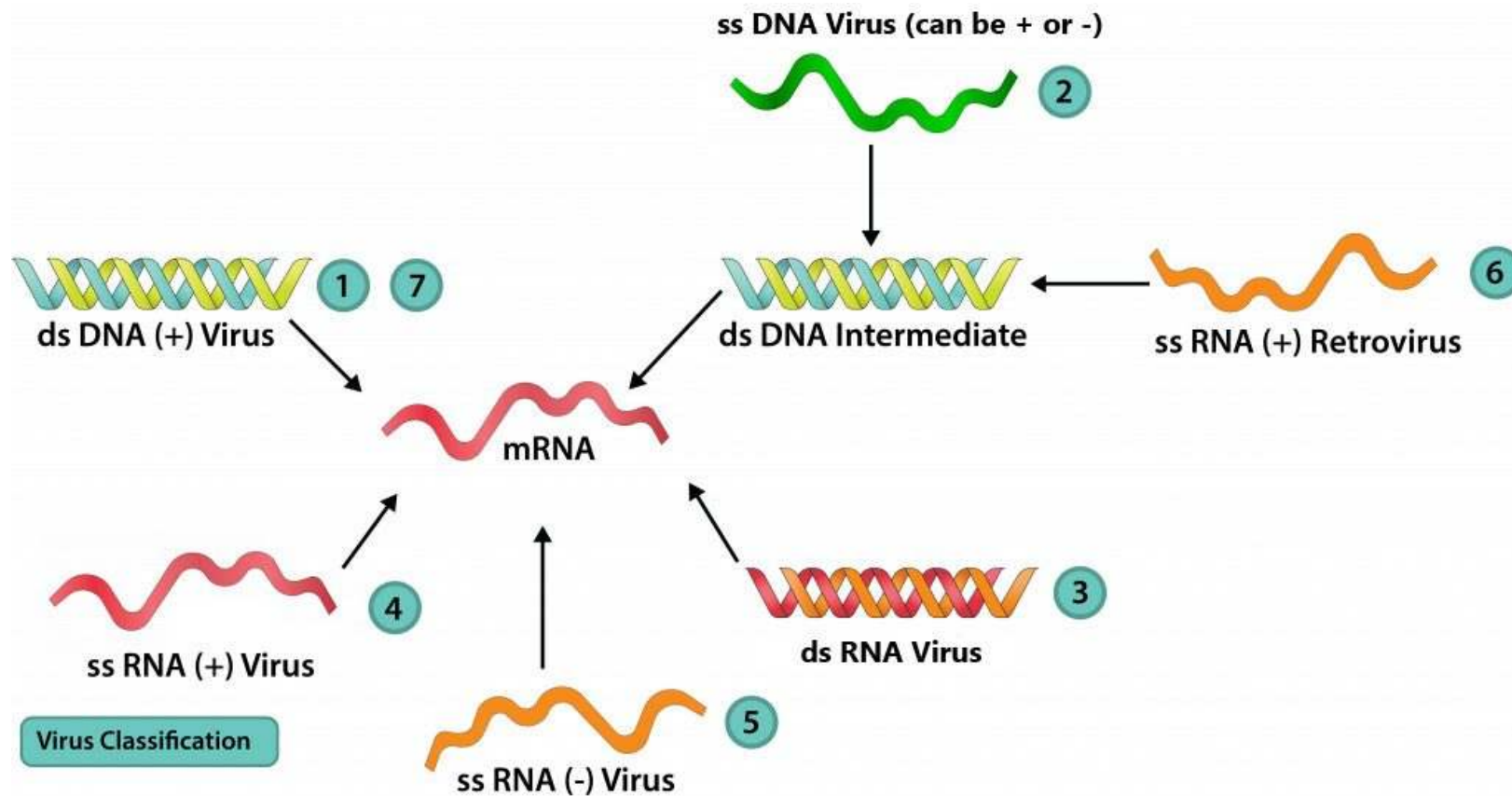
# Dogma da biologia molecular e



**FIG 1** The central dogma of molecular biology. The figure is redrawn from Crick's 1970 article. The solid arrows represent the mainstream routes of information flow, and the dashed arrows show [putative] "special routes" after Crick. Adapted from reference 10 with permission of Springer Nature.



# Material genético dos vírus



# Histórico

**COVID-19**

Coronavirus Disease 2019



<https://www.youtube.com/watch?v=ZMCVgtV2sG4&feature=youtu.be>



# Eventos mais relevantes na virologia



## 1796

**1796** Edward Jenner administers the first smallpox vaccine, which is heralded as the world's first vaccine. The vaccine consists of fluid from a cowpox blister, a virus similar to smallpox, which is scratched onto the skin of an 8-year old boy. When the boy is later inoculated with smallpox matter, no disease develops.



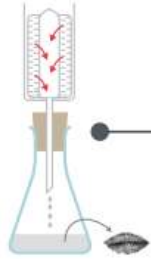
## 1885

**1885** French scientists, Louis Pasteur and Emile Roux, develop the first effective rabies vaccine. The virus is grown in rabbits and a vaccine is made from dried rabbit nervous system tissue, which is successfully administered to a boy that was bitten by a rabid dog.

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**1892** Dmitri Ivanovsky shows that tobacco mosaic disease, a single-stranded RNA plant virus, can be transmitted by extracts passed through porcelain Pasteur-Chamberland filters, which exclude the smallest known bacteria.

## 1892



**1898** Martinus Beijerinck replicates the filtration experiments performed by Dmitri Ivanovsky and calls the infectious agent that causes tobacco mosaic disease a "virus", which he describes as a "contagium vivum fluidum" or "contagious living fluid". Beijerinck along with Ivanovsky are considered to be the founders of virology.

## 1898

**1898** The first animal virus, known as foot and mouth disease virus, is discovered by German scientists, Friedrich Loeffler and Paul Frosch. They show that cows and sheep can be vaccinated against the virus using a heat-inactivated, filtered vesicle extract.



## 1901



**1901** Walter Reed heads the U.S. Army Yellow Fever Commission, which discovers that yellow fever is transmitted by the bite of an *Aedes aegypti* mosquito rather than by direct contact.



**1911** Peyton Rous discovers Rous sarcoma virus (RSV), the first oncogenic retrovirus to be described, which is found to cause sarcoma in chickens. Rous shares the Nobel Prize in Physiology or Medicine in 1966 for his discovery of tumor-inducing viruses.

## 1911



## 1933

**1933** Cottontail rabbit papillomavirus (CRPV) is discovered and is shown to be the first oncogenic DNA virus in 1935. CRPV causes skin tumors and warts that are typically located on the heads of infected rabbits.

## 1935

**1935** Wendell Stanley produces the first crystals of tobacco mosaic virus and shows that the virus remains active after crystallization. Crystallization of the virus was the first step toward proving that the virus is particulate.



## 1936

**1936** John Bittner reports that an infectious, filterable agent present in the milk of certain cancer-prone mouse strains can be transmitted to newborn, cancer-resistant mice by suckling and it can lead to the development of mammary tumors. This infectious agent later came to be known as mouse mammary tumor virus.



# Eventos mais relevantes na virologia

## 1937



**1937** Max Theiler grows the yellow fever virus in chicken eggs and produces a vaccine from an attenuated virus strain. In 1951, he receives the Nobel Prize in Physiology or Medicine for his discovery of an effective yellow fever vaccine, the only Nobel Prize ever awarded for the development of a virus vaccine.



**1951** Ludwik Gross identifies the first murine leukemia virus.

## 1951



## 1953

**1953** Jonas Salk announces on a national radio show and later reports in *The Journal of the American Medical Association* that he has successfully developed and tested an injectable, killed-virus vaccine against poliovirus, the virus responsible for poliomyelitis. Testing of the vaccine starts in 1954 and in 1955, it is announced that the vaccine is safe and effective. In 1962, an oral vaccine developed by Albert Sabin using a weakened form of the live virus becomes available.



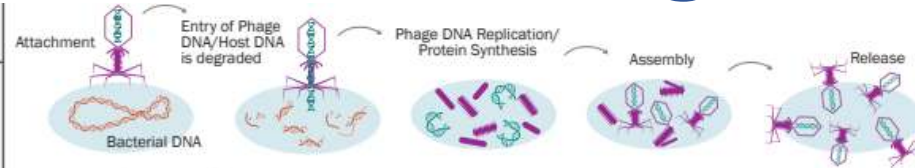
## 1955

**1955** Rosalind Franklin proposes the full structure of tobacco mosaic virus, suggesting that the virus contains a single strand of RNA that spirals in a helical groove inside the center of the viral proteins. Solving the structure of the tobacco mosaic plant virus paved the way for solving the structure of animal viruses, which Franklin's lab subsequently pursued, leading to a publication following her death that described the crystal structure of poliovirus.



## 1962

**1962** John Trentin reports that human adenovirus is capable of causing tumors in experimentally-infected animals. This is the first known human virus reported to be capable of inducing cancer.

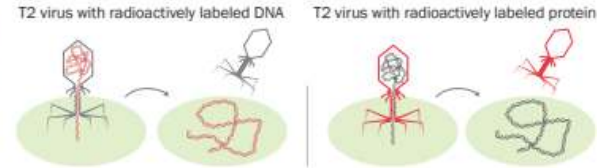


## 1939

**1939** Emory Ellis and Max Delbruck establish the concept of the one-step virus growth cycle, which serves as the basis for understanding viral replication and the virus life cycle. They demonstrate that virus particles do not grow, but rather are assembled from preformed components.

## 1952

**1952** Hershey and Chase demonstrate that DNA alone, not protein, enters a bacterial cell upon infection with enterobacteriophage T2, which is a virus that infects and kills *E. coli*.

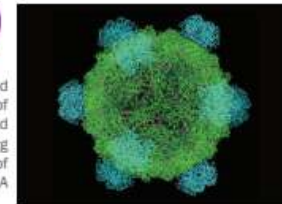


## 1954

**1954** John Franklin Enders, known as "The Father of Modern Vaccines", along with Thomas Huckle Weller and Frederick Chapman Robbins share the Nobel Prize in Physiology or Medicine for their discovery that poliovirus could be grown in cultures using various types of tissues without needing an intact organism. This finding allowed both inactivated and live polio vaccines to be produced for the first time and was critical to being able to create large quantities of different kinds of viruses for research.

## 1959

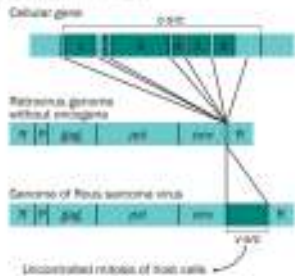
**1959** The Nobel Prize in Physiology or Medicine is jointly awarded to Severo Ochoa and Arthur Kornberg for their discovery of the mechanisms in the biological synthesis of ribonucleic acid and deoxyribonucleic acid. Ochoa discovered an enzyme that could synthesize RNA, while Kornberg discovered an enzyme that could synthesize DNA. Kornberg later showed that DNA synthesized in a test tube by purified enzymes could produce all of the features of a natural virus using the bacteriophage Phi X 174, a single-stranded DNA virus, as a model.



# Eventos mais relevantes na virologia

1970

1970 The first retroviral oncogene, a Src, is discovered in Rous sarcoma virus. The gene encodes a tyrosine kinase involved in cell growth and differentiation.



1970 Reverse transcriptase is discovered by Howard Temin in RSV viruses and independently isolated by David Baltimore from two RNA tumor viruses, RMT1 and RMT2. The enzyme is used by retroviruses to generate complementary DNA from an RNA genome, which is then stably integrated into the chromosomal DNA of the host. Temin and Baltimore are jointly awarded the Nobel Prize in Physiology or Medicine in 1975, along with Francis Crick, for their discovery.

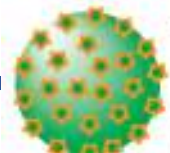


1976

1976 The first known outbreaks of Ebola virus disease (EVD) occur in South Sudan and in the Democratic Republic of the Congo, and are found to be caused by two distinct subtypes of Ebolavirus. While the natural host of Ebola remains unknown, bats are now believed to be the most likely reservoir.

1980

1980 Robert Gallo discovers the first human retrovirus, Human T-lymphotropic virus (HTLV-1), by visualizing viral particles in cultured human T-cell lymphoma cells.



1983

1983-1984 Harold zur Hausen shows that two strains of the human papillomavirus (HPV) cause most cases of cervical cancer, a discovery for which he was awarded jointly the Nobel Prize in Physiology or Medicine in 2008, along with Lui Montagnier and Pierrette Barre-Sinoussi, who discovered HIV.

1965

1965 Berach Blumberg and his colleagues discover a new antigen in the serum of an Australian aborigine that reacts with an antibody in the sera from patients with leukemia who had received blood transfusions. This antigen is called the Australia antigen, and is later found to be a surface antigen of the hepatitis B virus (HBV). In addition to discovering HBV, Blumberg later develops a screening test for the virus and an approach for developing a vaccine. In 1976, he shares the Nobel Prize in Physiology or Medicine for his discovery of the new mechanisms for the origin and dissemination of infectious diseases.



1971



1971 The measles, mumps, rubella (MMR) vaccine is developed by Maurice Hilleman at Merck Pharmaceutical Co. The vaccine is a mixture of the three attenuated viruses. Over his lifetime, Hilleman is credited with developing over 40 vaccines and saving millions of lives through his efforts.

1975

1975 Garuch Blumberg discovers a link between chronic hepatitis B virus (HBV) infection and hepatocellular carcinoma (HCC). This link is confirmed in a 1982 paper by Stanley, R.P. et al. published in The Lancet, which reports that chronic HBV infection is associated with a 500-fold increase in the risk of developing HCC.



1977

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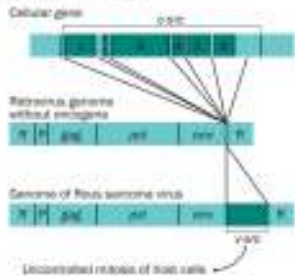




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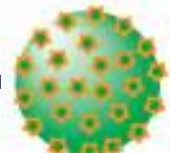


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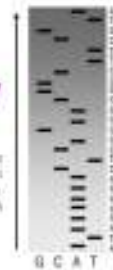
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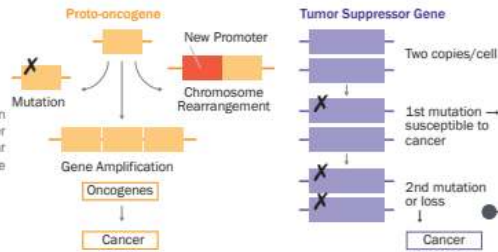
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# Eventos mais relevantes na virologia

## 1988

1988 Ed Harlow and David Livingston demonstrate that viruses can promote cancer either by activating the products of cellular proto-oncogenes or by inactivating the products of cellular tumor suppressor genes.

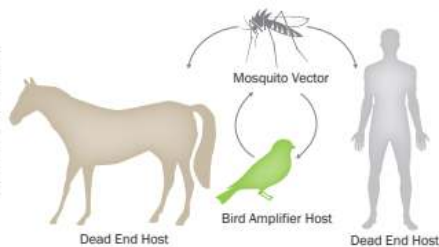


## 1989

1989 Michael Houghton and his colleagues at Chiron Corporation along with Dan Bradley at the CDC discover the hepatitis C virus (HCV). Chronic HCV infection is found to be associated with hepatocellular carcinoma (HCC).

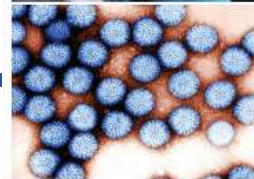
## 1999

1999 The first documented cases of West Nile Virus (WNV) infection in the Western Hemisphere are recorded in New York City in August of this year, following reports of a number of severe cases of encephalitis and avian deaths. During August and September, 59 patients are hospitalized with WNV infection. This initial outbreak is followed by years of progressive spread of the virus throughout the U.S., with the largest annual epidemic of WNV in North America occurring in 2003.



## 2006

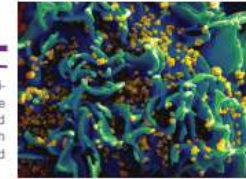
2006 A vaccine protecting against the two cancer-causing strains of human papillomavirus (HPV) is approved by the U.S. Food and Drug Administration. While more than 100 HPV types have been identified, HPV 16 and HPV 18 are the two strains that have been found to cause 70% of cervical cancers. The HPV vaccine approved in this year targets both of these strains and two other low-risk HPV types, HPV 6 and HPV 11.



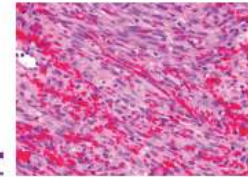
2006 The U.S. Food and Drug Administration approves a vaccine for preventing rotavirus, a double-stranded RNA virus that is easily spread among children. Infection with rotavirus can have devastating complications including severe diarrhea, abdominal pain, vomiting, and even death, particularly in developing countries.

## 1984

1984 Luc Montagnier and Françoise Barré-Sinoussi discover the human immunodeficiency virus (HIV), a retrovirus that attacks lymphocytes, which is later shown to be the causative agent of AIDS. Luc Montagnier and Françoise Barré-Sinoussi are awarded jointly the Nobel Prize in Physiology or Medicine in 2008 for their discovery, along with Harald zur Hausen, who identified the link between human papillomaviruses and cervical cancer.



## 1994



1994 Yuan Chang and Patrick Moore discover Kaposi's sarcoma herpesvirus (KSHV) in Kaposi's sarcoma tissue from AIDS patients. In addition to being the causative agent of Kaposi's sarcoma, KSHV is also associated with Castleman's disease, primary effusion lymphoma, and KSHV inflammatory cytokine syndrome.

## 2003

2003 The Centers for Disease Control and Prevention and Canada's National Microbiology Laboratory identify the severe acute respiratory syndrome (SARS) coronavirus genome, which is later confirmed to be the causative agent of SARS.



## 2005

2005-2006 A severe Chikungunya outbreak occurs on the islands of Mauritius and Réunion in the Indian Ocean with more than 272,000 cases being reported. This outbreak is followed by an outbreak in India in 2006 and 2007 during which more than 1,500,000 cases of Chikungunya or dengue fever are reported. In 2007, a localized outbreak occurs in northern Italy, the first cases to ever be reported in Europe. Chikungunya is found for the first time in the Americas in late 2013 on islands in the Caribbean.



## 2009

2009 The World Health Organization declares the H1N1/Influenza A, or swine flu, outbreak that occurs in this year to be a global pandemic. The H1N1 virus responsible for the outbreak is found to have a unique combination of swine, avian, and human genes that had not been seen before. Although the 2009 pandemic was caused by human-to-human transmission, it was called "swine flu" because genetic analysis of the virus showed that it was most similar to H1N1 viruses with swine origins. President Obama declared the swine flu outbreak in the U.S. to be a national emergency in October of this year.



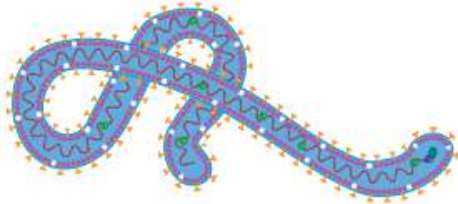
R&D SYSTEMS  
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# Eventos mais relevantes na virologia

## 2011

**2011** Rinderpest, a contagious viral disease of ruminant mammals, primarily cattle, is declared to be fully eradicated by veterinary epidemiologists, making it only the second disease in history following smallpox to be globally eradicated.



## 2014

**2014** The largest Ebola outbreak in history occurs in West Africa, resulting in the death of more than 11,300 people in Guinea, Liberia, and Sierra Leone. In September of this year, the CDC confirms the first laboratory-confirmed case of Ebola to be diagnosed in the U.S., which was found in a man who had traveled from Liberia to Texas.

## 2018

**2018** The world's second largest Ebola outbreak on record begins in August of this year with four cases being confirmed in North Kivu Province in the Democratic Republic of the Congo (DRC). On July 17, 2019, the outbreak is declared a Public Health Emergency of International Concern by the World Health Organization as there are more than 2,500 cases and more than 1,600 deaths reported by the DRC Ministry of Health. By March 16, 2020, more than 3,400 cases of Ebola virus disease are confirmed, and more than 2,260 people have died since the outbreak was first declared.

## 2012

**2012** The first case of Middle East Respiratory Syndrome (MERS) is reported in Saudi Arabia. It is found to be caused by Middle East Respiratory Syndrome coronavirus (MERS-CoV), which is thought to have come from an animal source, possibly camels, in the Arabian Peninsula.



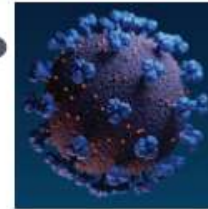
**2016** Due to the rapid spread of the Zika virus across Latin America and the Caribbean along with the dramatic increase in the number of reported cases of prenatal microcephaly and adult neurological disorders such as Guillain-Barré syndrome in Brazil and other areas affected by the virus, the World Health Organization declares a public health emergency of international concern.



## 2016

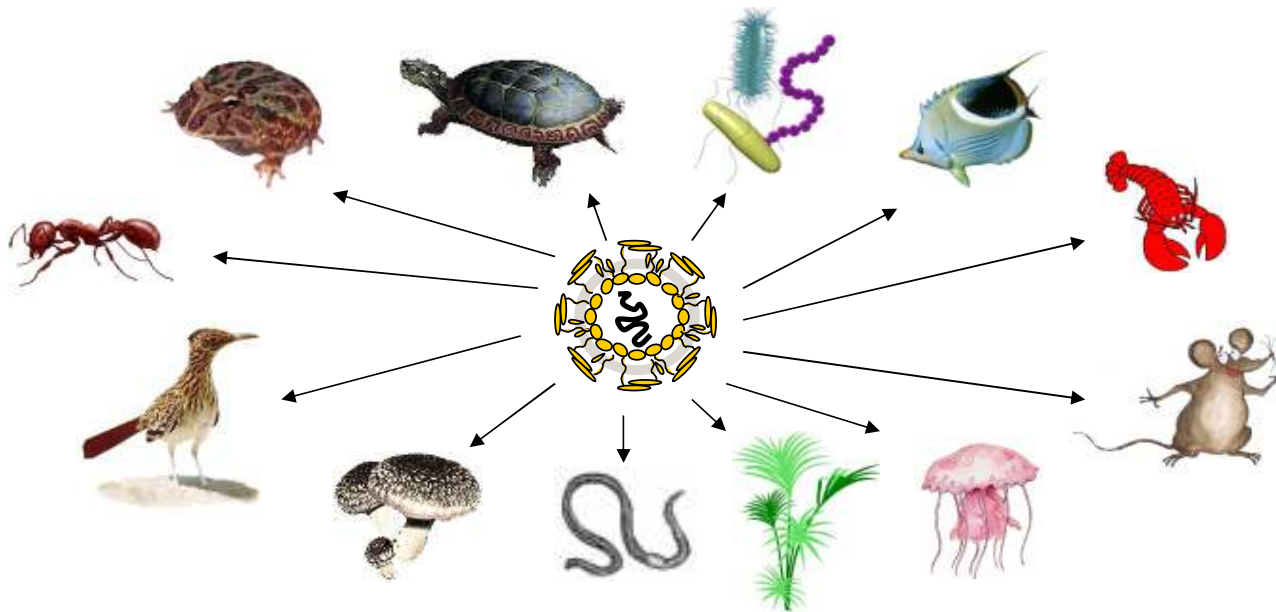
## 2019

**2019** In late December of this year, a novel coronavirus called severe acute respiratory syndrome (SARS)-CoV-2, is found to be responsible for an outbreak of respiratory illness that was first reported in the city of Wuhan in the Hubei Province of China. The disease caused by SARS-CoV-2 is designated as coronavirus disease 19 (COVID-19), which in severe cases, can lead to potentially life-threatening complications including pneumonia and respiratory failure. On January 30, 2020, the outbreak is declared a global health emergency by the World Health Organization (WHO) as nearly 8,000 cases are reported worldwide and there is evidence that the virus can be transmitted from person-to-person. Six weeks later on March 11, 2020, the WHO declares COVID-19 to be a pandemic as there are over 118,000 cases reported in more than 114 countries and territories around the world, and 4,291 people are reported to have died from the disease.



# Ubiquidade dos vírus

- Todos e qualquer organismo é potencialmente parasitado por algum tipo de vírus

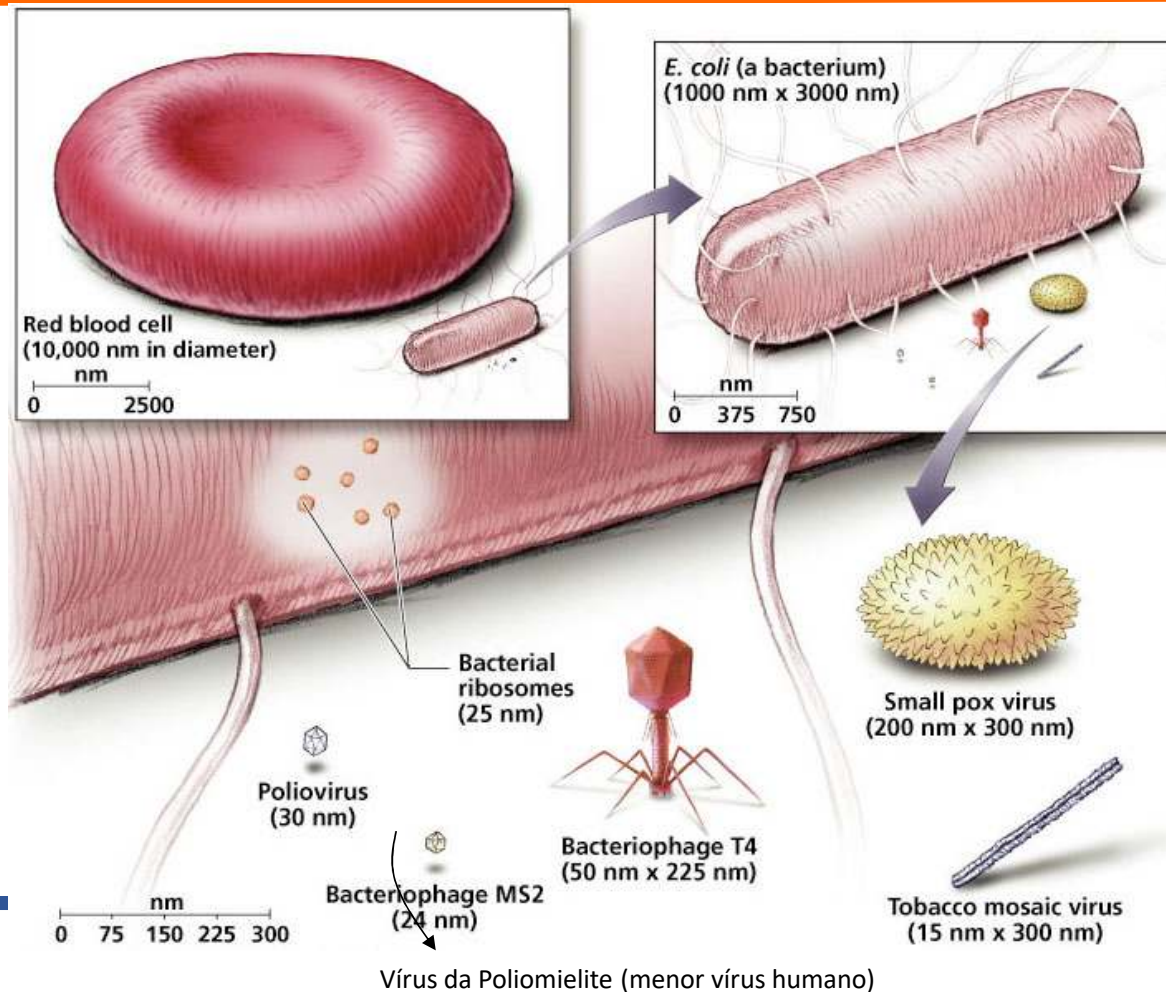


- Consequência evolutiva

Natureza predatória: todo vírus é um parasita obrigatório



# Tamanho dos Vírus



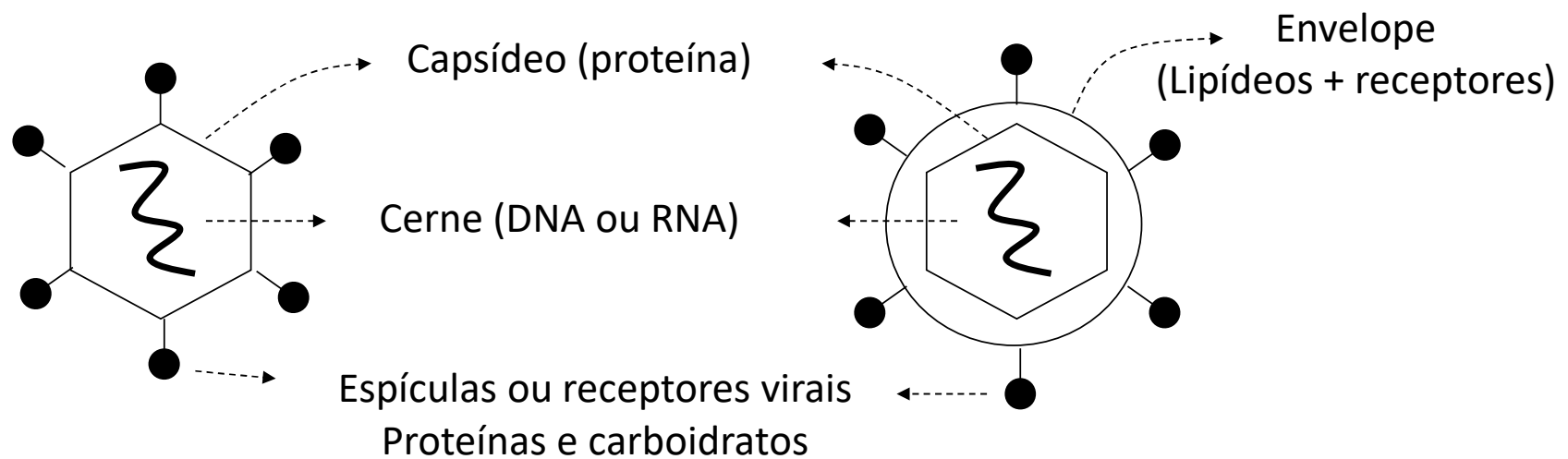
Tamanho da partícula: 15 a 300 nm

1nm = 1/10.000.000 cm

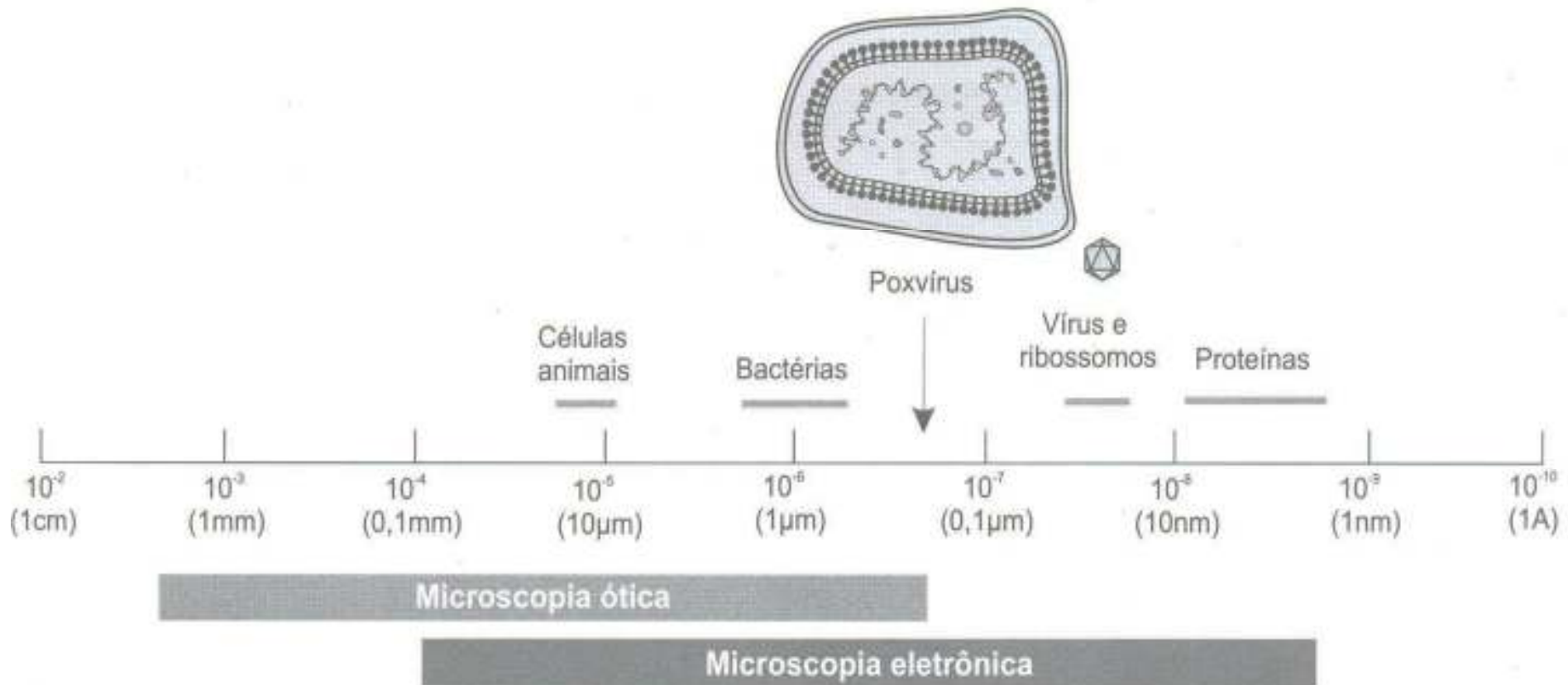


# Estrutura Viral Básica

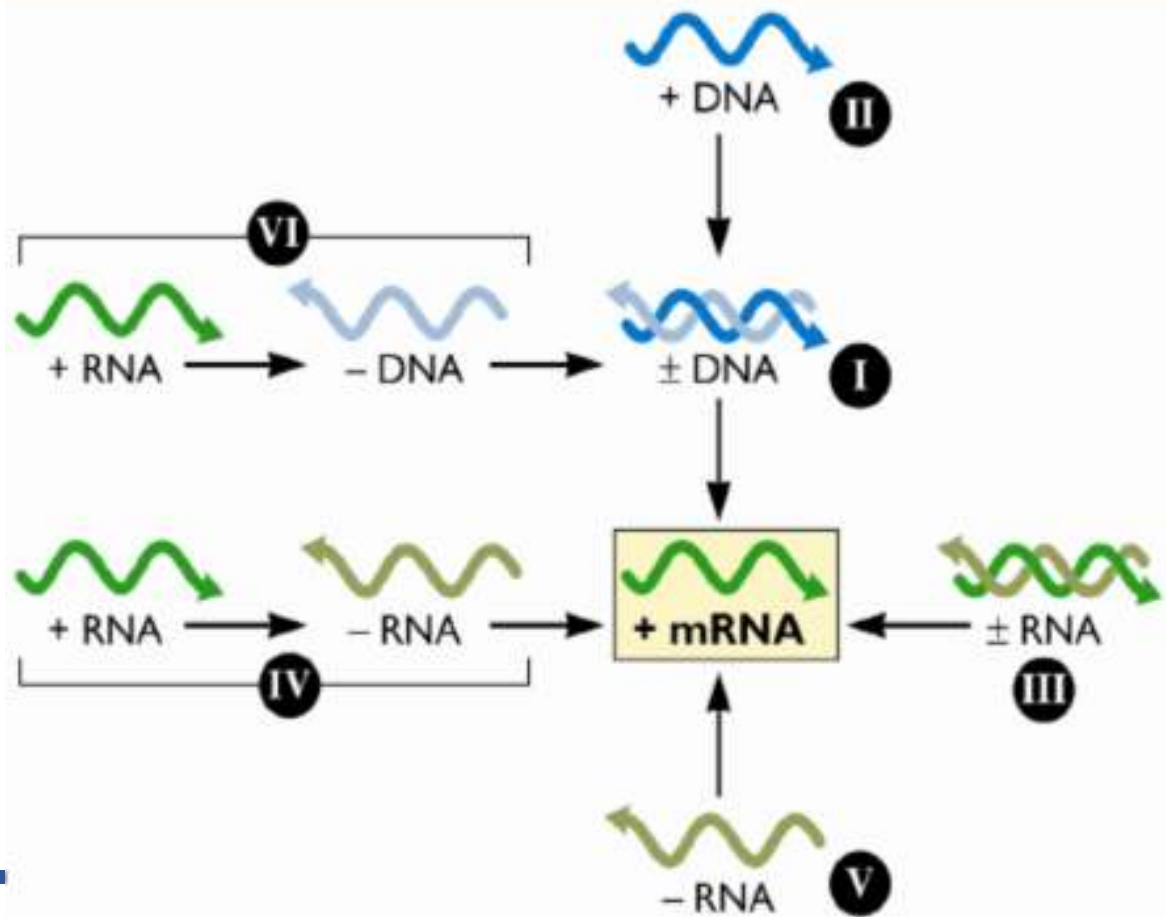
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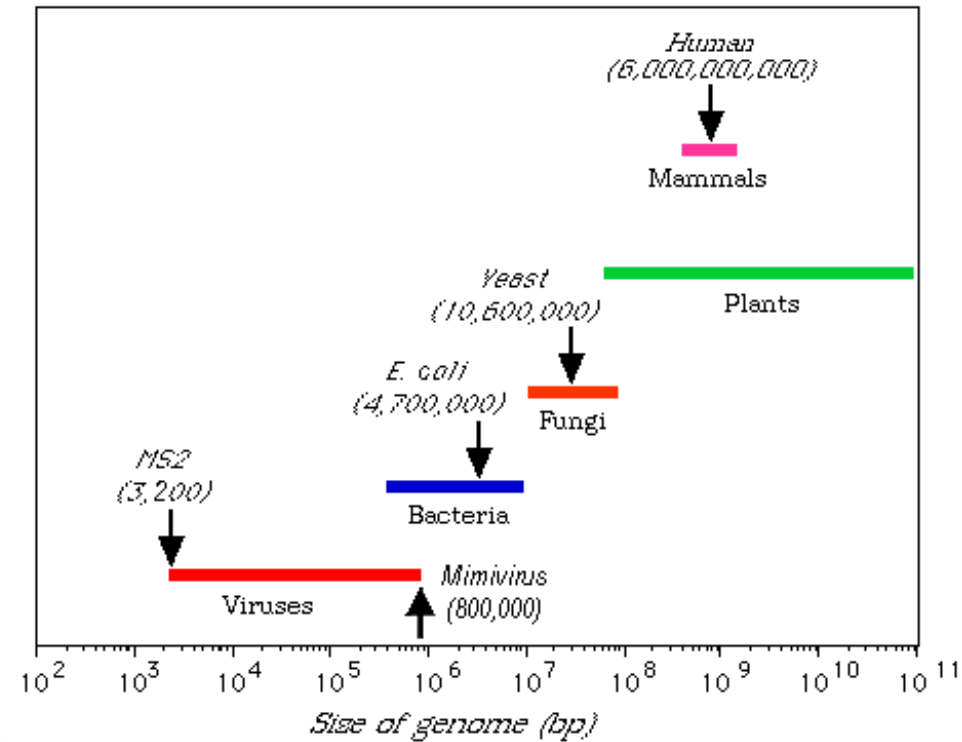
# Tamanho



# Genoma Viral



Comparison of Genome Size:





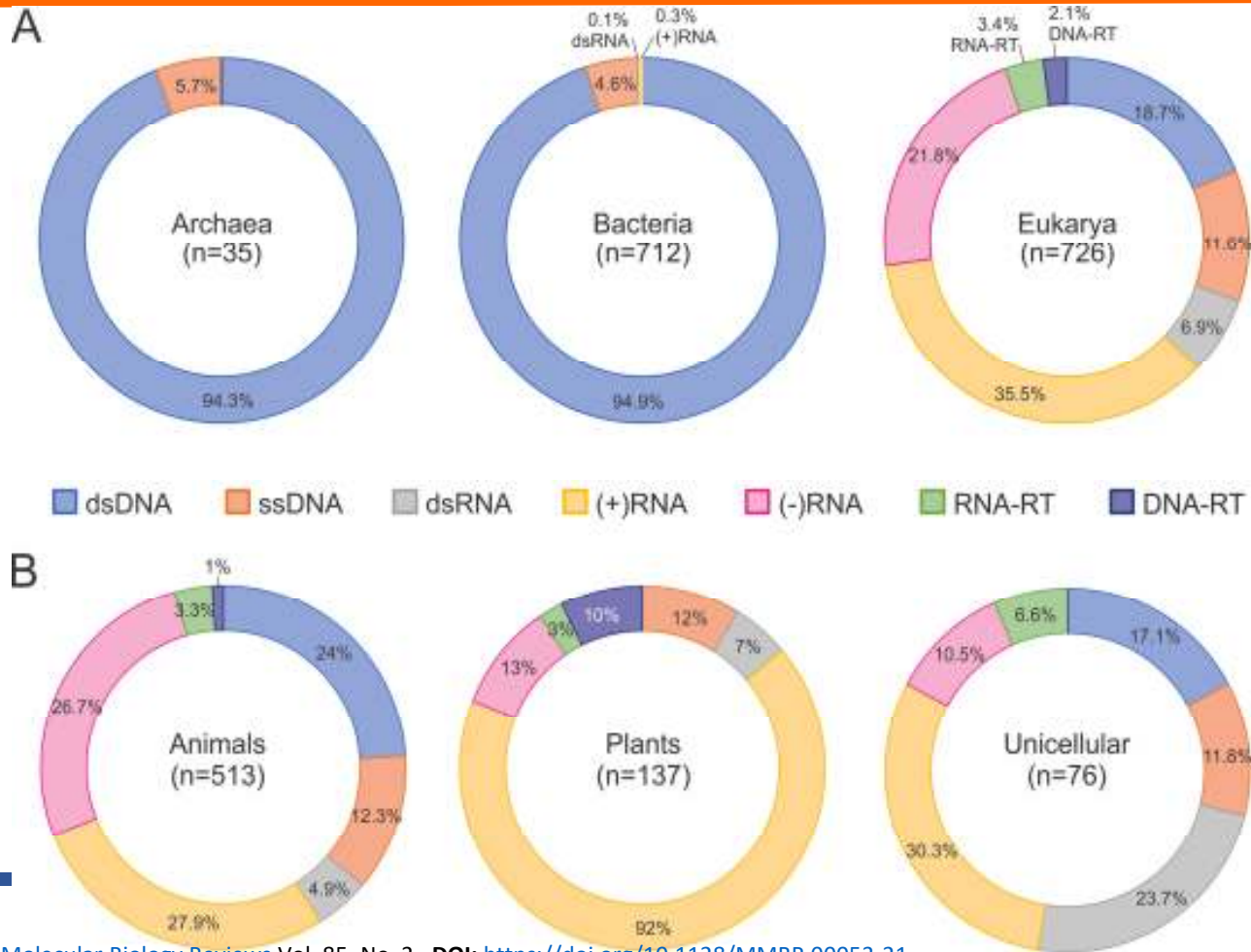
# Ácido Nucleico

---

- simples fita ou dupla fita
- ssDNA, dsDNA, ssRNA e dsRNA
- linear ou circular
- segmentado ou não-segmentado (contínuo)
- ex: 1,2 milhões pares de bases



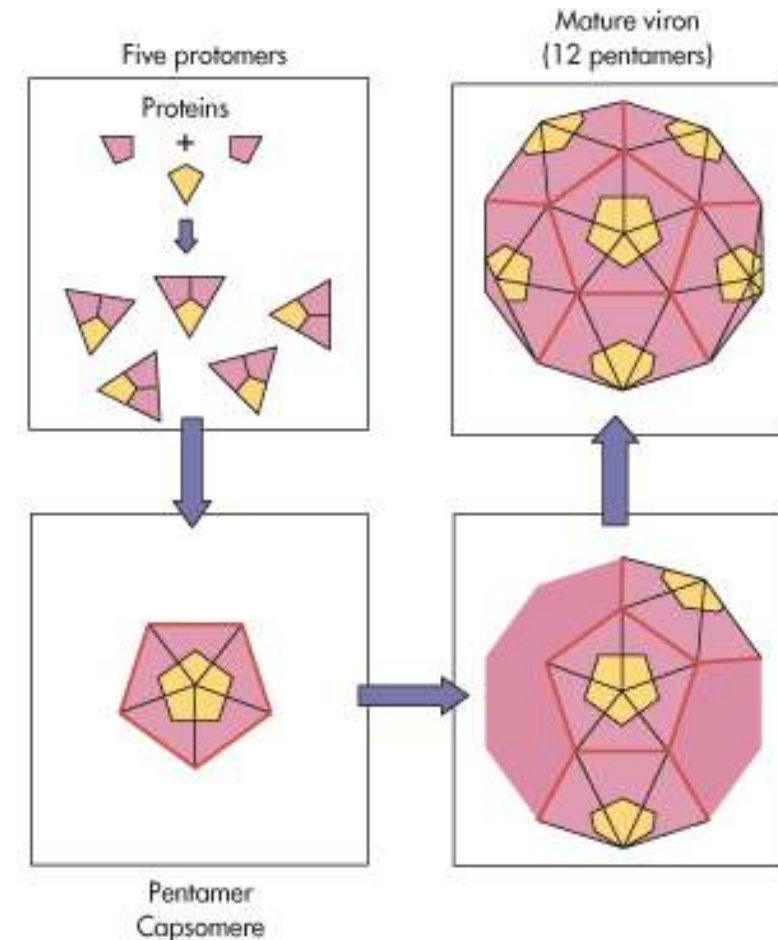
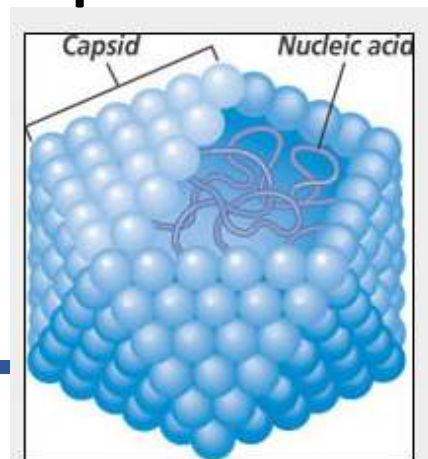
# Ácido nucleico



# Capsídeo

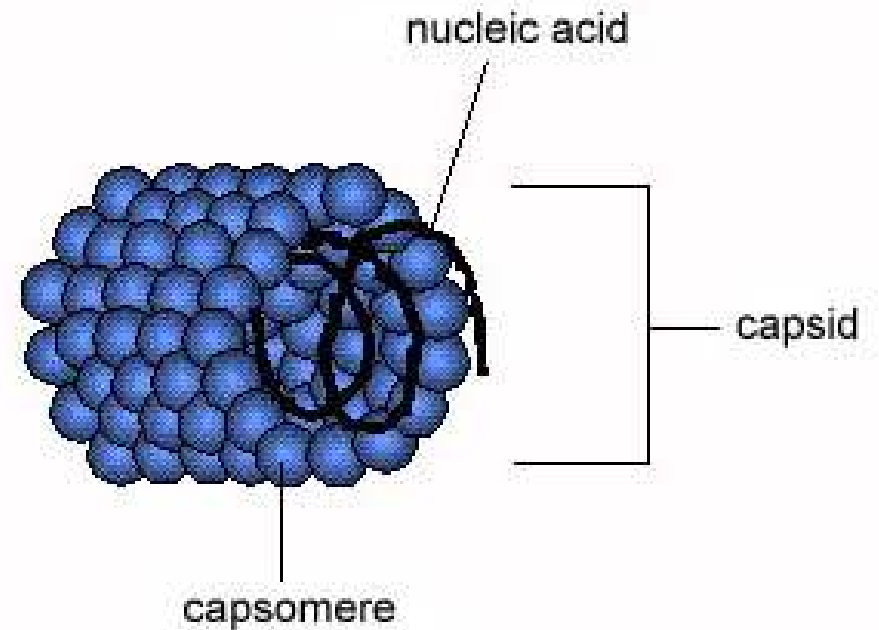
Capsídeo: função de proteger o material genético

- subunidades estruturais: capsômeros
- 1 ou mais tipos de proteínas (protômeros)



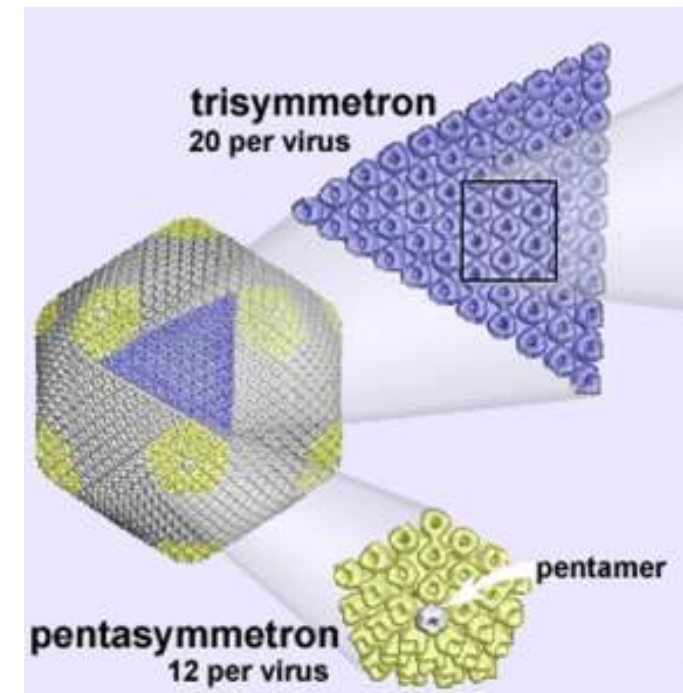
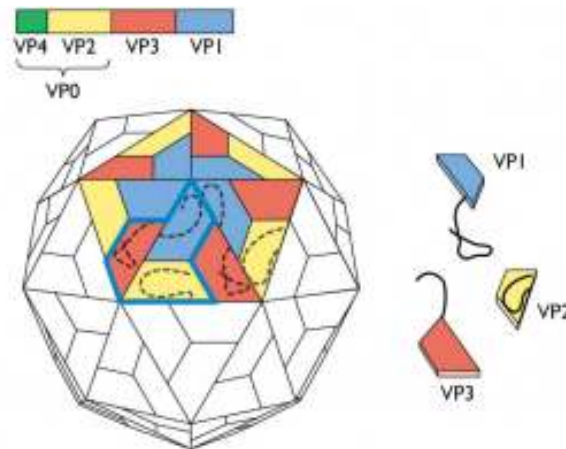
# Capsídeo

- Morfologia viral baseada na estrutura do capsídeo
  - helicoidais: lembram longos bastonetes, genoma no interior de capsídeo cilíndrico oco. Ex.: raiva

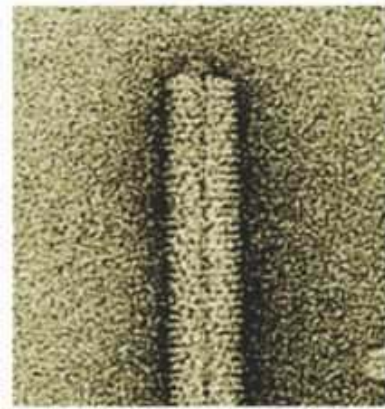
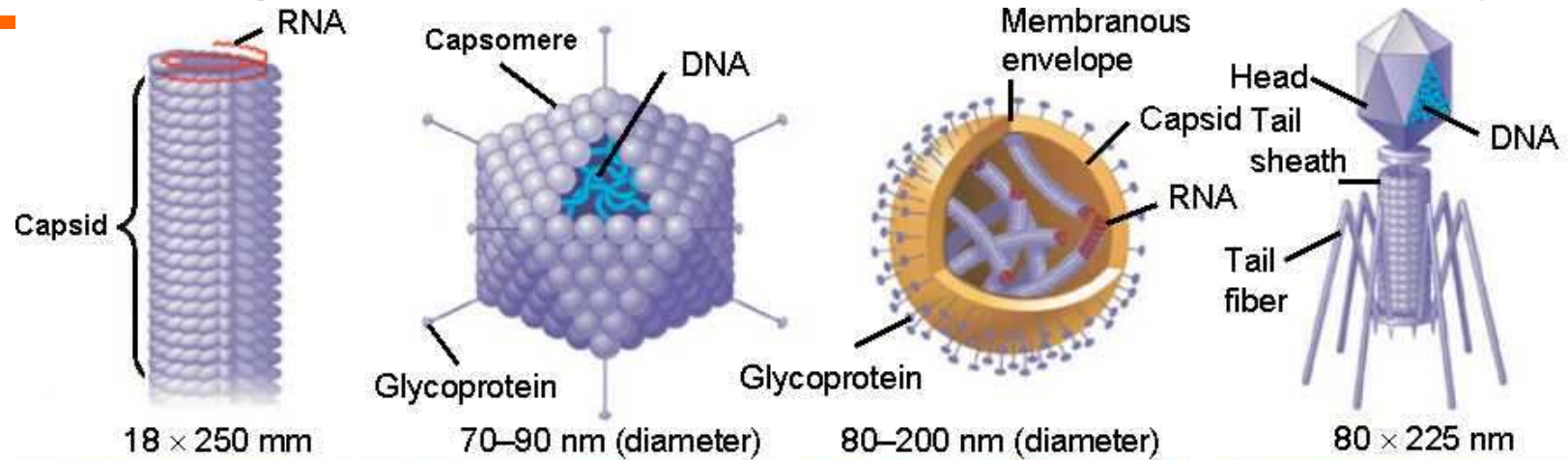


# Capsídeo

- poliédricos
- Capsídeo tem forma de icosaédrico.
- Capsômeros de cada face formam um triângulo equilátero.
- Ex.: adenovírus



# Morfologia baseada na estrutura do capsídeo



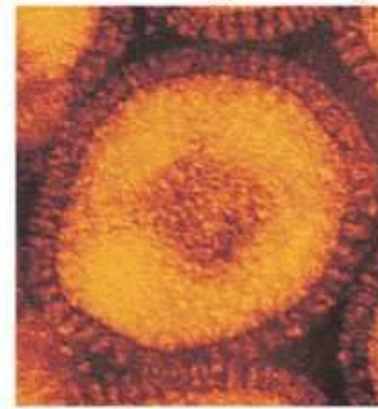
20 nm

(a) Tobacco mosaic virus



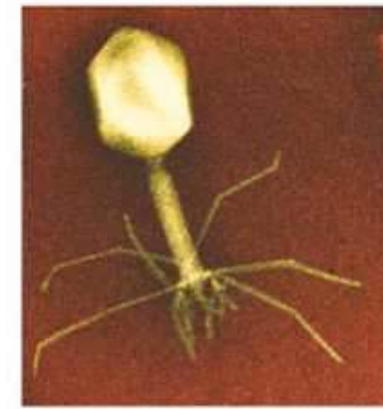
50 nm

(b) Adenoviruses



50 nm

(c) Influenza viruses



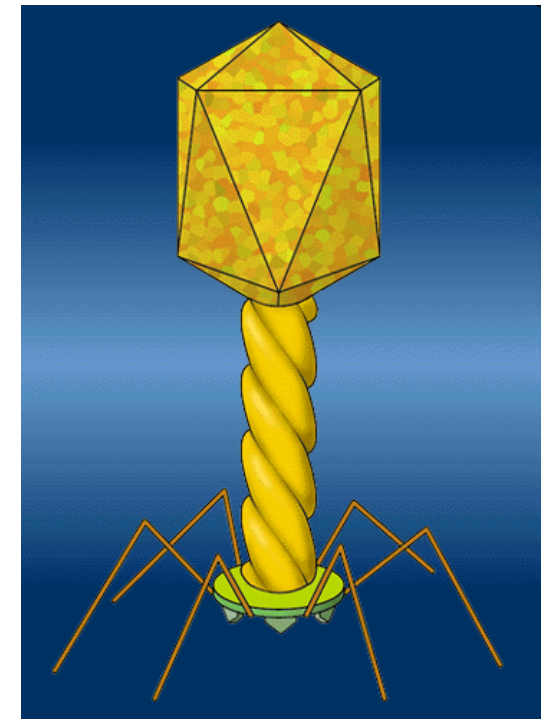
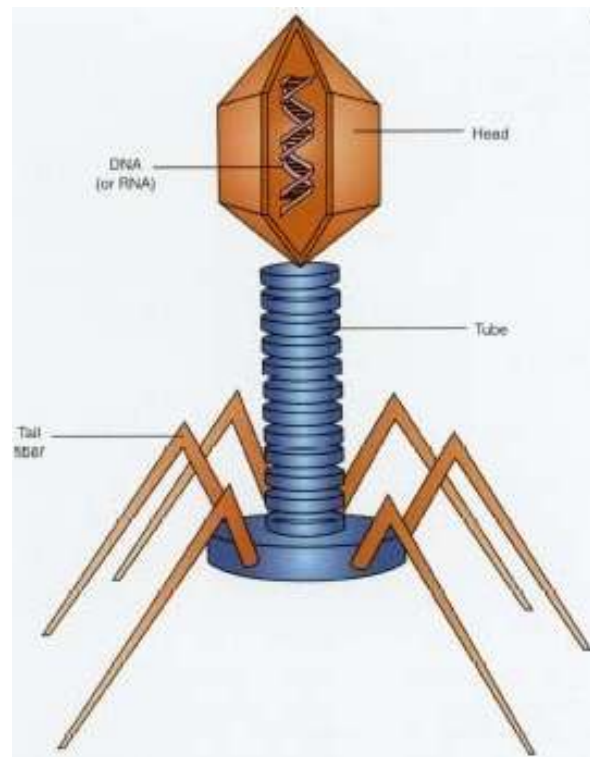
50 nm

(d) Bacteriophage T4



# Capsídeo

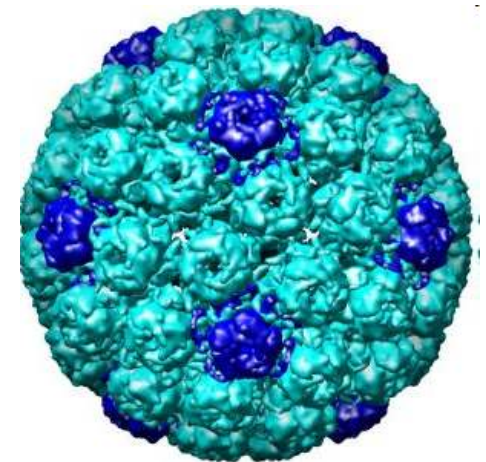
- Morfologia baseada na estrutura do capsídeo
  - complexos: poliédrico + estruturas adicionais (helicoidal).
  - Ex.: bacteriófagos



# Proteínas virais

---

- **Estruturais:**
  - Participam da construção e “arquitetura”/estrutura do vírus ⇒ presentes no vírion
- **Não-estruturais:**
  - Produzidas durante o ciclo replicativo, não participam da “arquitetura” do vírion ⇒ atividades enzimáticas e/ou regulatórias





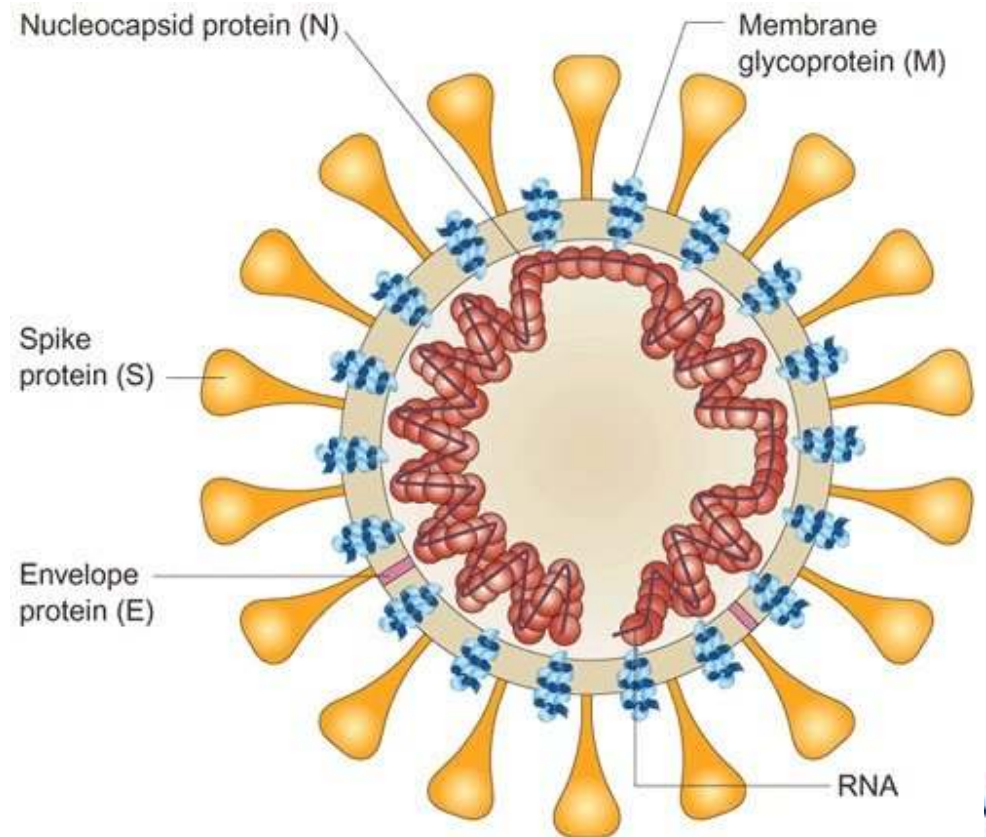
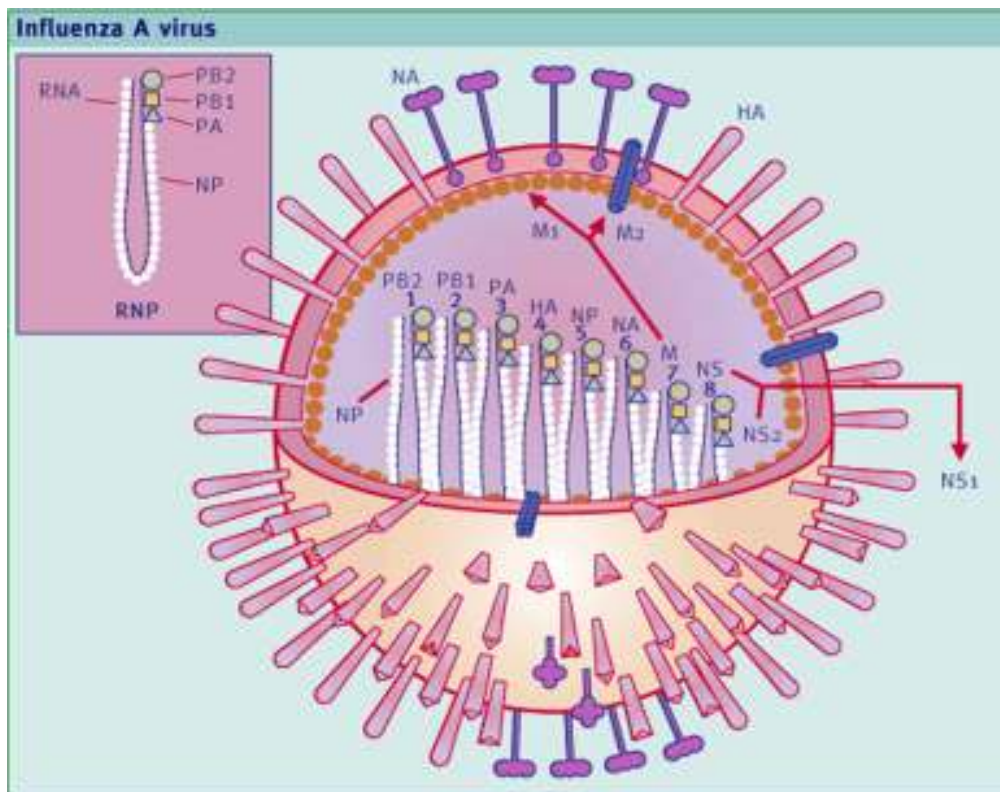
# Proteínas virais

---

- lipídeos (fosfolipídeos e colesterol) + proteínas virais + carboidratos
- derivado de membranas da célula hospedeira (plasmática, retículo endoplasmático, Golgi) através de mecanismo denominado brotamento
- lábil: ácidos, bases, calor, detergente, ressecamento
- espículas ou peplômeros:

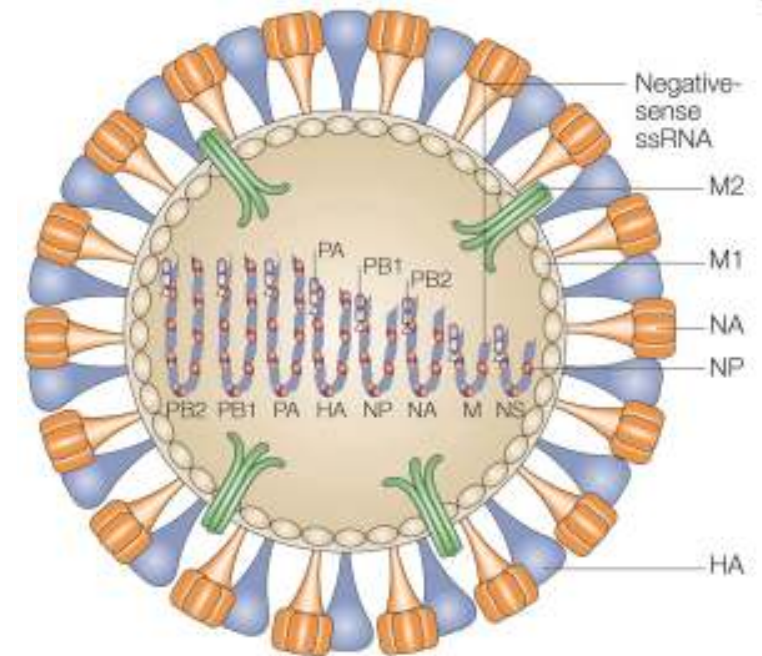


# Envelope viral



# Matriz

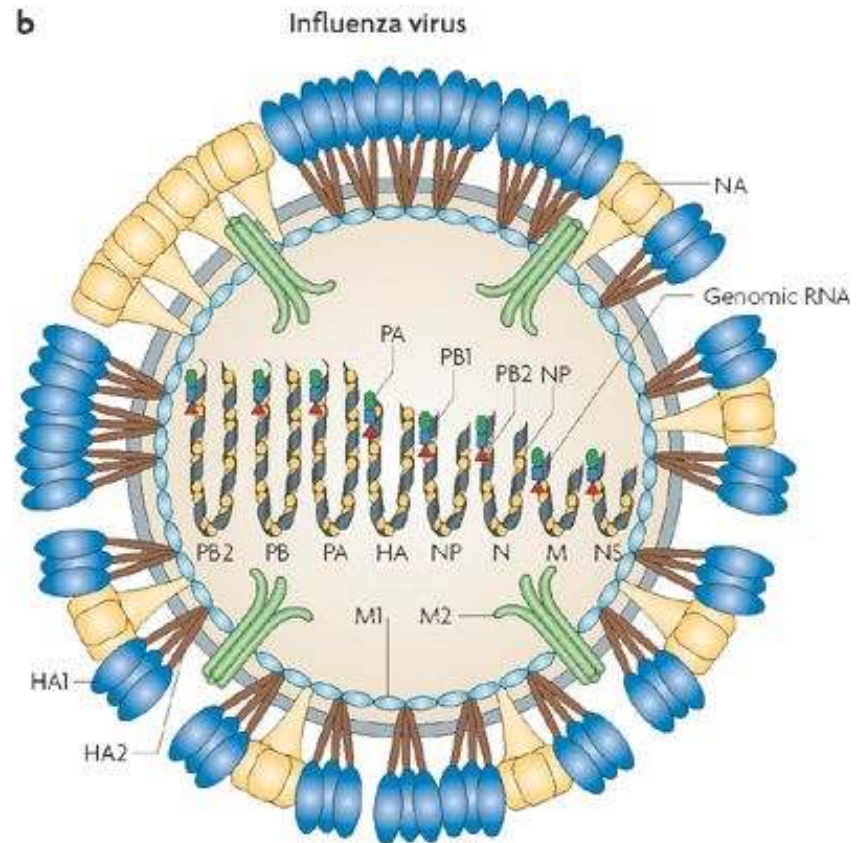
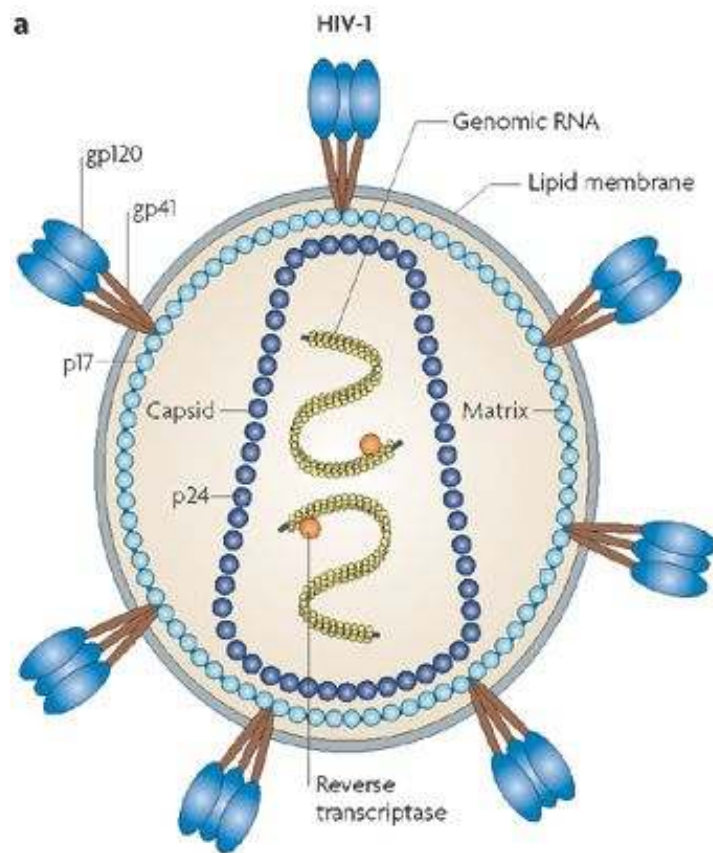
- camada protéica recobrando externamente o nucleocapsídeo
- faz interação entre nucleocapsídeo e envelope
- glicosilada



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Nature Reviews | Microbiology



# Matriz

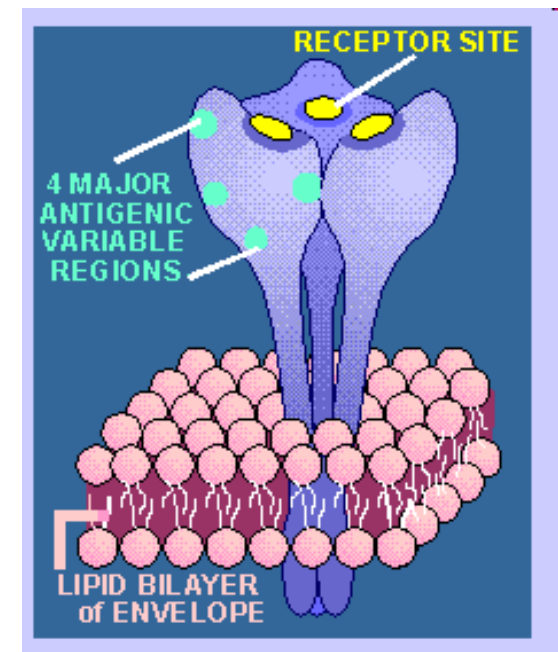


# Proteínas do envelope

---

→ monômeros, heterodímeros, tetrâmeros      homo      ou  
trímeros      ou

→ geralmente: proteínas integrais



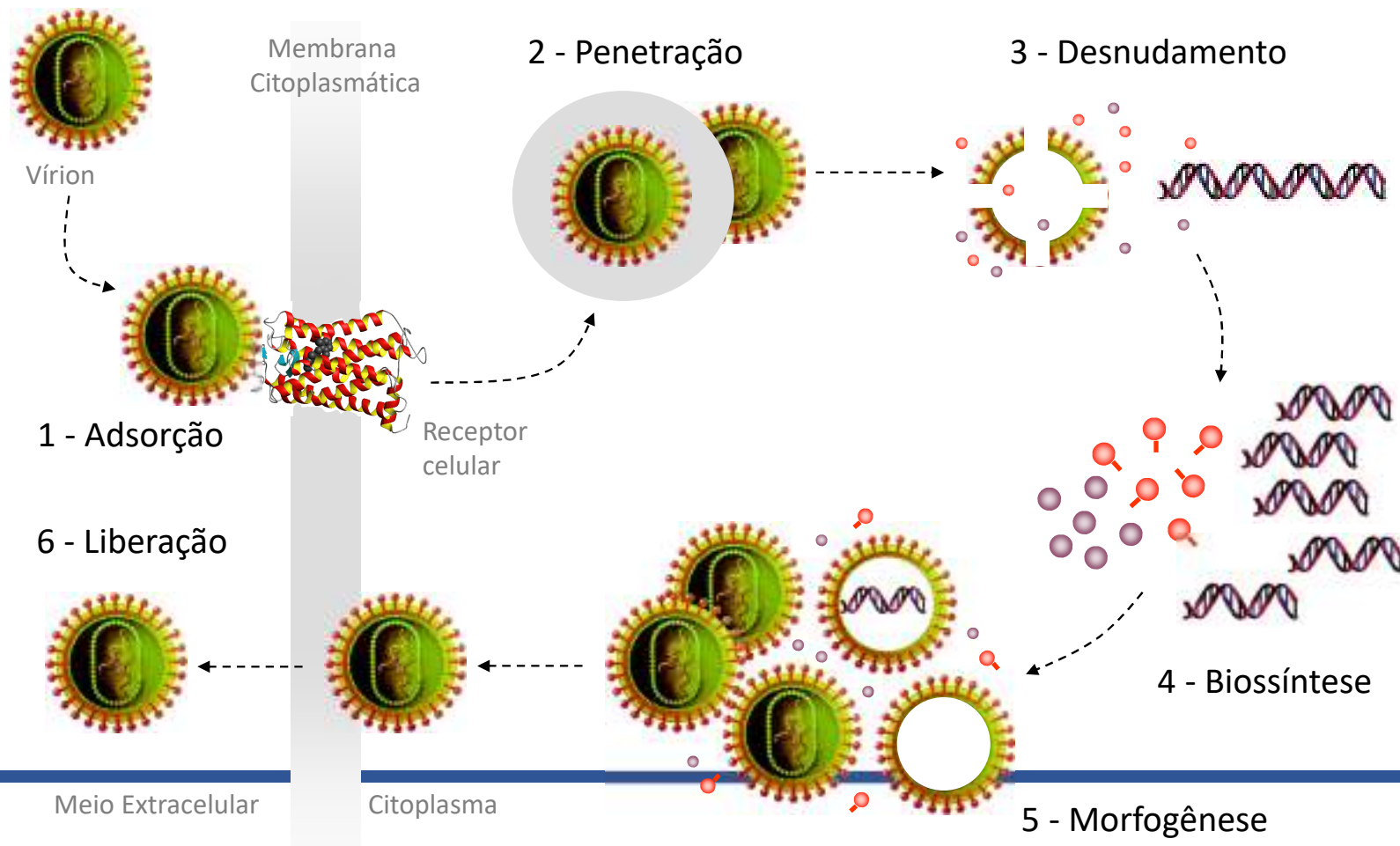
# Replicação viral – 6 fases

---

- Adsorção: ligação com os receptores
- Penetração: endocitose ou fusão
- Desnudamento ou Descapsidação
- Biossíntese: ácido nucléico + proteínas virais
- Morfogênese ou montagem das partículas virais
- Liberção (lise ou brotamento)



# Replicação Viral - 6 fases



---

# Taxonomia viral





# Caracteres discriminatórios entre diferentes espécies virais

Similaridade de seqüência genômica

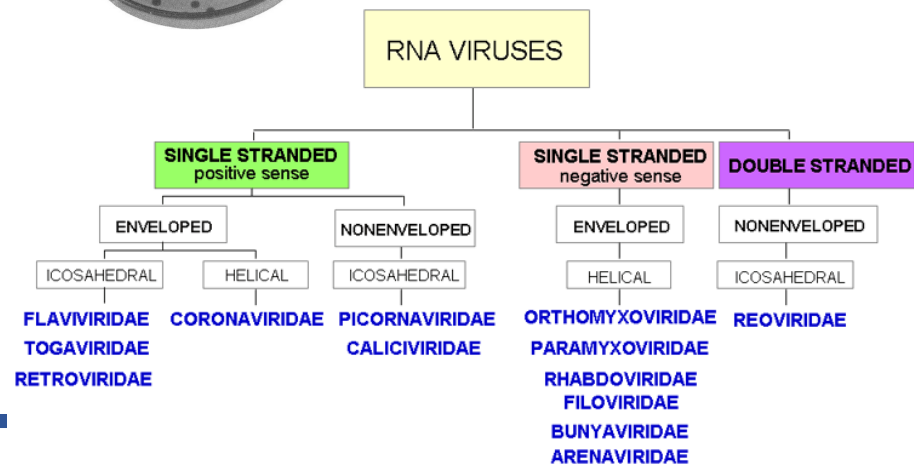
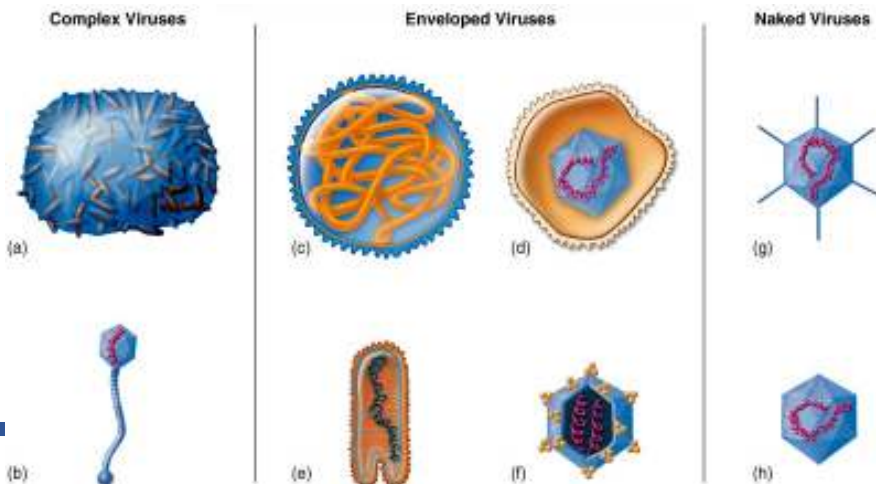
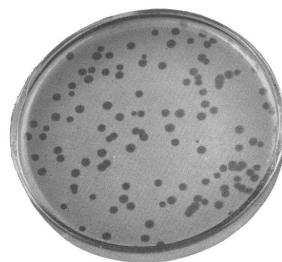
Hospedeiros naturais (amplitude)

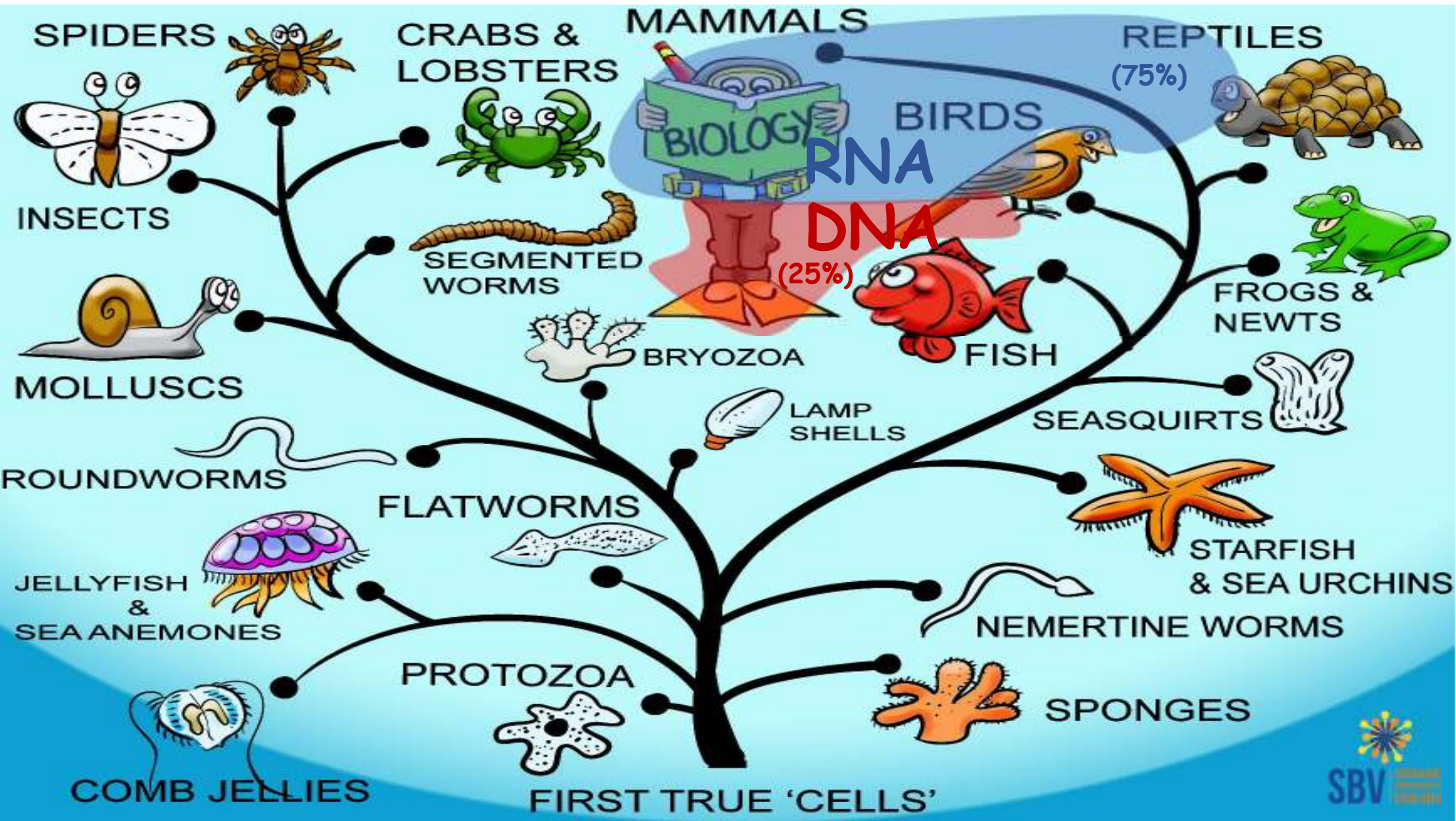
Tropismo tecidual e celular

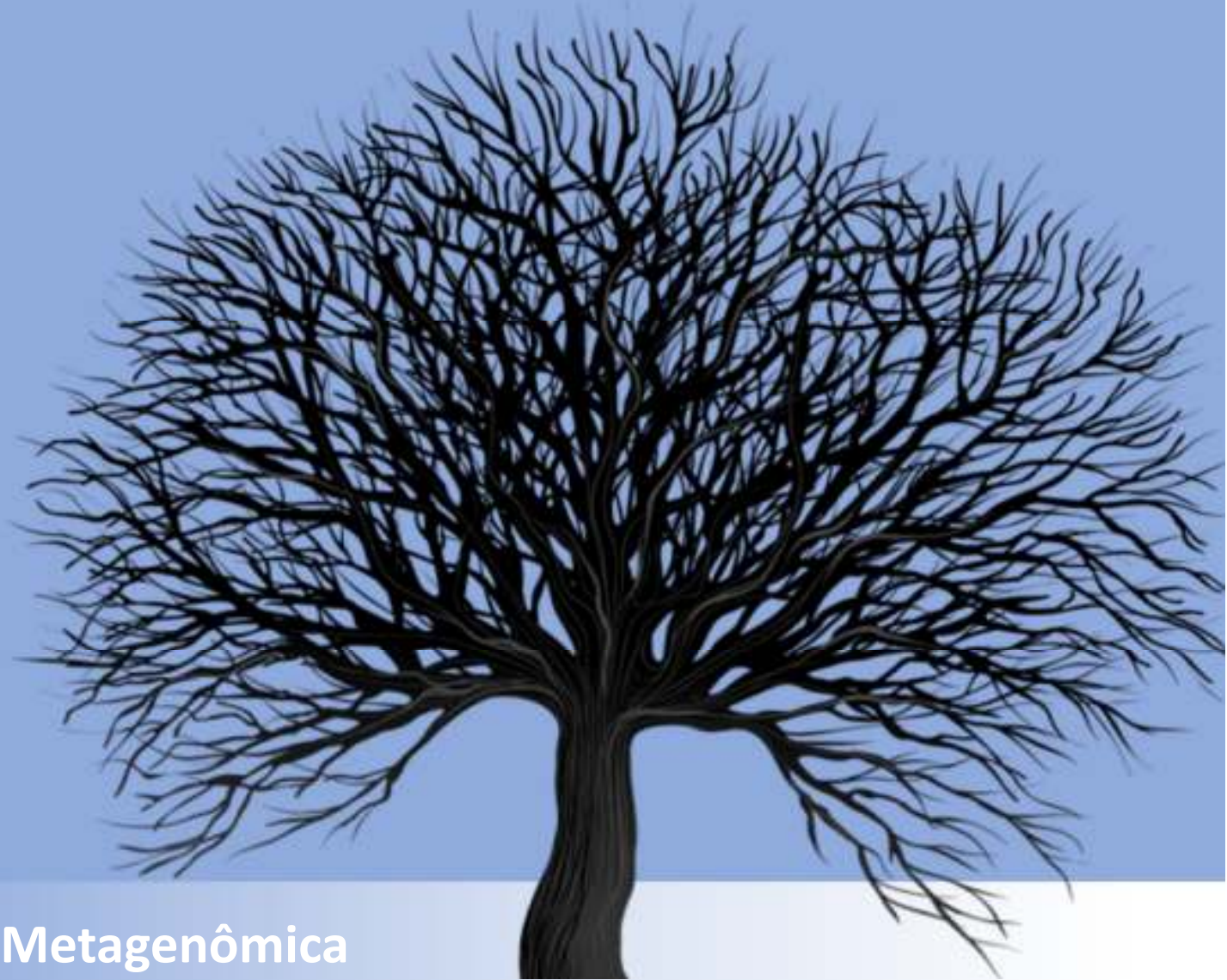
Patogenicidade e citopatogenicidade

Propriedades físico-químicas dos vírions

Propriedades antigênicas dos vírions







1ª Revolução Metagenômica  
DNA-Seq

---

**“The number of virus particles on Earth is on the order of  $10^{31}$ ”**

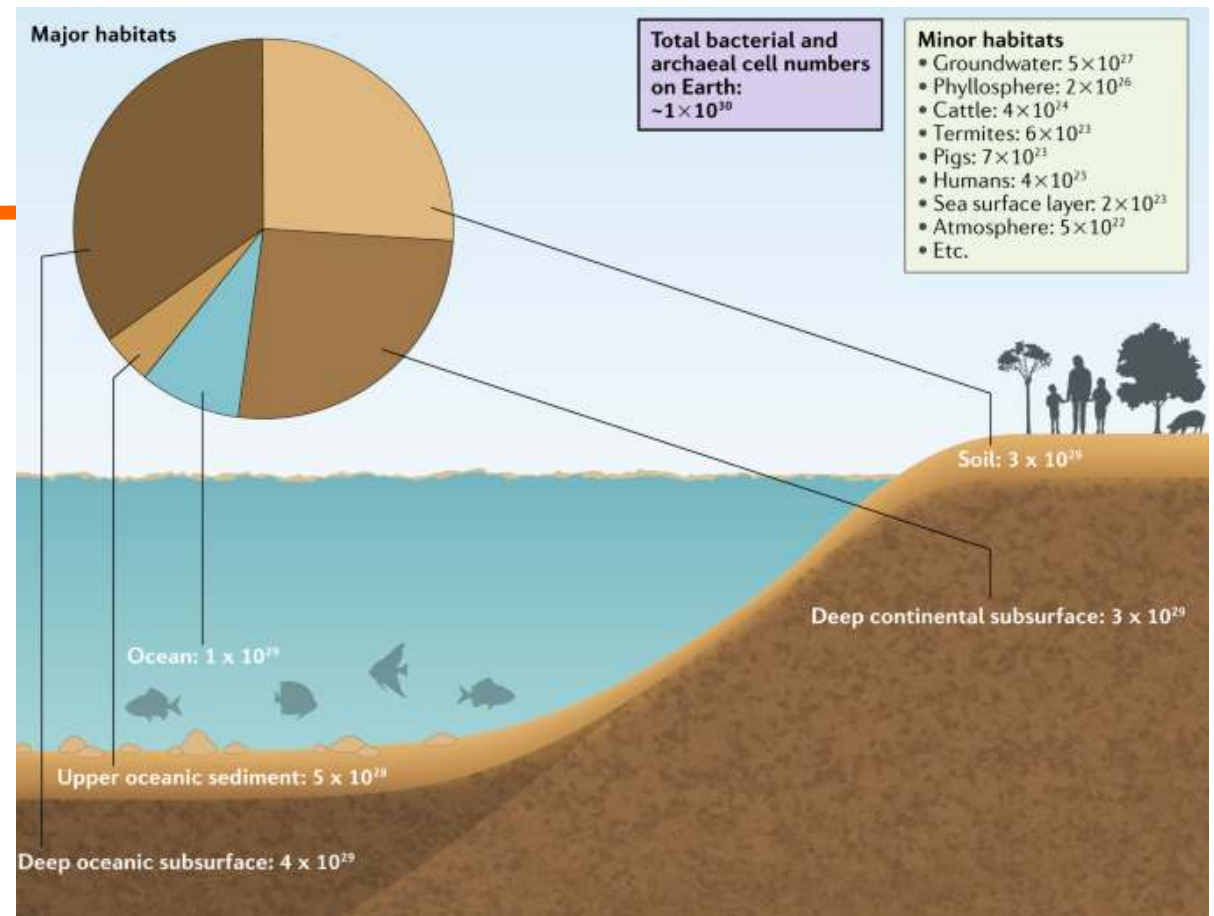
**Hendrix et al., 1999**

R. W. Hendrix, M. C. Smith, R. N. Burns, M. E. Ford, and G. F. Hatfull, Proc Natl Acad Sci U S A  
96:2192–2197, 1999



## Vírus em Bactérias

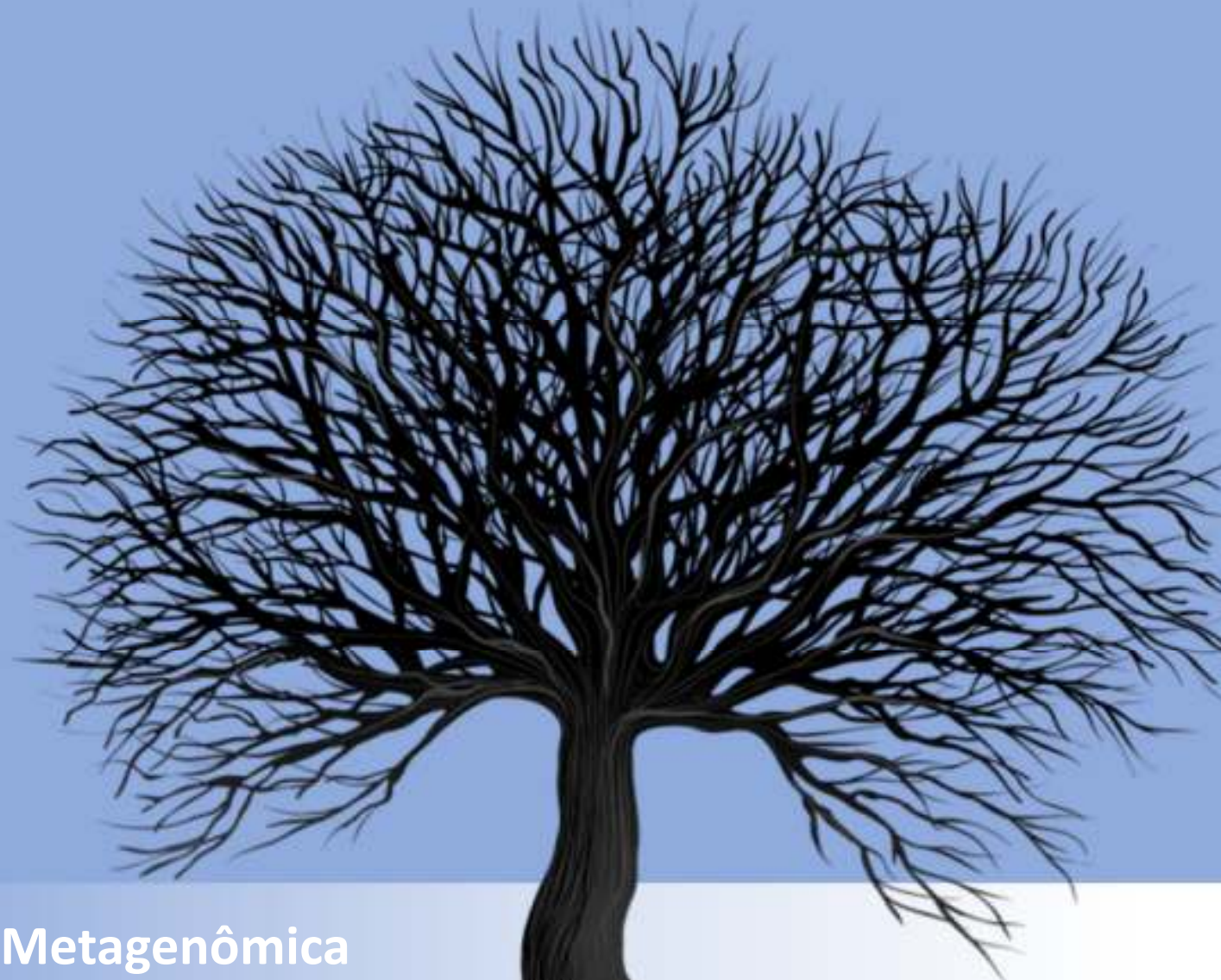
ssRNA+  
ssDNA  
dsDNA



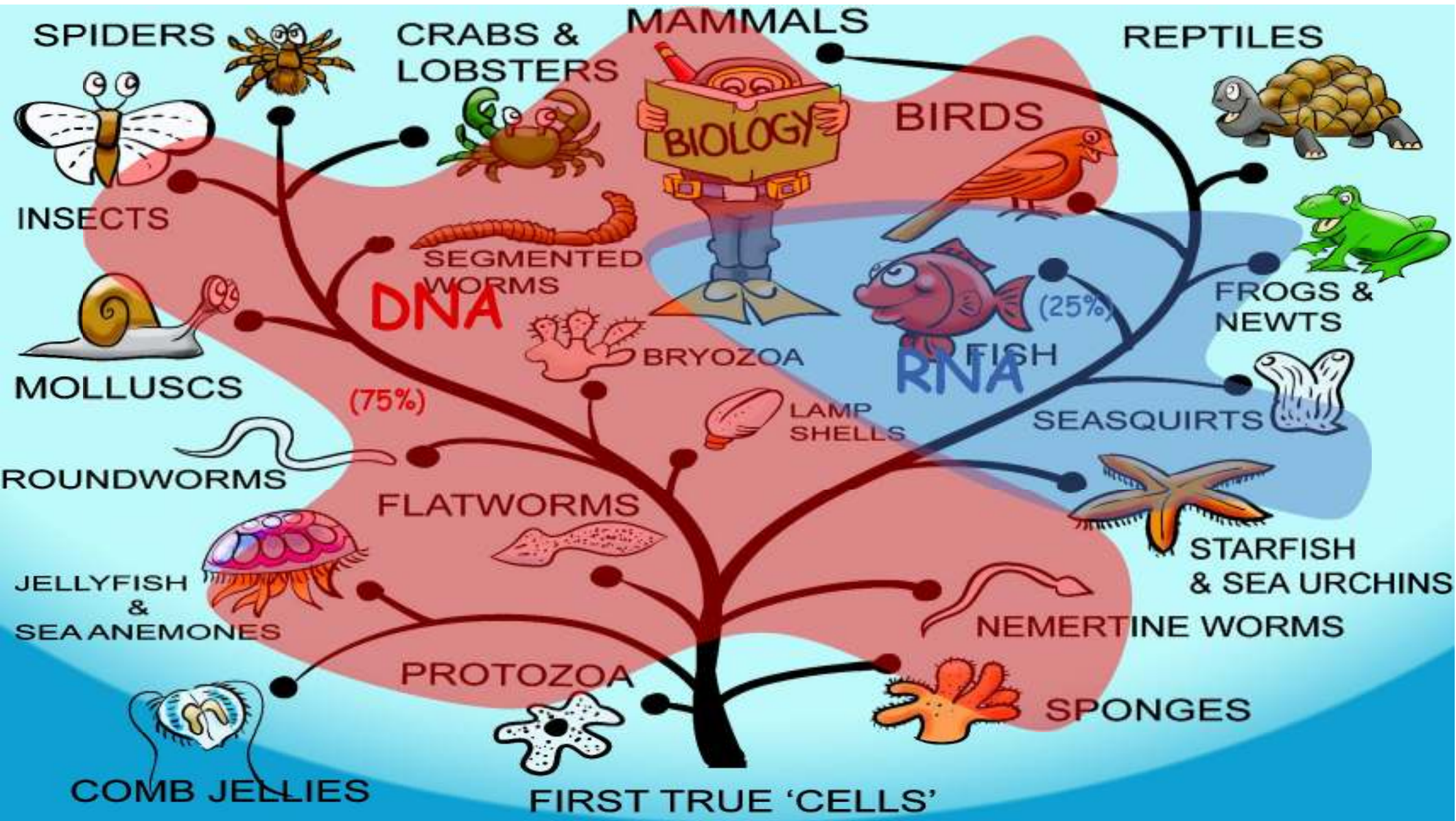
“Because phage particles are typically present in five- to 10-fold excess over bacterial cells, it is plausible that **tailed phages** are not only the most abundant organisms on Earth but, in fact, constitute an absolute majority of organisms.”

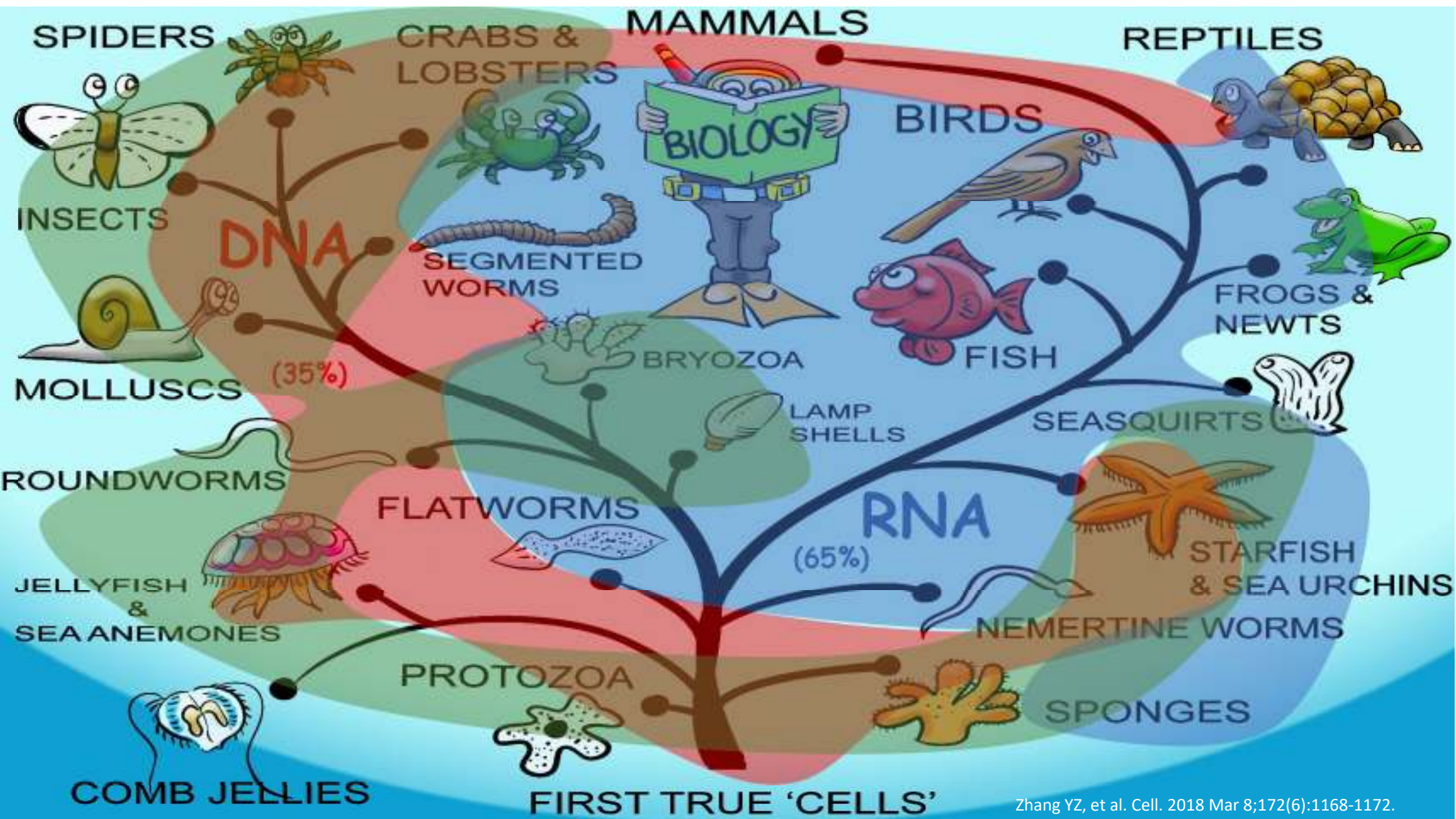
Nasir et al. Front Microbiol. 2014; 5:194 / Hendrix et al. Proc Natl Acad Sci U S A 96:2192–2197, 1999.



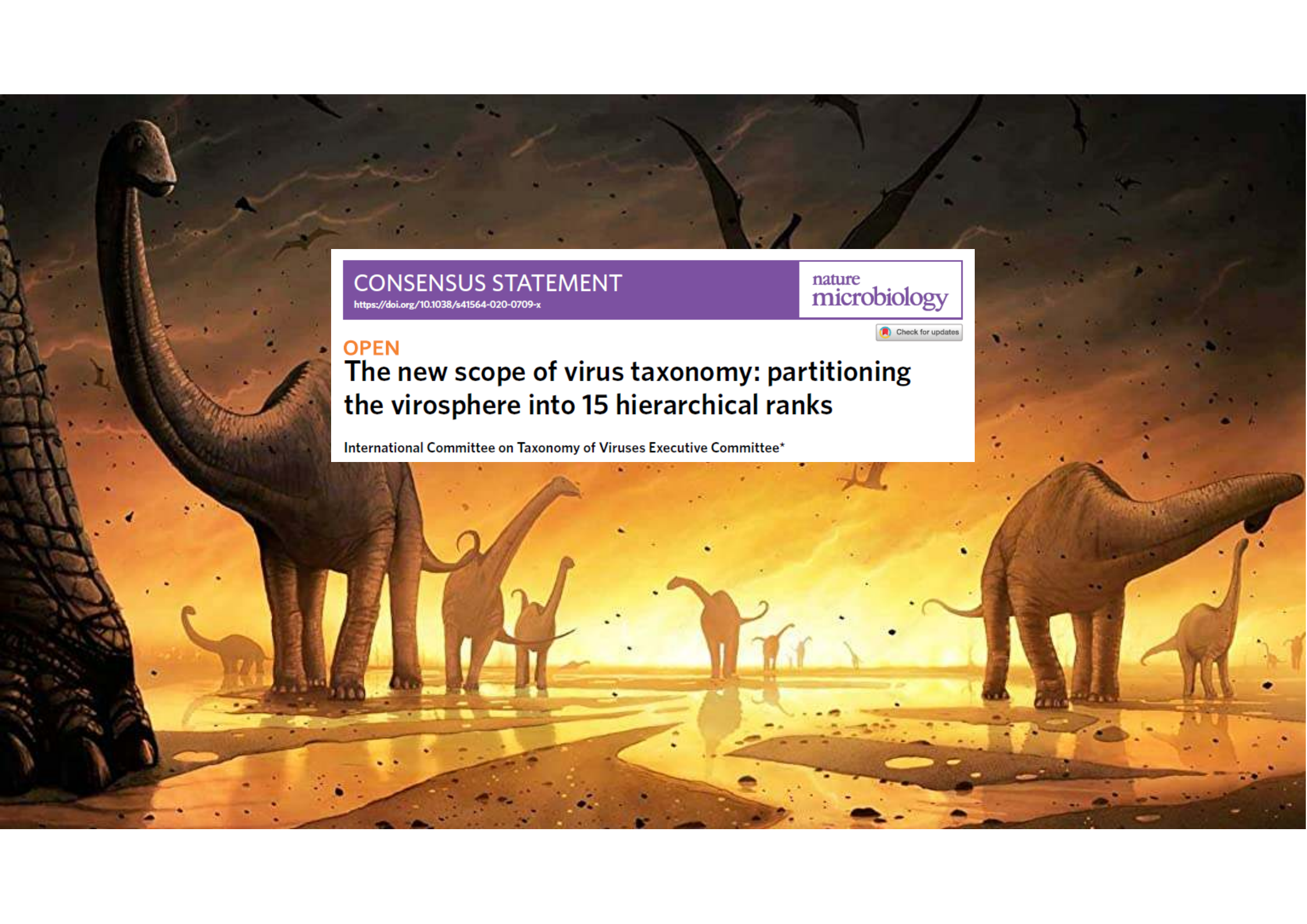


2ª Revolução Metagenômica  
RNA-Seq









CONSENSUS STATEMENT

<https://doi.org/10.1038/s41564-020-0709-x>

nature  
microbiology



OPEN

## The new scope of virus taxonomy: partitioning the virosphere into 15 hierarchical ranks

International Committee on Taxonomy of Viruses Executive Committee\*

# Base de Dados para a Taxonomia Viral

---

- International Committee on Taxonomy ICTV



É órgão Internacional que governa a nomenclatura e a relação taxonômica de todos os vírus

→ Archaea  
→ Bactérias  
→ Fungos  
→ Algas  
→ Plantas  
→ Animais

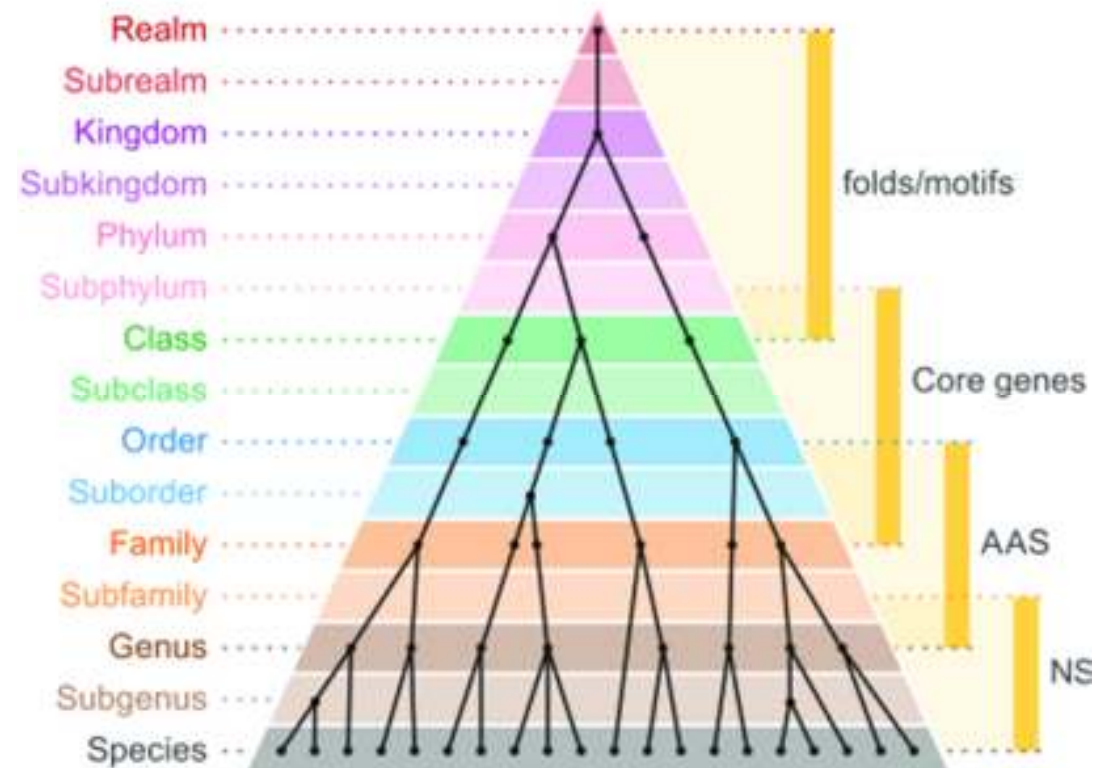
Até 2015: Apenas de Ordem a Espécie

Depois de 2015: de Domínio a Espécie

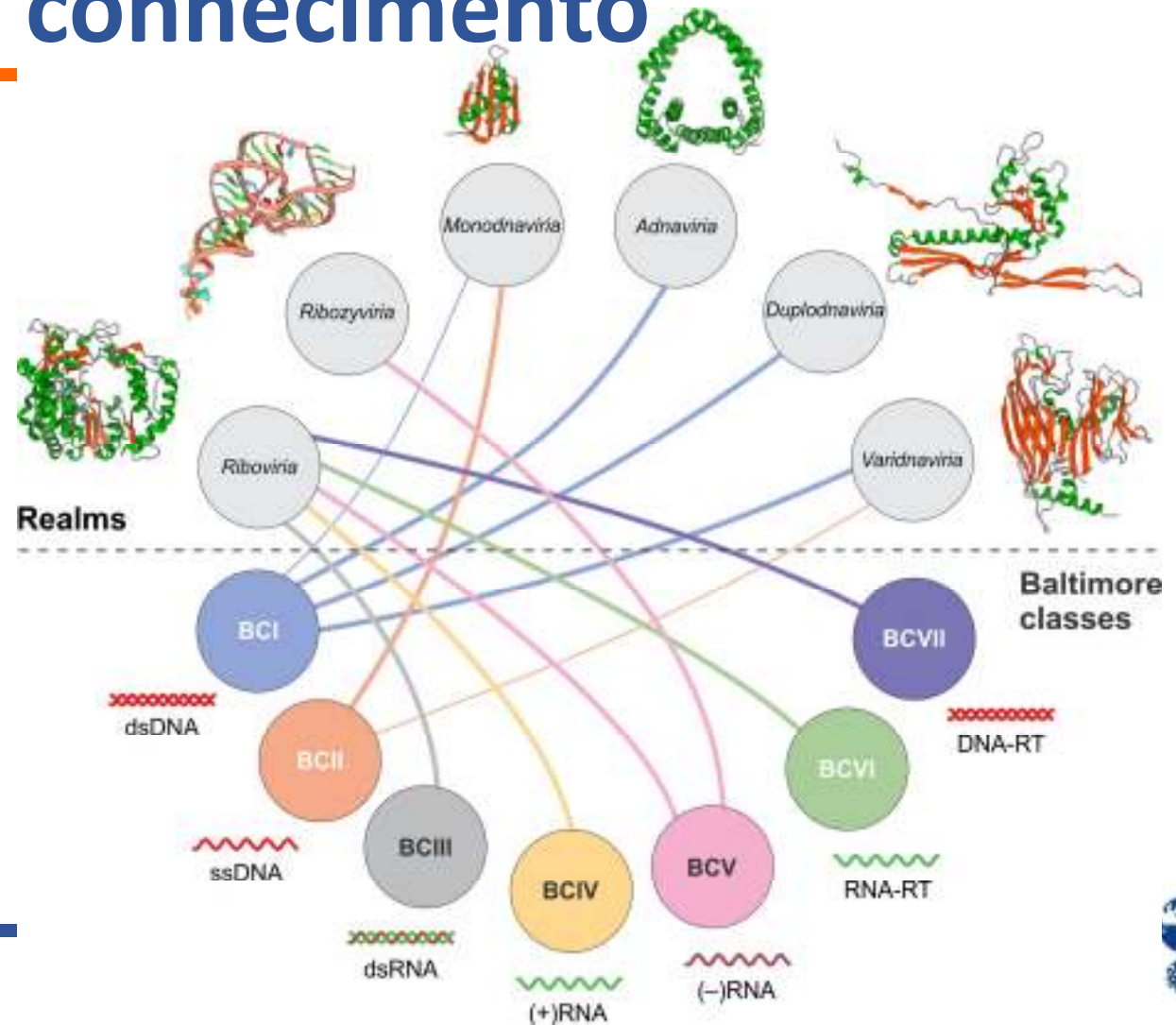


# ICTV

- **Princípio 1.** A taxonomia de vírus deve refletir a história evolutiva dos vírus
- **Princípio 2.** As propriedades do vírus podem orientar a atribuição de classificações para maximizar sua utilidade
- **Princípio 3.** Uma taxonomia evolutiva é apenas um dos muitos meios possíveis para classificar vírus
- **Princípio 4.** Atribuições taxonômicas de vírus inferidos a partir de sequências metagenômicas requerem rigoroso controle de qualidade de sequência



# A evolução do conhecimento





# Taxonomia Viral

- **Realm:** *Riboviria* i
- **Kingdom:** *Orthornavirae* Realm: *Riboviria* i
- + **Phylum:** *Duplornaviricota* Kingdom: *Orthornavirae* i
- + **Phylum:** *Kitrioviricota* Kingdom: *Orthornavirae* i
- + **Phylum:** *Lenarviricota* Kingdom: *Orthornavirae* i
- **Phylum:** *Negarnaviricota* Kingdom: *Orthornavirae* i
- + **Subphylum:** *Haploviricotina* Phylum: *Negarnaviricota* i
- **Subphylum:** *Polyploviricotina* Phylum: *Negarnaviricota* i
- + **Class:** *Ellioviricetes* Subphylum: *Polyploviricotina* i
- **Class:** *Insthoviricetes* Subphylum: *Polyploviricotina* i
- **Order:** *Articulavirales* Class: *Insthoviricetes* i
- + **Family:** *Amnoonviridae* Order: *Articulavirales* i
- **Family:** *Orthomyxoviridae* Order: *Articulavirales* i
- **Genus:** *Alphainfluenzavirus* Family: *Orthomyxoviridae* i
- **Species:** *Alphainfluenzavirus influenzae* Genus: *Alphainfluenzavirus* i

Ex: Vírus da Influenza



---

# Como identificar os vírus?



# Isolamento viral

---

## 1. Cultivo celular

Efeitos citopáticos (CPE)  
Hemabsorção  
Imunodeteção

## 2. Ovos embrionados

Lesões na CAM  
Hemaglutinação  
Corpúsculos de inclusão

## 3. Animais

Doença ou morte



# Isolamento viral

---

- Inoculação em animais
- Inoculação em ovos embrionados
- Cultivos celulares
  - 3 tipos
  - Métodos imunológicos





# Isolamento viral em ovos embrionados

---



# Isolamento viral em ovos embrionados

---



# Isolamento viral em cultivo celular



# Isolamento viral em cultivo celular

---

- Cultivos primários (células diplóides)
- Linhas celulares
  - Cultivo semi contínuo
    - Diplóide, 40 passagens
  - Cultivo contínuo
    - Número de passagens indefinido



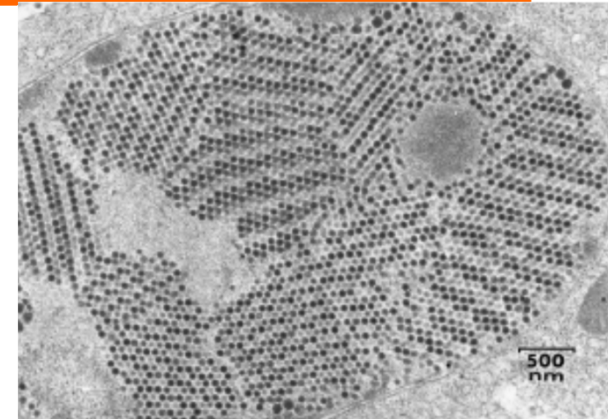
# Isolamento viral em cultivo celular

---

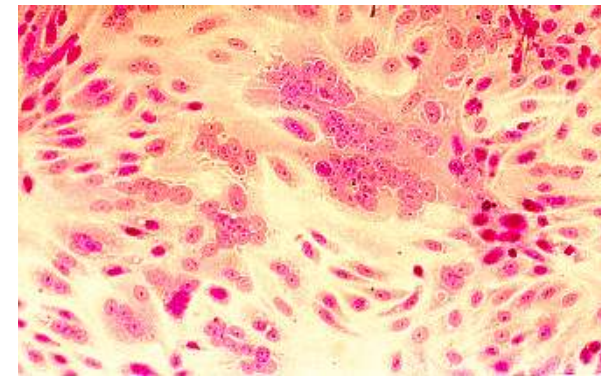
- EFEITO CITOPÁTICO
- É o dano que o vírus causa à célula !
  - Lise
  - Arredondamento
  - Vacuolização
  - Formação de sincícios
  - Inclusões
  - Picnose
  - Apoptose



# Isolamento viral em cultivo celular



Adenovirus – Células de Fígado



Formação de Sincícios



# Isolamento viral em animais

---



# Detecção do agente (direto)

---

- 1. Microscopia eletrônica de transmissão ou varredura** morfologia dos agentes
- 2. Detecção do antígeno** imunofluorescência, ELISA, IHC, teste rápido, etc.
- 3. Microscopia de luz**  
Histopatologia  
Corpúsculo de inclusão
- 4. Detecção do genoma** Técnicas moleculares, hibridização com sondas específicas de ácidos nucleicos PCR, RT-PCR





# Microscopia Eletrônica

Light Microscopy

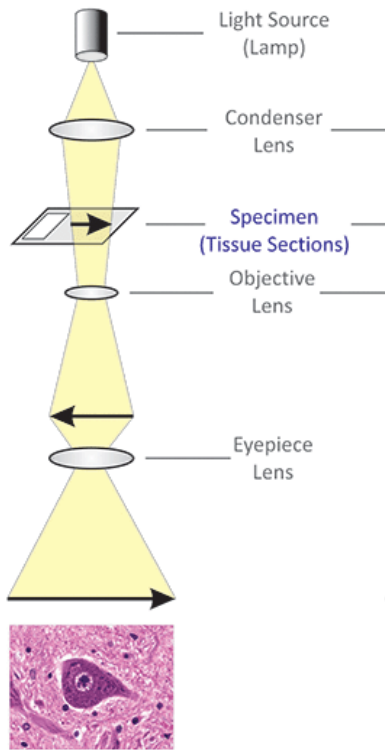
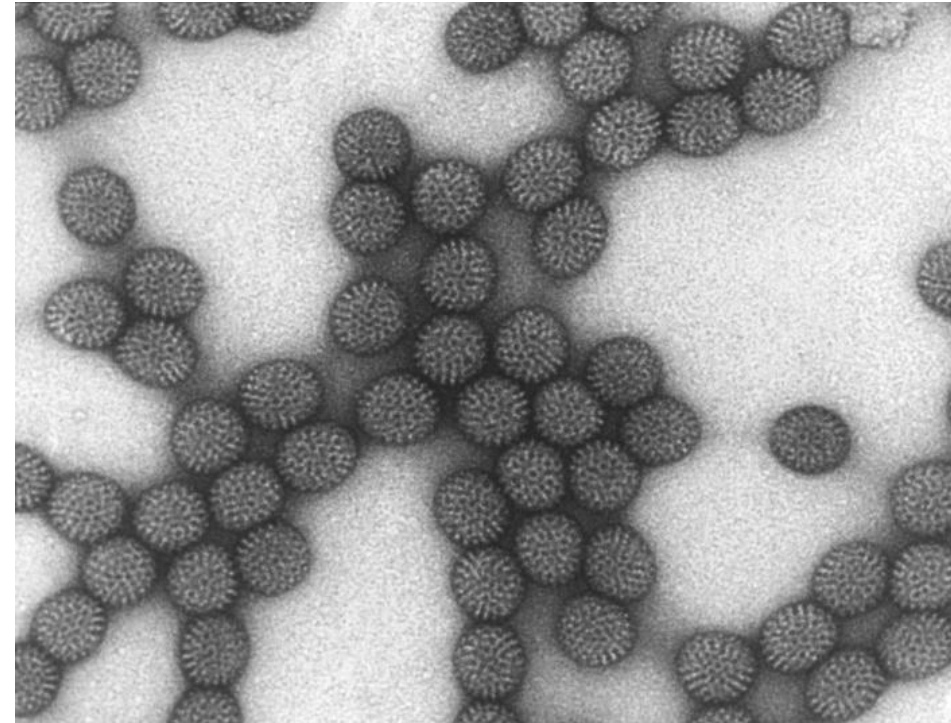
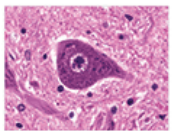


Image Viewed Directly



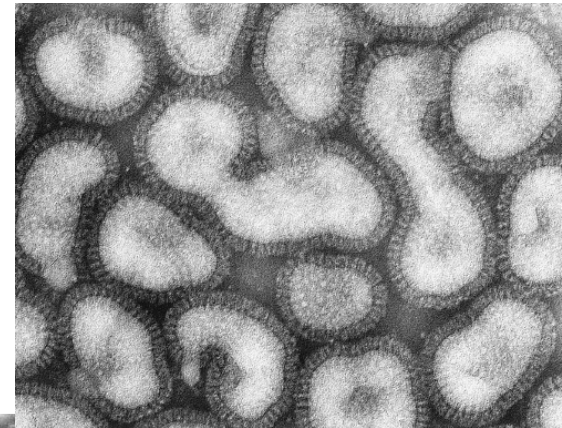
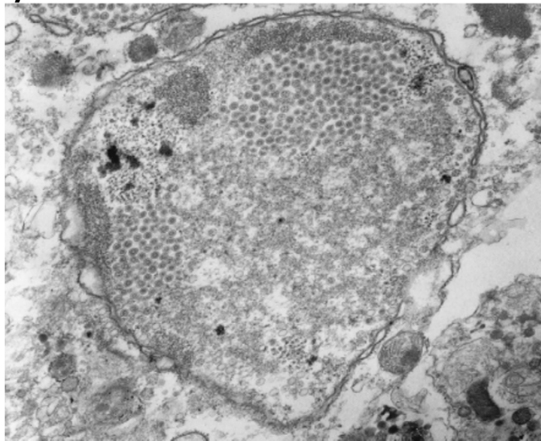
Photos of Rotavirus | CDC



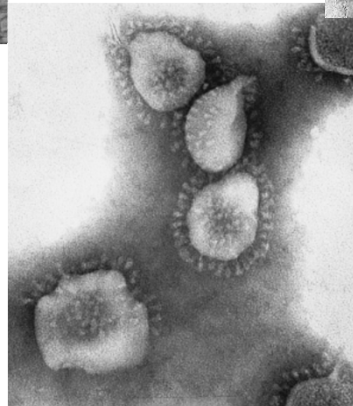
# Microscopia eletrônica de transmissão

---

*Herpesvirus 1*



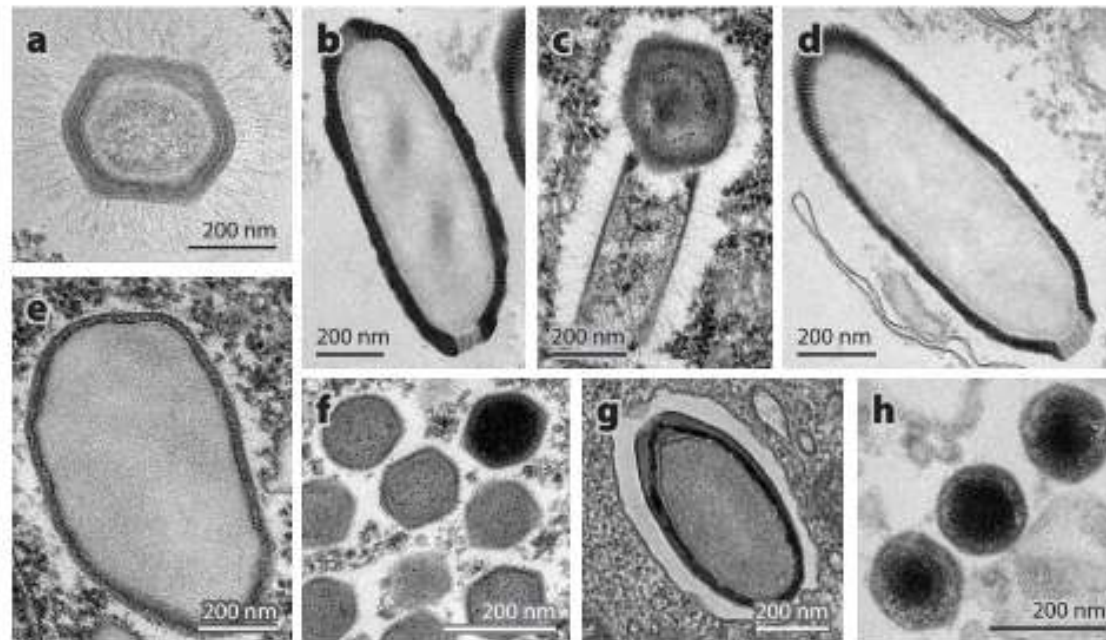
*Influenzavirus A*



*Coronavirus*



# Microscopia eletrônica de transmissão

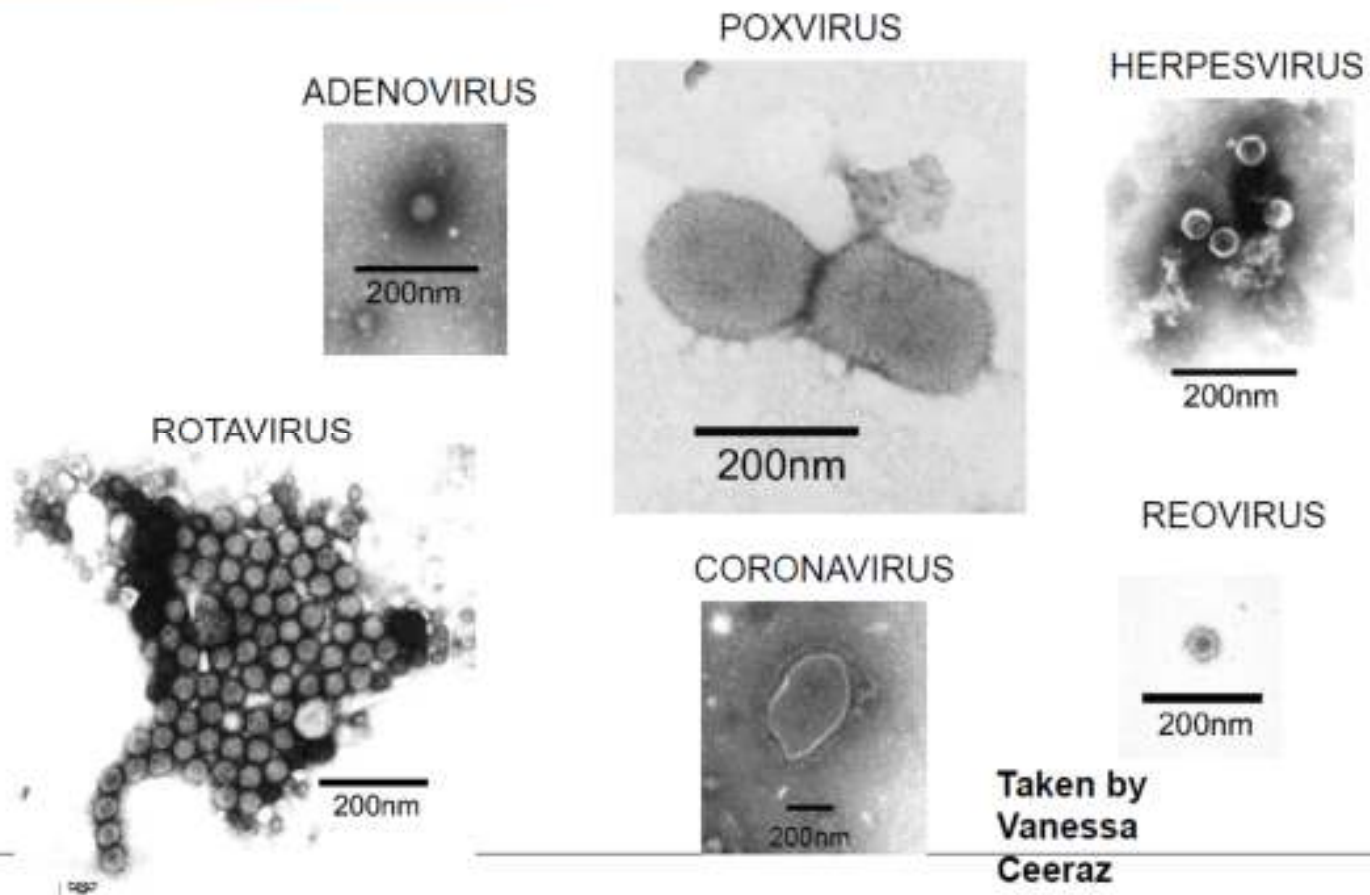


**Figure 1**

Diversity of amoebae-infecting viruses. Electron microscopy images evidence the structural diversity and complexity of viruses isolated from different amoebae hosts: (a) mimivirus, (b) cedratvirus, (c) tupanvirus, (d) pithovirus, (e) orpheovirus, (f) marseillevirus, (g) pandoravirus, and (h) kaumoebavirus.

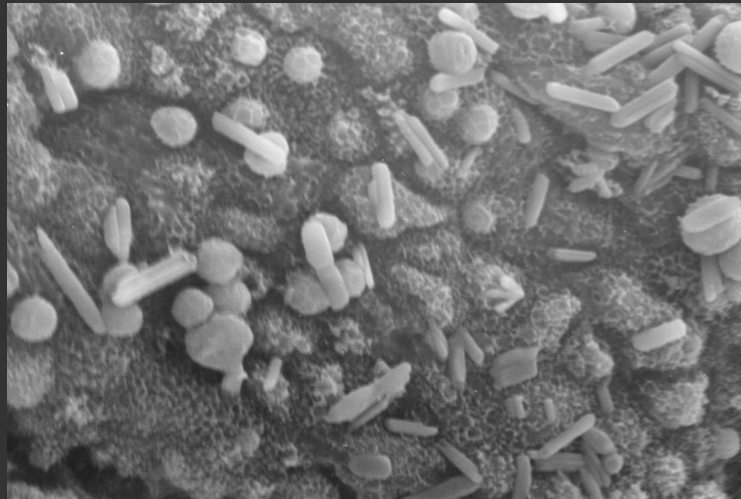


# Microscopia eletrônica de transmissão



# Microscopia eletrônica de varredura

---



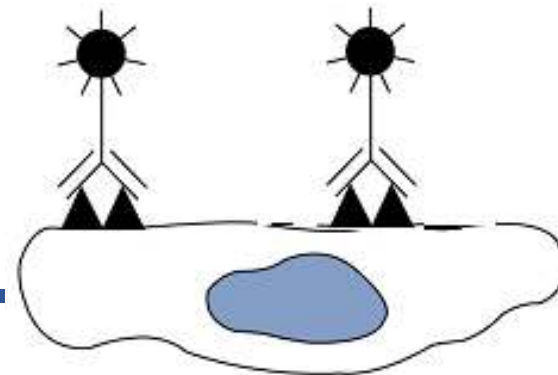
Traquéia mostrando colonização epitelial por *E. coli* e *C. baileyi* com perda severa de cílios. Grupo II, 13 dias de idade, 2000 X.



# Imunohistoquímica e imunofluorescência

---

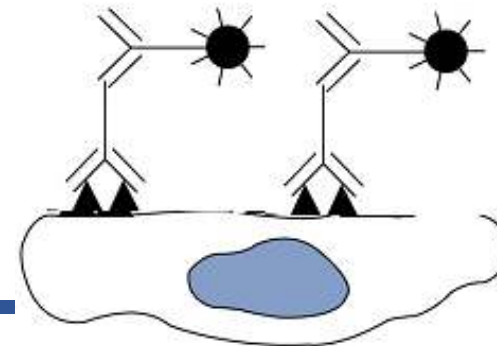
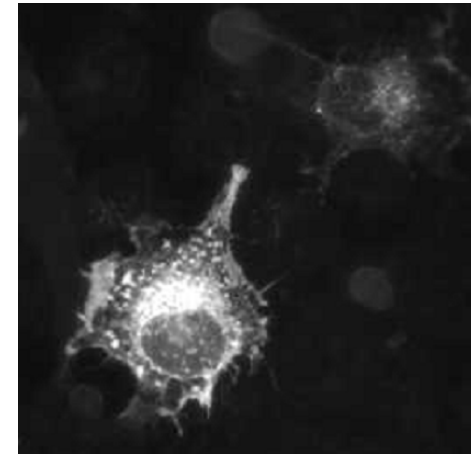
- **Direta**
  - Anticorpo marcado sobre material infectado
- Material infectado (suspeito)
- Anticorpo específico marcado com FITC ou Peroxidase
- Luz UV ou luz branca



# Imunohistoquímica e imunofluorescência

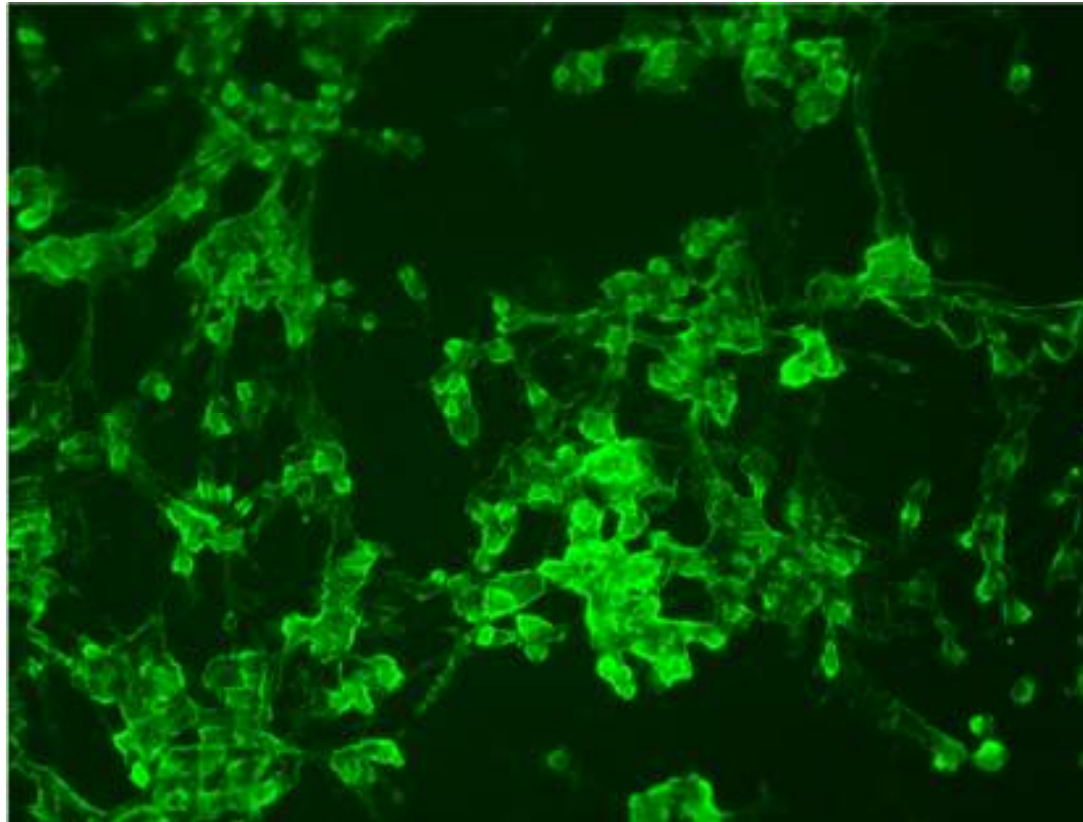
- **Indireta**

- Utilização de dois anticorpos
- Material infectado (células ou tecidos)
- Soro específico (ex: soro galinha)
- Conjugado anti-IgG específico (ex. anti-IgG galinha/FITC ou Peroxidase)
- Luz UV ou Luz branca



# Identificação viral

---



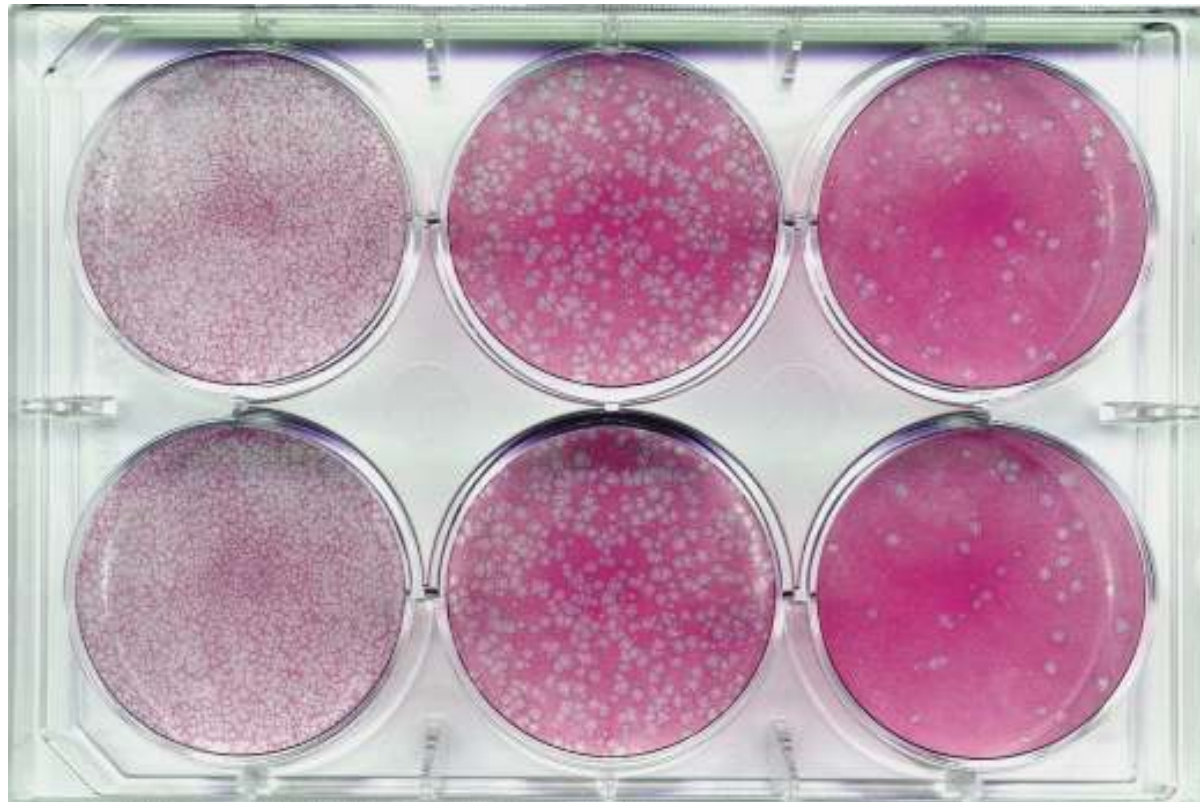
IFA – VERO infectadas com NDV





# Identificação viral

---

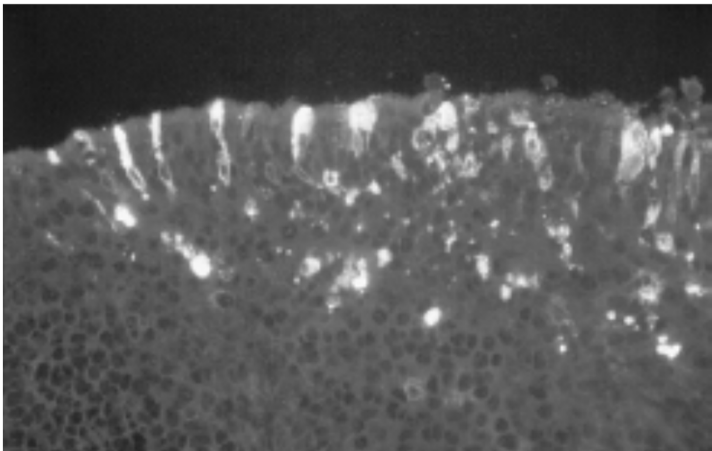


PFU

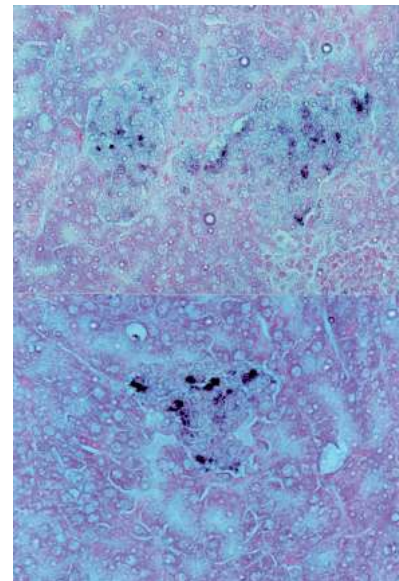


# Imunohistoquímica e imunofluorescência

---



Coronavirus - Intestino



Adenovirus - Pâncreas



# Testes rápidos de detecção

---



Positivo

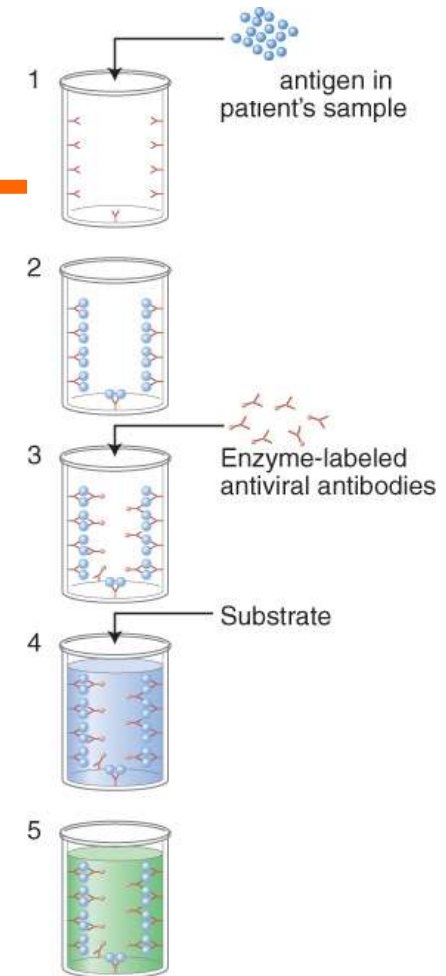
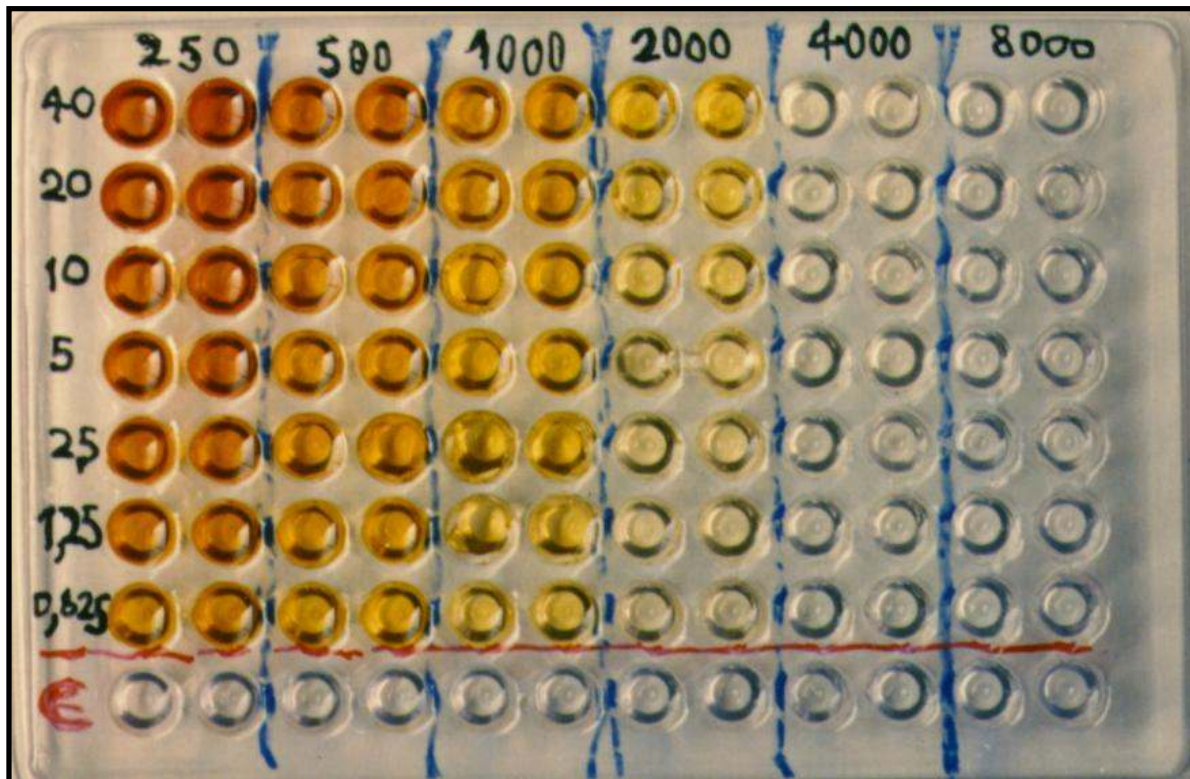


Negativo

<http://www.cfsph.iastate.edu/video.php?link=antigen-detection>



# ELISA (DIRETO)

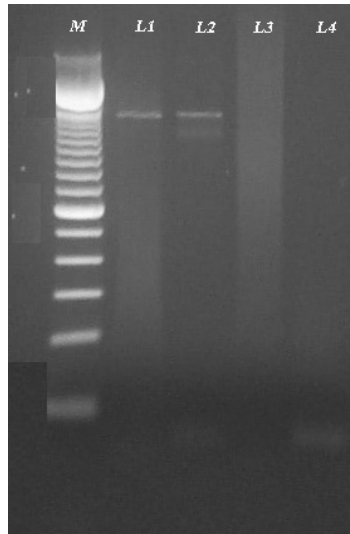


<http://www.cfsph.iastate.edu/video.php?link=idexx-elisa>

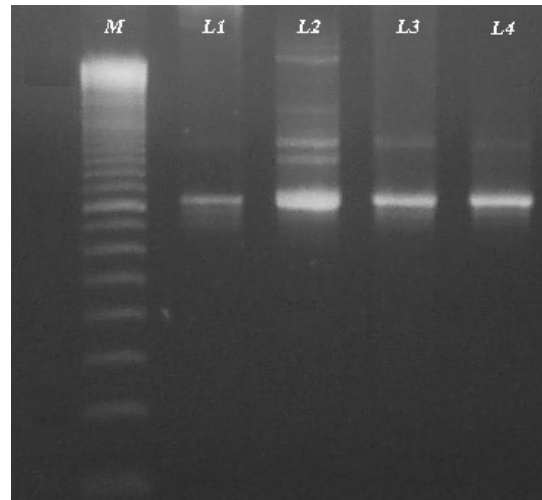


# Biologia Molecular

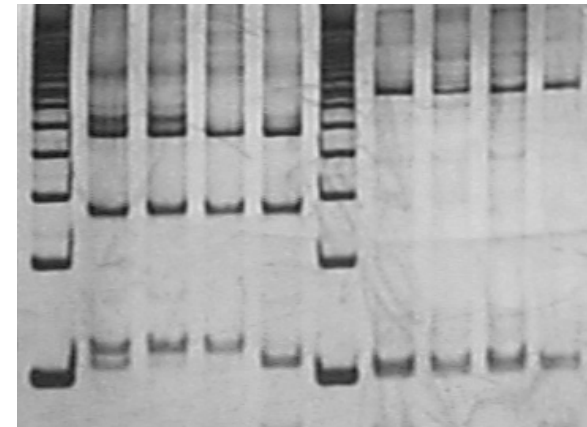
---



PCR



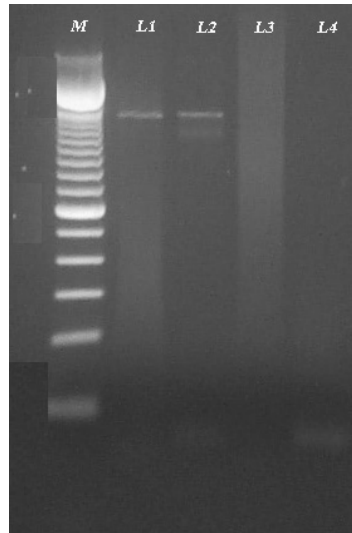
RT-PCR



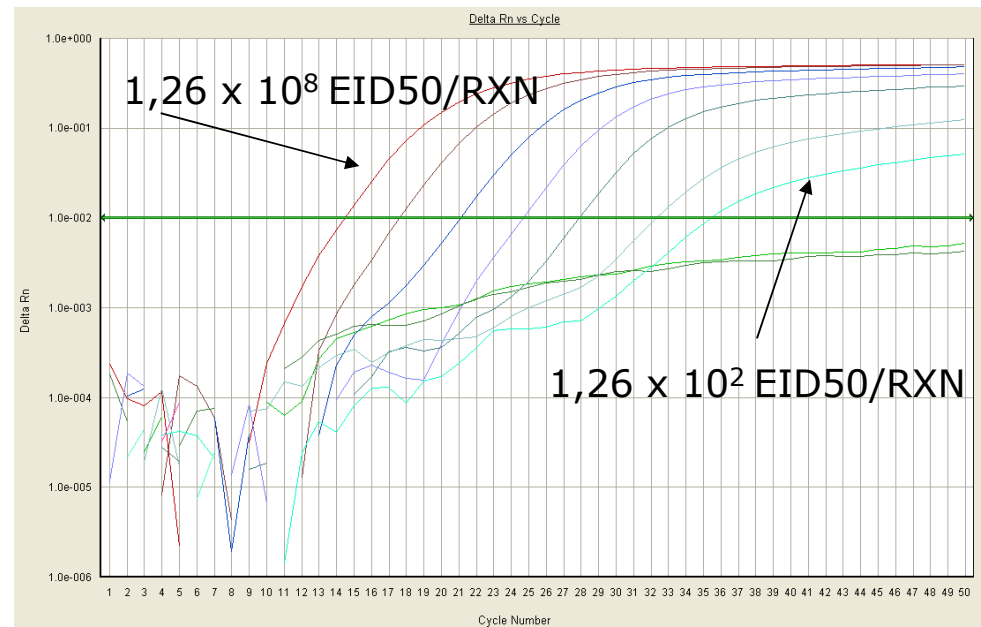
PCR-RFLP



# Biologia Molecular



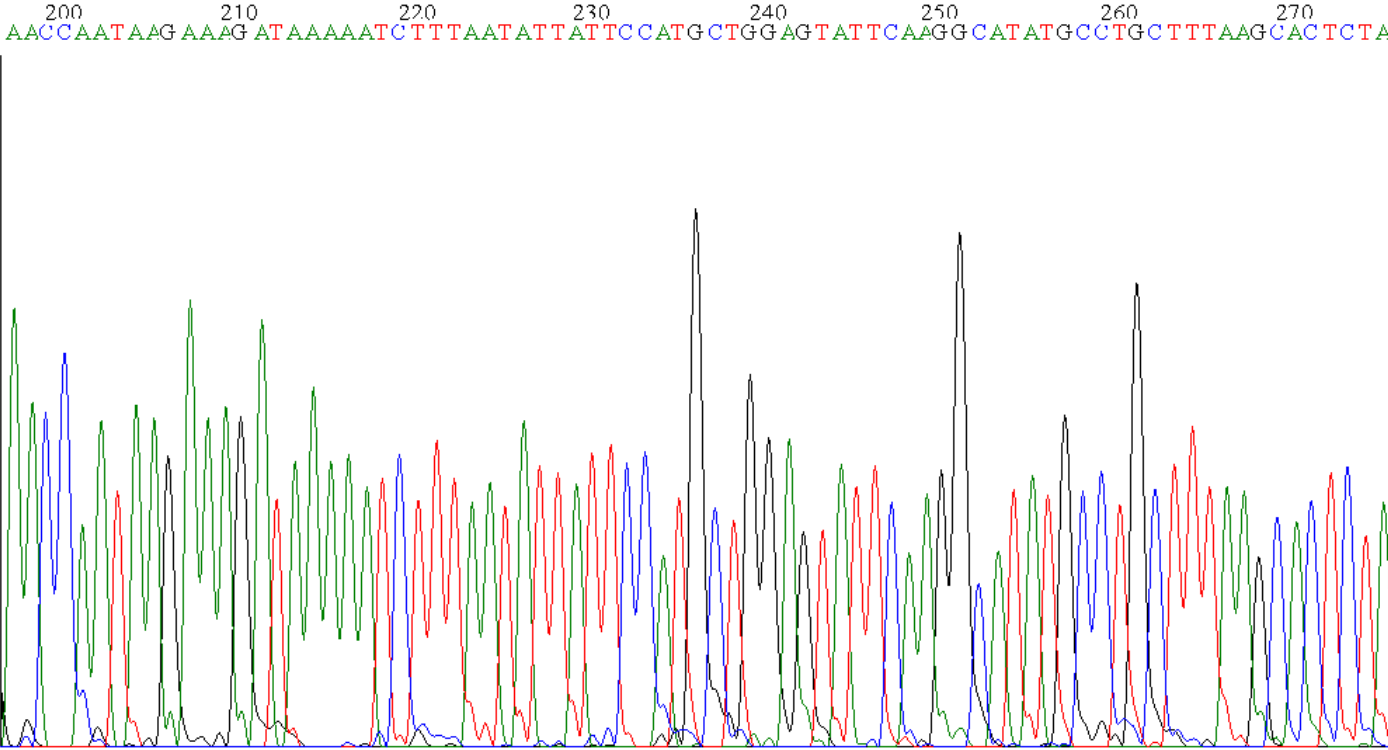
RT- PCR



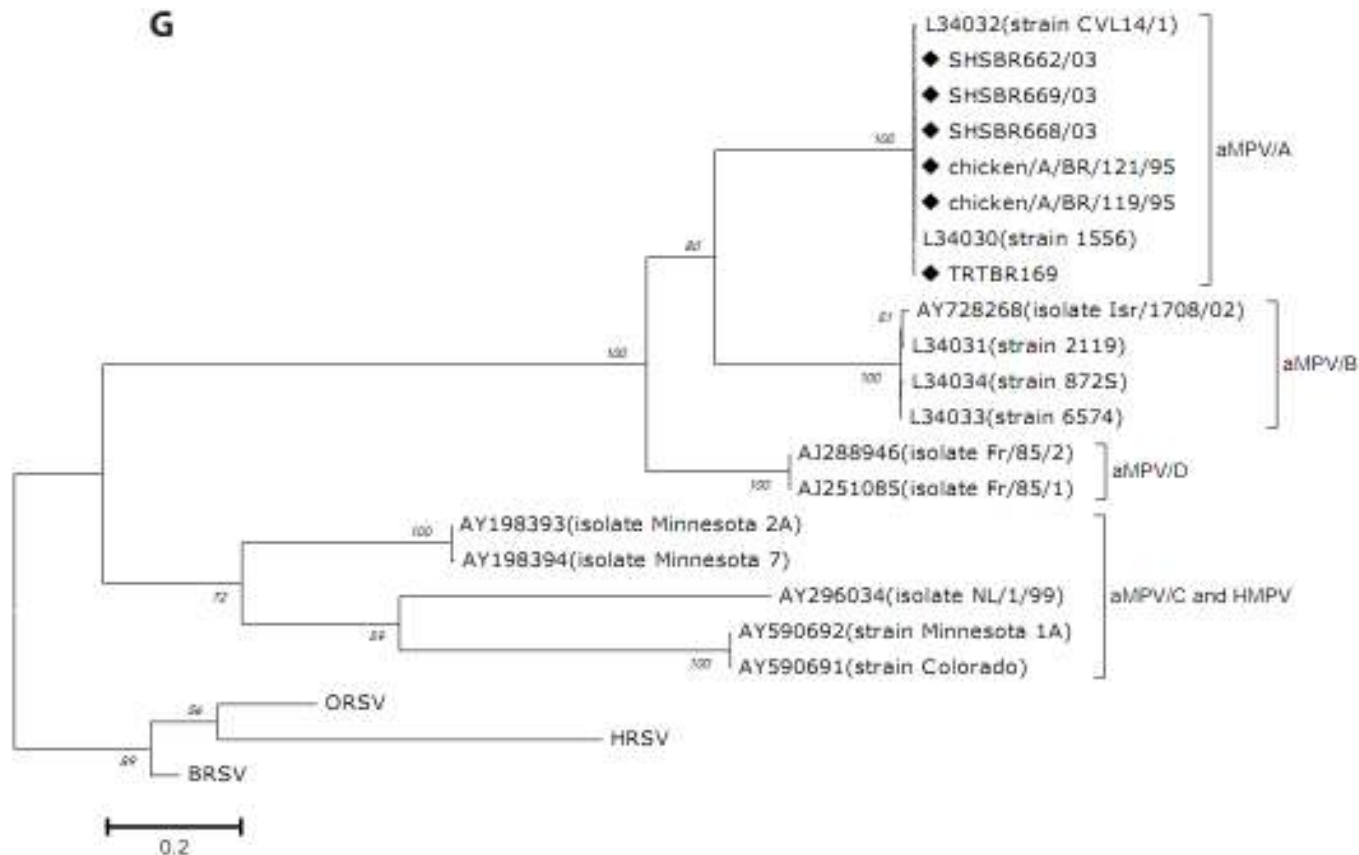
RT-qPCR



# Sequenciamento



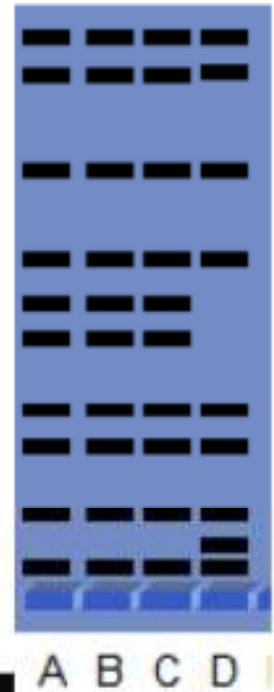
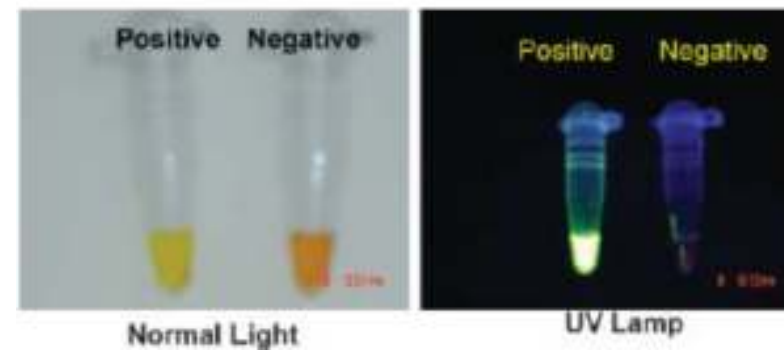
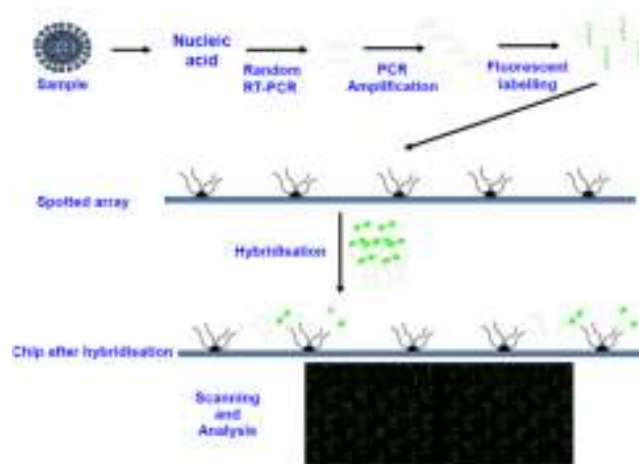
# Epidemiologia molecular



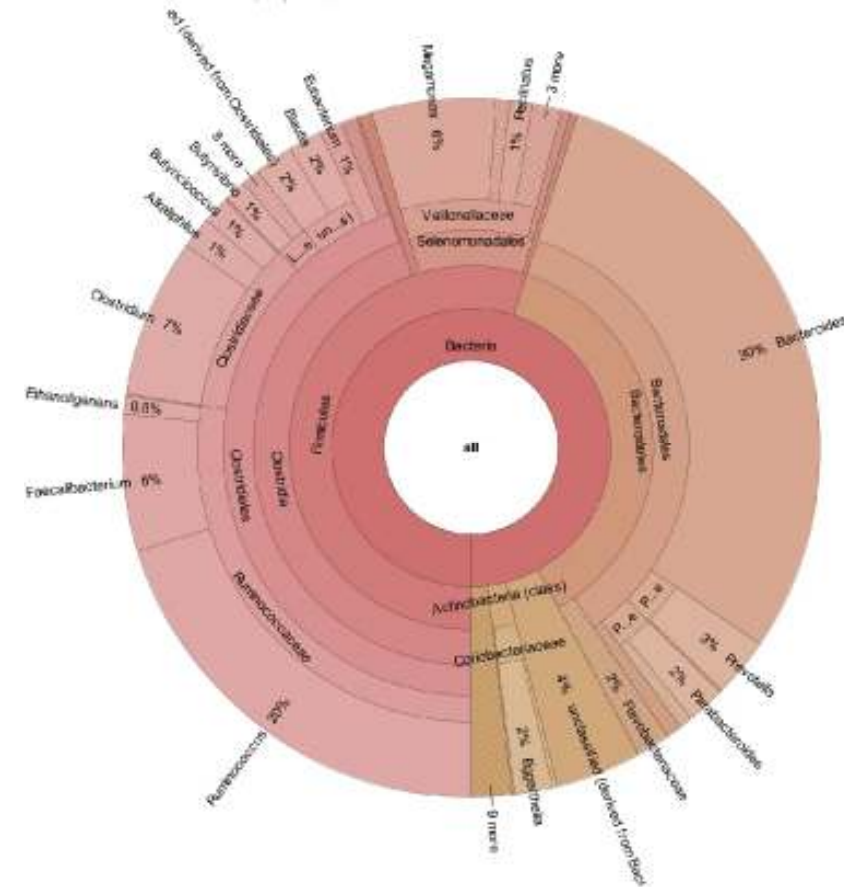


# Outras técnicas moleculares

- DNA fingerprint
- LAMP
- Microarray



# Metagenômica



---

# Importância dos vírus e os alimentos



# Vírus de origem alimentar

---

- Apenas algumas partículas são necessárias para produzir doenças;
- Um elevado número de partículas virais é derramado nas fezes de pessoas infectadas (até 10<sup>11</sup> partículas por grama de fezes relatadas para rotavírus);
- Os vírus precisam de células vivas específicas para se replicar e, portanto, não podem fazê-lo em alimentos ou água;
- Os vírus de origem alimentar normalmente são bastante estáveis fora do hospedeiro e são resistentes ao ácido.



# Vírus transmitidos por alimentos ou água

Likelihood of food- or waterborne transmission of enterically transmittable viruses, according to the type of illness associated with infection

| Likelihood of food- or waterborne transmission | Illness   |                                |                          |
|--|---|--------------------------------|--------------------------|
|  | Gastroenteritis   | Hepatitis                      | Other                    |
| Common   | Norovirus   | Hepatitis A virus              |                          |
| Occasionally                                   | Enteric adenovirus (types 40/41)<br>Rotavirus (group A–C)<br>Sapovirus<br>Astrovirus<br>Coronavirus<br>Aichivirus | Hepatitis E virus (waterborne) | Enterovirus <sup>a</sup> |

<sup>a</sup>Enteroviruses (e.g. poliovirus) are associated with a range of symptoms, including neurological symptoms.



# Vírus nos produtos de animais

Beato *et al.*

**Table 1.** Summary of data available in the literature on the presence of HPAI viruses in poultry commodities

| Commodity | Species                               | Strain  | Experimental (E) or natural (N) infection | Infecting dose (EID <sub>50</sub> log <sub>10</sub> ) | Titres (log <sub>10</sub> EID <sub>50</sub> /g) detected in commodity | Reference (first author) |
|-----------|---------------------------------------|---|---|---|---|--------------------------|
| Meat      | Chickens                              | A/duck/Anyang/AVL-1/01 (H5N1)   |   | 6/0.1 ml  | 5.3 to 5.5  | Tumpey, Kishida, Swayne  |
|           |                                       | A/chicken/Pennsylvania/1370/1983 (H5N2)   | E   | 6/0.1 ml  | 2.2 to 3.2  |                          |
|           |                                       | A/tern/South Africa/61 (H5N3)   |   | 7/0.1 ml  | >4  |                          |
|           | Turkeys                               | A/turkey/Italy/4580/99 (H7N1)   | E   | 7/0.1 ml  | 4.38  | Toffan                   |
|           |                                       | A/duck/Anyang/AVL-1/01 (H5N1)   | E and N                                   | 6/0.1 ml  | 3 to 4  | Tumpey, Li               |
|           | Ducks                                 | A/goose/Vietnam/3/2005 (H5N1)   | E   | 6/0.1 ml  | 3   | Pantin-Jackwood          |
|           |                                       | A/Vietnam/1203/2004, A/ThailandPB/6231/2004, A/crow/Thailand/2004, A/Egret/HK/757.2/2002 (H5N1) | E   | 5/0.1 ml  | 4 to 6 (2-week-old birds)   |                          |
|           |                                       | A/egret/HK/757.2/2002 (H5N1)  | E   | 8/0.1 ml  | 2 to 4 (5-week-old birds)   |                          |
|           |                                       | A/chicken/Yamaguchi/7/2004 (H5N1)   | E   | 7/0.1 ml  | 1.5   |                          |
|           |                                       | A/duck/Vietnam/12/2007 (H5N1)   |   |   |   |                          |
| Eggs      | Turkeys                               | A/turkey/Ontario/7732/66 (H5N9)   | N   | Not reported  | Not reported  | Cappucci, Narayan        |
|           |                                       | H5N2 (Virginia/1985)  | E   | Not reported  | Not reported  |                          |
|           | Chickens                              | H5N2 (Virginia/1985)  | N   | Not reported  | Not reported  | Cappucci                 |
|           |                                       | Ducks and geese   | H5N1 (strain not reported)                | N   | Not reported  | Not reported             |
|           | Quail                                 | H5N1 (strain not reported)  | N   | Not reported  | 4.6 to 6.2  | Promkuntod               |
| Feathers  | Chickens, turkeys, quail, guinea fowl | A/chicken/Yamaguchi/7/2004, A/chicken/Miyazaki/K11/2007, A/chicken/Hong Kong/220/1997           | E   | 5.8 to 6.2/0.1 ml                                     | Not investigated  | Perkins                  |
|           | Ducks                                 | A/chicken/Yamaguchi/7/2004, A/chicken/Miyazaki/K11/2007   | E   | 8/0.1 ml  | Not investigated  | Yamamoto                 |
|           | Turkeys                               | H5N1 (strain not reported)  | N   | Not available   | Not investigated  | Slomka                   |
| Liver     | Ducks                                 | A/chicken/Vietnam/12/2005 (H5N1)  | E   | 7/0.1 ml  | Not reported  | Beato                    |
| Blood     | Chickens                              | A/tern/South Africa/61 (H5N3)   | E   | 7/0.1 ml  | 4   | Kishida, Swayne          |
|           |                                       | A/chicken/Pennsylvania/1370/1983 (H5N2)   |   | 6/0.1 ml  | Not reported  |                          |
|           | Pigeons, geese                        | A/turkey/Ontario/7732/66 (H5N9)   | E   | 8/0.5 ml  | Not recovered   | Narayan                  |
|           |                                       | Turkeys   | A/turkey/Italy/4580/99 (H7N1)             | E   | 6/0.1 ml  |                          |
|           | Ducks                                 | A/turkey/Ontario/7732/66 (H5N9)   |   | 8.7/0.5 ml  | 2.7 to 3.7  | Beato, Narayan           |
|           |                                       | A/chicken/Vietnam/12/2005 (H5N1)  | E   | 7/0.1 ml  | Not reported  |                          |
|           |                                       | A/turkey/Ontario/7732/66 (H5N9)   | E   | 8/0.5 ml  | Not recovered   |                          |
| Skin      | Ducks                                 | A/chicken/Yamaguchi/7/2004,   | E   | 8/0.1 ml  | 2.5 to 4.4  | Yamamoto                 |
|           |                                       | A/chicken/Yamaguchi/7/2004  | E   | 8/0.1 ml  | 3.5   |                          |
|           | Geese                                 | A/chicken/Miyazaki/K11/2007   | E   |   | 4.5   | Yamamoto                 |



# Vírus nos produtos de animais

**Table 2.** Summary of data available in literature on the presence of LPAI viruses in poultry commodities

| Commodity | Species         | Strain   | Natural (N) or experimental (E) infection | Infecting dose (EID <sub>50</sub> log <sub>10</sub> ) | Titres (log <sub>10</sub> EID <sub>50</sub> /g) | Reference (first author) |
|-----------|-----------------|--|---|---|---|--------------------------|
| Meat      | Chickens        | A/chicken/aq-Y-55/01 (H9N2);                                   | E   | 7/0.1 ml  | 1.6 to 2  | Kishida                  |
|           |                 | A/chicken/aq-Y-135/01(H9N2)                                    | E   | 7/0.1 ml  | 1.6 to 2  |                          |
|           | Turkeys         | A/turkey/Italy/3675/99 (H7N1)                                  | E   | 6/0.1 ml  | No infectious virus detected                    | Toffan                   |
|           |                 | A/turkey/Virginia/159512/2002 (H7N2)                           | E   | 6/0.1 ml  |   |                          |
|           |                 | A/chicken/New York/21586-8/99 (H7N2)                           | E   | 6/0.1 ml  |   |                          |
| Eggs      | Ducks           | No data available  |   |   |   |                          |
|           | Turkeys         | A/turkey/California/meleagrium/64; A/turkey/California/5142/66 | E   | 2.25/0.2 ml   | No infectious virus detected                    | Shalaby                  |
|           | Chickens        | A/chicken/Alabama/7395/75 (H4N8)                               | N   | Not reported  | No infectious virus detected                    | Shalaby                  |
| Feathers  | Ducks and geese | Not available  |   |   |   |                          |
| Liver     | Avian species   | Not available  |   |   |   |                          |
| Blood     | Chickens        | A/chicken/aq-Y-55/01 (H9N2)                                    | E (co infection with <i>S. aureus</i> )   | 6/0.1 ml  | Not reported                                    | Kishida, Swayne          |
|           |                 | A/chicken/Beijing/2/97 (H9N2)                                  |   | 6/0.1 ml  |   |                          |
|           | Turkeys         | A/turkey/Italy/3675/99 (H7N1)                                  | E   | 6/0.1 ml  | <1  | Toffan                   |
|           | Ducks           | No data available  |   |   |   |                          |



# Vírus nos produtos de animais

## Foodborne Germs and Illnesses

[Español \(Spanish\)](#) | [Kreyòl \(Creole\)](#) | [Print](#)



CDC estimates that each year 48 million people get sick from a foodborne illness, 128,000 are hospitalized, and 3,000 die.

## Causes of Food Poisoning

Top 5 Germs Causing Illness, Hospitalizations, and Deaths From Food Eaten in the United States

| Illnesses                            | Hospitalizations                     | Deaths                               |
|--------------------------------------|--------------------------------------|--------------------------------------|
| 1. <b>Norovirus</b>                  | 1. <i>Salmonella</i> (non-typhoidal) | 1. <i>Salmonella</i> (non-typhoidal) |
| 2. <i>Salmonella</i> (non-typhoidal) | 2. <b>Norovirus</b>                  | 2. <i>Toxoplasma gondii</i>          |
| 3. <i>Clostridium perfringens</i>    | 3. <i>Campylobacter</i>              | 3. <i>Listeria monocytogenes</i>     |
| 4. <i>Campylobacter</i>              | 4. <i>Toxoplasma gondii</i>          | 4. <b>Norovirus</b>                  |
| 5. <i>Staphylococcus aureus</i>      | 5. <i>E. coli</i> O157               | 5. <i>Campylobacter</i>              |

- *Salmonella* can cause [salmonellosis](#) and [typhoid fever and paratyphoid fever](#).
- [Botulism](#) is most often caused by [Clostridium botulinum](#).
- Some other germs that cause foodborne illness include [Cryptosporidium](#), [Cyclospora](#), [hepatitis A virus](#), [Shigella](#), and [Yersinia](#).
- See a complete [A-Z index of foodborne illnesses](#).





# Norovírus no Brasil

## Vírus apontado como causador de surto de diarreia é encontrado em rio de Florianópolis

Agente foi detectado no Rio do Brás, que chega até areia da praia de Canasvieiras, totalmente imprópria para banho. Especialista aponta que falta de saneamento básico pode ajudar na circulação do vírus.

Por Joana Caldas e Juan Todescatt, g1 SC e NSC TV

23/01/2023 06h57 · Atualizado há 2 meses



SEGUNDO JORNAL

Prova do Masterchef Espanha deixa 40 pessoas com intoxicação alimentar



ALEMÃHA

Brasileiras que tiveram mal

## Norovírus: como prevenir o microrganismo por trás do surto de diarreia em Florianópolis



Medidas de prevenção incluem práticas de higiene e de consumo adequado de alimentos



Medidas de prevenção incluem práticas de higiene e de consumo adequado de alimentos  
Foto: Shutterstock/Unsplash



# Rotavírus no Brasil

≡ MENU | g1 PERNAMBUCO



São Lourenço da Mata registrou mais de 6 mil casos de rotavírus.

Mais de 6,2 mil casos de diversas viroses, incluindo o rotavírus humano, foram registrados entre novembro de 2022 e janeiro deste ano, em **São Lourenço da Mata**, no Grande **Recife** (veja vídeo acima).

**Errata:** O **g1** errou ao publicar na reportagem que os casos registrados eram só de rotavírus humano. Esse dado foi repassado pela assessoria de comunicação da Unidade de Pronto Atendimento (UPA) do município. Além disso, os casos foram registrados entre novembro de 2022 e janeiro de 2023. As informações foram corrigidas às 7h52 desta quinta (9).

Segundo a UPA da cidade, essas doenças atingem majoritariamente crianças e causam diarreia, vômito e desidratação, podendo levar à morte (veja aqui os sintomas).

- [Compartilhar no WhatsApp](#)



# Muito obrigada

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