

BMM 0160 – Microbiologia Básica para Farmácia

Classificação dos vírus

Cultivo de vírus animais

Profa. Patricia C. B. Beltrão Braga

Depto de Microbiologia- ICB/USP



CLASSIFICAÇÃO DOS VÍRUS

Existem dois sistemas:

- Sistema hierárquico
- Sistema de Baltimore

CLASSIFICAÇÃO DOS VÍRUS

Sistema hierárquico de classificação de vírus

- Criado em 1962 por Lwoff, Horne e Tournier
- Considera a estrutura phylum - classe - ordem - família - subfamília - género - espécie - cepa/tipo
- Princípio: os vírus devem ser agrupados em função de suas características em comum e não em função das características das células que infectam.



André Lwoff

CLASSIFICAÇÃO DOS VÍRUS

São utilizadas 4 características fundamentais:

- Tipo de ácido nucléico (DNA ou RNA)
- Simetria do capsídeo
- Presença de envelope
- Dimensões do vírion ou capsídeo

Taxonomia

- Parte crítica da virologia!
 - ***The International Committee on Taxonomy of Viruses Universal System of Virus Taxonomy = ICTV***
(Committee of the Virology Division of the International Union of Microbiological Societies)

Taxonomia

6 orders, 87 families, 19 subfamilies, 348 genera, and 2,290 species.

Order: -virales
Families: -viridae
Subfamilies: -virinae
Genera: -virus

TABLE 2.2 Taxonomy of the Order <i>Mononegavirales</i>						
Order	Family	Subfamily	Genus	Type species	Host	
<i>Mononegavirales</i>	<i>Bornaviridae</i>		<i>Bornavirus</i>	<i>Borna disease virus</i>	V	
			<i>Vesiculovirus</i>	<i>Vesicular stomatitis Indiana virus</i>	V, I	
	<i>Rhabdoviridae</i>			<i>Lyssavirus</i>	<i>Rabies virus</i>	V
				<i>Ephemerovirus</i>	<i>Bovine ephemeral fever virus</i>	V, I
				<i>Novirhabdovirus</i>	<i>Infectious hematopoietic necrosis virus</i>	V
				<i>Cytorhabdovirus</i>	<i>Lettuce necrotic yellows virus</i>	P, I
				<i>Nucleorhabdovirus</i>	<i>Potato yellow dwarf virus</i>	P, I
				<i>Marburgvirus</i>	<i>Lake Victoria marburgvirus</i>	V
				<i>Ebolavirus</i>	<i>Zaire ebolavirus</i>	V
	<i>Paramyxoviridae</i>	<i>Paramyxovirinae</i>		<i>Rubulavirus</i>	<i>Mumps virus</i>	V
				<i>Avulavirus</i>	<i>Newcastle disease virus</i>	V
				<i>Respirovirus</i>	<i>Sendai virus</i>	V
				<i>Henipavirus</i>	<i>Hendra virus</i>	V
				<i>Morbillivirus</i>	<i>Measles virus</i>	V
		<i>Pneumovirinae</i>		<i>Pneumovirus</i>	<i>Human respiratory syncytial virus</i>	V
			<i>Metapneumovirus</i>	<i>Avian metapneumovirus</i>	V	

CLASSIFICAÇÃO/Taxonomia

- Segundo o hospedeiro
 - Vírus de vertebrados
 - Vírus de invertebrados
 - Vírus de plantas
 - Vírus de bactérias (Bacteriófagos)
 - Vírus de fungos (Micovírus)
- Segundo o tipo de ácido nucleico
 - vírus de DNA
 - Virus de RNA

CLASSIFICAÇÃO/Taxonomia

- Presença ou ausência de envoltório:
 - envelopados
 - não envelopados
- Estrutura e simetria do capsídeo
- Composição química
- Homologia nucleotídica

Vírus de vertebrados

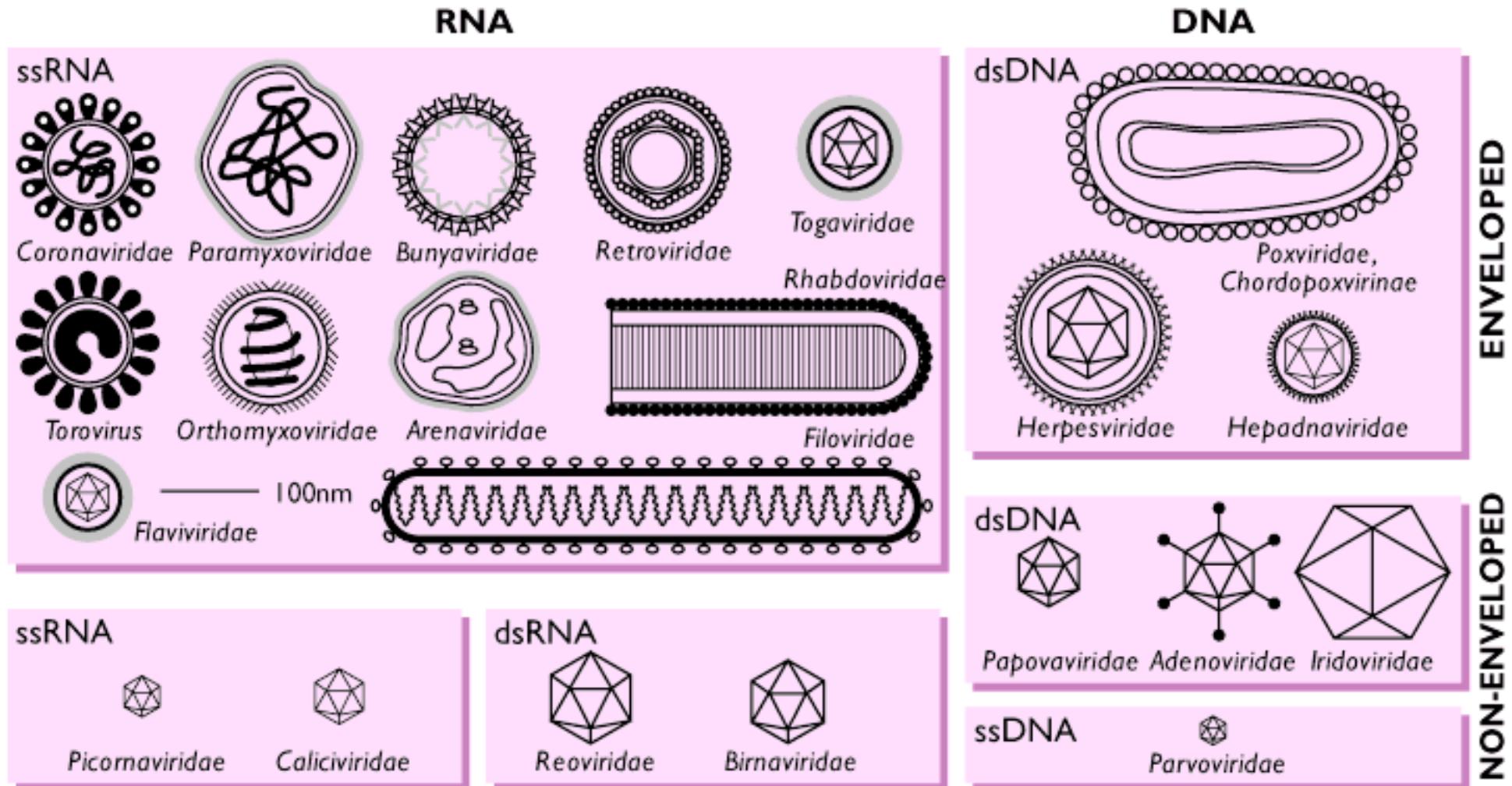


Fig. 5 Diagrammatic representation of the families of viruses infecting vertebrates, grouped according to the nature and strandedness of their genome and the presence or absence of an envelope. Reproduced with permission from Springer-Verlag.

Vírus de bactérias

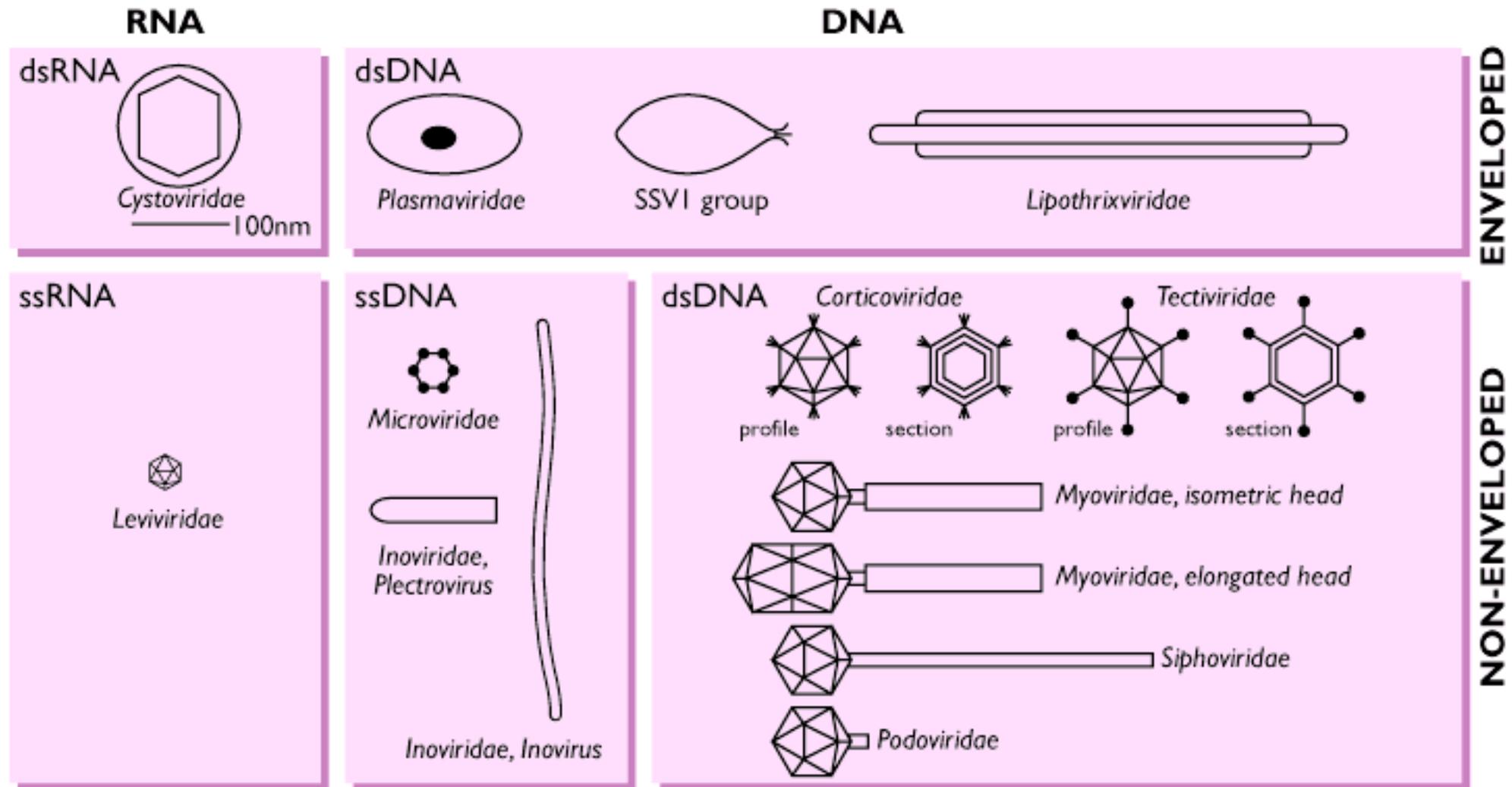


Fig. 1 Diagrammatic representation of the families of viruses infecting bacteria, group according to the nature and strandedness of their genome and the presence or absence of an envelope. Reproduced with permission from Springer-Verlag.

TABLE 2.1 Summary Characteristics of Vertebrate Virus Families

Family	Nucleocapsid morphology	Envelope	Virion morphology	Genome*	Host†
dsDNA viruses					
<i>Adenoviridae</i>	Icosahedral	No	Icosahedral	1 ds linear, 26–48 kb	V
<i>Alloherpesviridae</i>	Icosahedral	Yes	Spherical, tegument	2 ds linear, 135–294 kb	V
<i>Asfviridae</i>	Icosahedral	Yes ^c	Icosahedral	1 ds linear, 165–190 kb	V, I
<i>Herpesviridae</i>	Icosahedral	Yes	Spherical, tegument	1 ds linear, 125–240 kb	V
<i>Iridoviridae</i>	Icosahedral	No ^d	Icosahedral	1 ds linear, 140–303	V, I
<i>Papillomaviridae</i>	Icosahedral	No	Icosahedral	1 ds circular, 7–8 kb	V
<i>Polyomaviridae</i>	Icosahedral	No	Icosahedral	1 ds circular, 5 kb	V
<i>Poxviridae</i>	Ovoid	Yes	Ovoid	1 ds linear, 130–375 kb	V, I
ssDNA viruses					
<i>Anellovirus</i>	Icosahedral	No	Icosahedral	1 – circular, 2–4 kb	V
<i>Circoviridae</i>	Icosahedral	No	Icosahedral	1 – or ± circular, 2 kb	V
<i>Parvoviridae</i>	Icosahedral	No	Icosahedral	1 +, – or ± linear, 4–6 kb	V, I
dsDNA reverse transcribing viruses					
<i>Hepadnaviridae</i>	Icosahedral	Yes	Spherical	1 ds circular, 3–4 kb	V
ssRNA reverse transcribing viruses					
<i>Metaviridae</i>	Spherical	Yes	Spherical	1 + linear, 4–10 kb	F, I, P, V
<i>Retroviridae</i>	Spherical, rod or cone shaped	Yes	Spherical	1 + linear dimer, 7–13 kb	V
dsRNA viruses					
<i>Birnaviridae</i>	Icosahedral	No	Icosahedral	2 ds linear, 5–6 kb	V, I
<i>Picobirnaviridae</i>	Icosahedral	No	Icosahedral	3 ds linear, 4 kb	V
<i>Reoviridae</i>	Icosahedral	No	Icosahedral, layered	10–12 ds linear, 19–32 kb	V, I, P, F
Negative sense ssRNA viruses					
<i>Bornaviridae</i>	ND*	Yes	Spherical	1 – linear, 9 kb	V
<i>Deltavirus</i>	Isometric	Yes	Spherical	1 – circular, 2 kb	V
<i>Filoviridae</i>	Helical filaments	Yes	Bacilliform, filamentous	1 – linear, 19 kb	V
<i>Orthomyxoviridae</i>	Helical filaments	Yes	Pleomorphic, spherical	6–8 – linear, 10–15 kb	V
<i>Paramyxoviridae</i>	Helical filaments	Yes	Pleomorphic, spherical, filamentous	1 – linear, 13–18 kb	V
<i>Rhabdoviridae</i>	Coiled helical filaments	Yes	Bullet shaped	1 – linear, 11–15 kb	V, I, P
Positive sense ssRNA viruses					
<i>Arteriviridae</i>	Linear, asymmetric	Yes	Spherical	1 + linear, 13–16 kb	V
<i>Astroviridae</i>	Icosahedral	No	Icosahedral	1 + linear, 6–8 kb	V
<i>Caliciviridae</i>	Icosahedral	No	Icosahedral	1 + linear, 7–8 kb	V
<i>Coronaviridae</i>	Helical	Yes	Spherical	1 + linear, 26–32 kb	V
<i>Flaviviridae</i>	Spherical	Yes	Spherical	1 + linear, 9–13 kb	V, I
<i>Hepevirus^e</i>	Icosahedral	No	Icosahedral	1 + linear, 7 kb	V
<i>Nodaviridae</i>	Icosahedral	No	Icosahedral	2 + linear, 4–5 kb	V, I
<i>Picomaviridae</i>	Icosahedral	No	Icosahedral	1 + linear, 7–9 kb	V
<i>Togaviridae</i>	Icosahedral	Yes	Spherical	1 + linear, 10–12 kb	V, I
Ambisense ssRNA viruses					
<i>Arenaviridae</i>	Filamentous	Yes	Spherical	2 ± linear, 11 kb	V
<i>Bunyaviridae</i>	Filamentous	Yes	Spherical	3 – or ± linear, 11–19 kb	V, I, P
Subviral agents: prions					
Prions	—	—	—	—	V, F

CLASSIFICAÇÃO DOS VÍRUS

Sistema de Baltimore

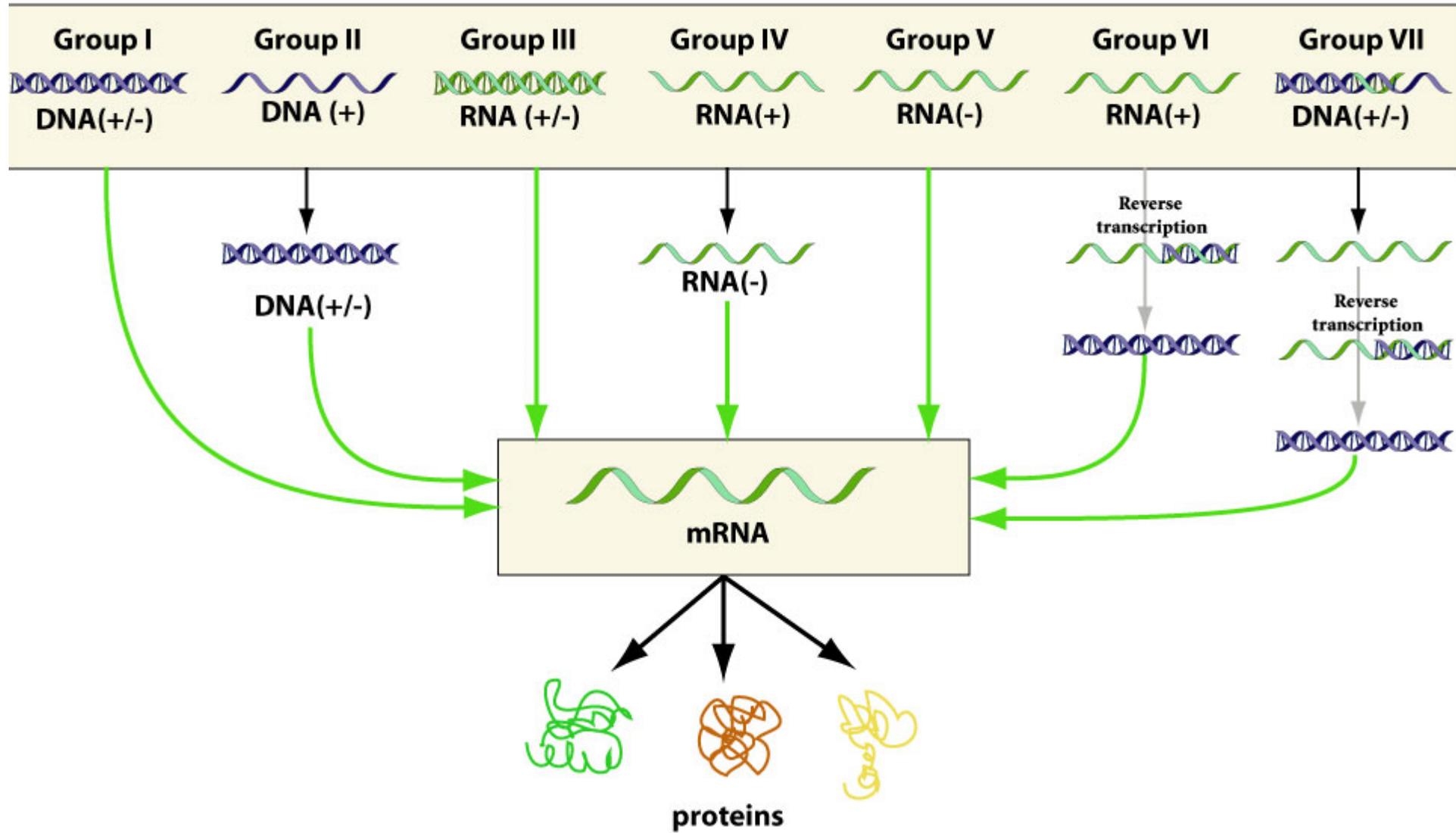


David Baltimore

- Baseado nas diferentes estratégias de replicação viral e na dependência absoluta dos vírus do sistema de tradução protéica da célula.
- Princípio: todos vírus devem produzir mRNA a partir do genoma para sintetizar proteínas e replicar seu material genético. O mecanismo varia entre os diferentes tipos de vírus.

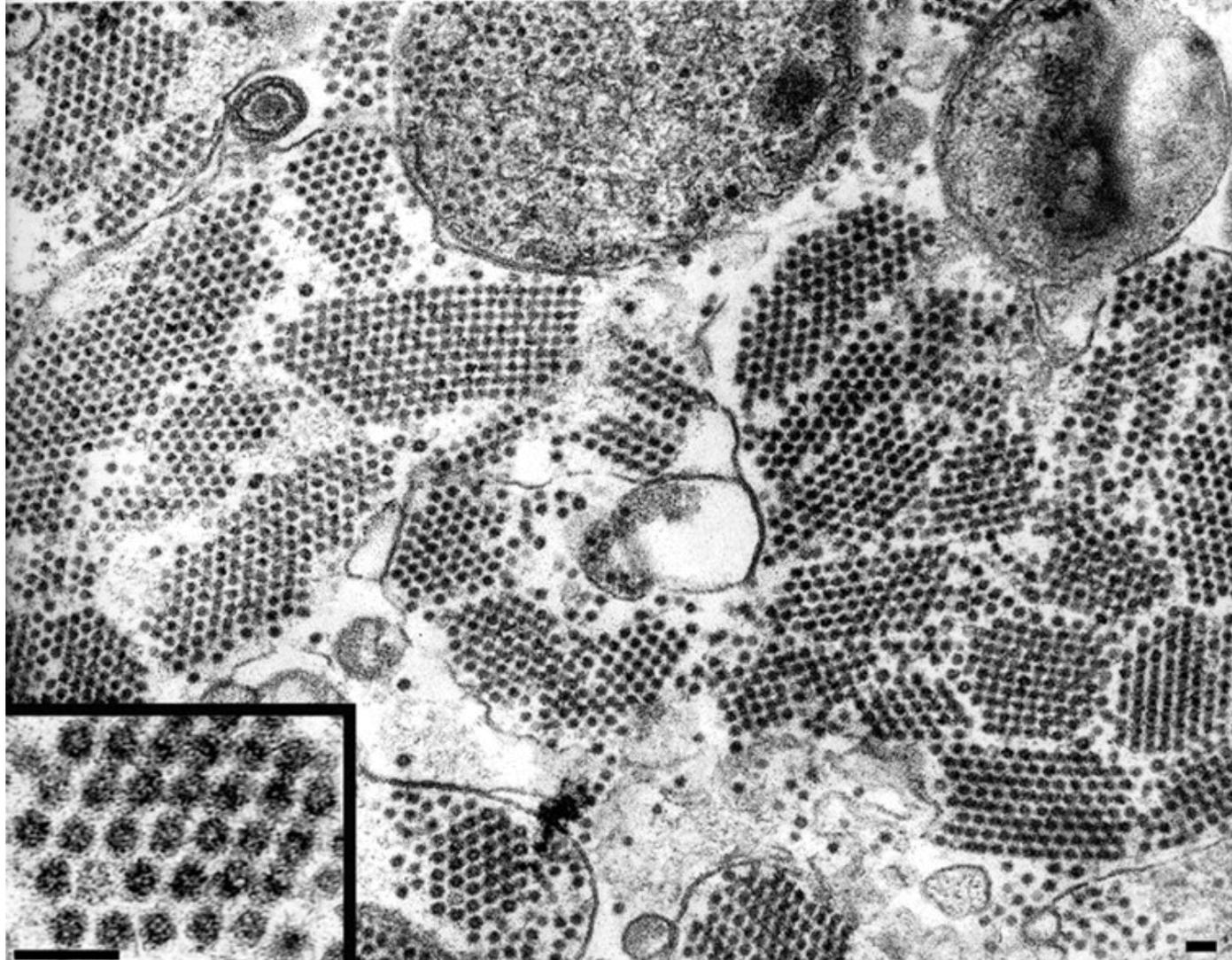
CLASSIFICAÇÃO DOS VÍRUS

Sistema de Baltimore



Cultivo de vírus animais

Vírus são parasitas intracelulares obrigatórios



Membrana corioalantóide de embrião de galinha infectado com vírus da varíola de vaca (cowpox), A e B.



Cultura de células

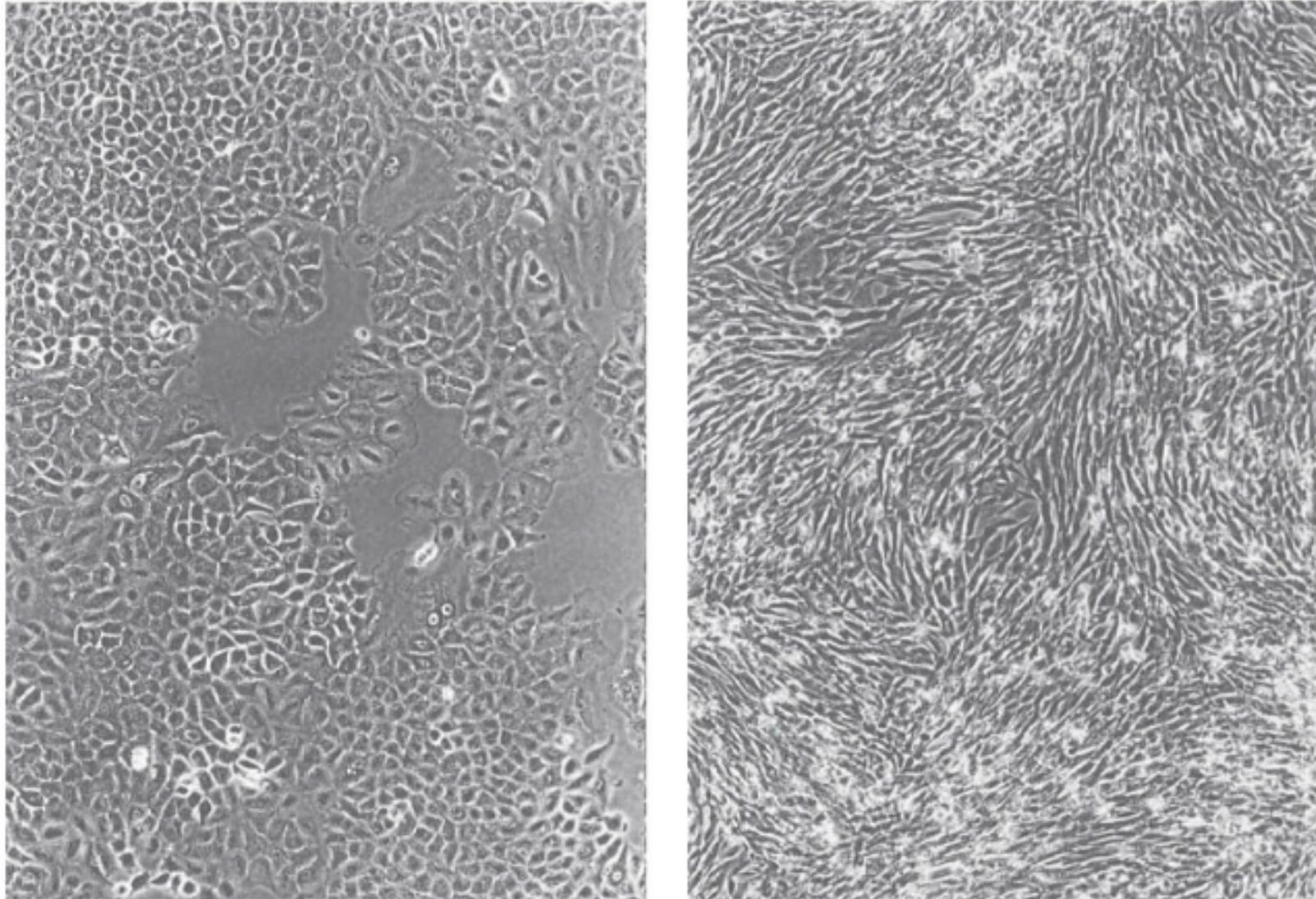
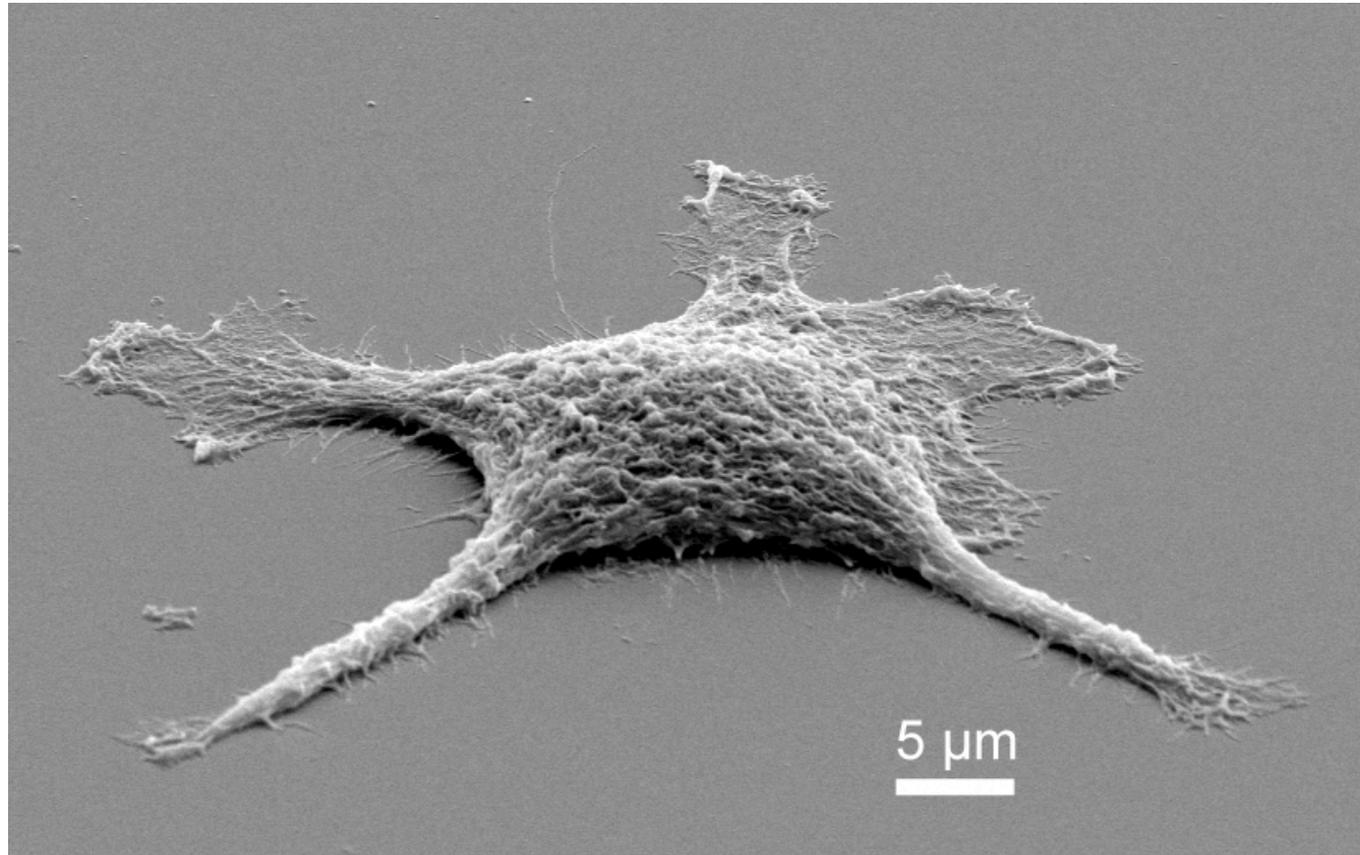


FIGURE 24. Cultured cell types. Phase contrast photomicrographs are shown. **A:** Epithelial-like cells, A549, a human lung carcinoma cell line, a slightly subconfluent monolayer. **B:** Fibroblast-like cells, BHK, a baby hamster kidney cell line. (A549 cell culture courtesy of J. I. Lewis. BHK cell culture courtesy of D. Holmes and Dr. S. Moyer.)

Tipos de culturas celulares

1. Primárias
2. Secundárias ou permanentes (“linhagens celulares”)

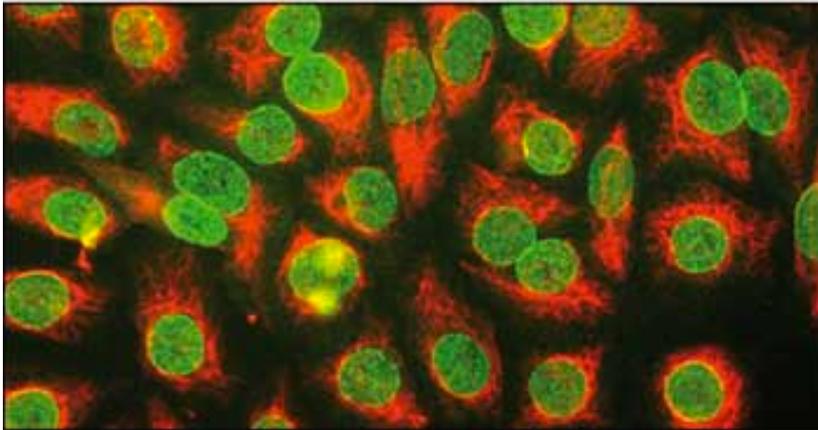


Henrietta Lacks

**1 agosto 1920
4 outubro 1951
(31 anos)**



Henrietta Lacks e David Lacks

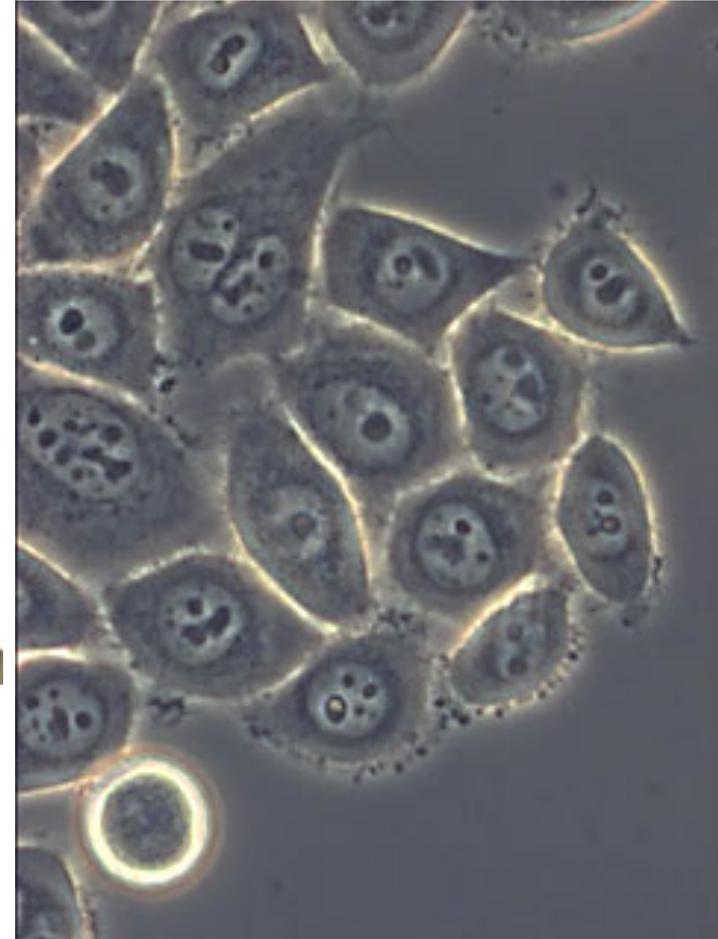
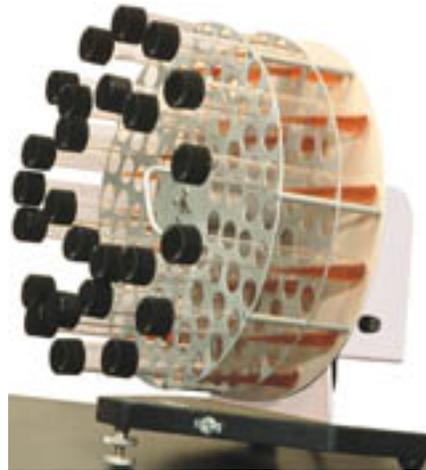


Células HeLa

George Otto Gey (1899-1970)
Professor, Johns Hopkins



Mary Kubicek, assistente do Dr George Gey
21 anos, fisiologia



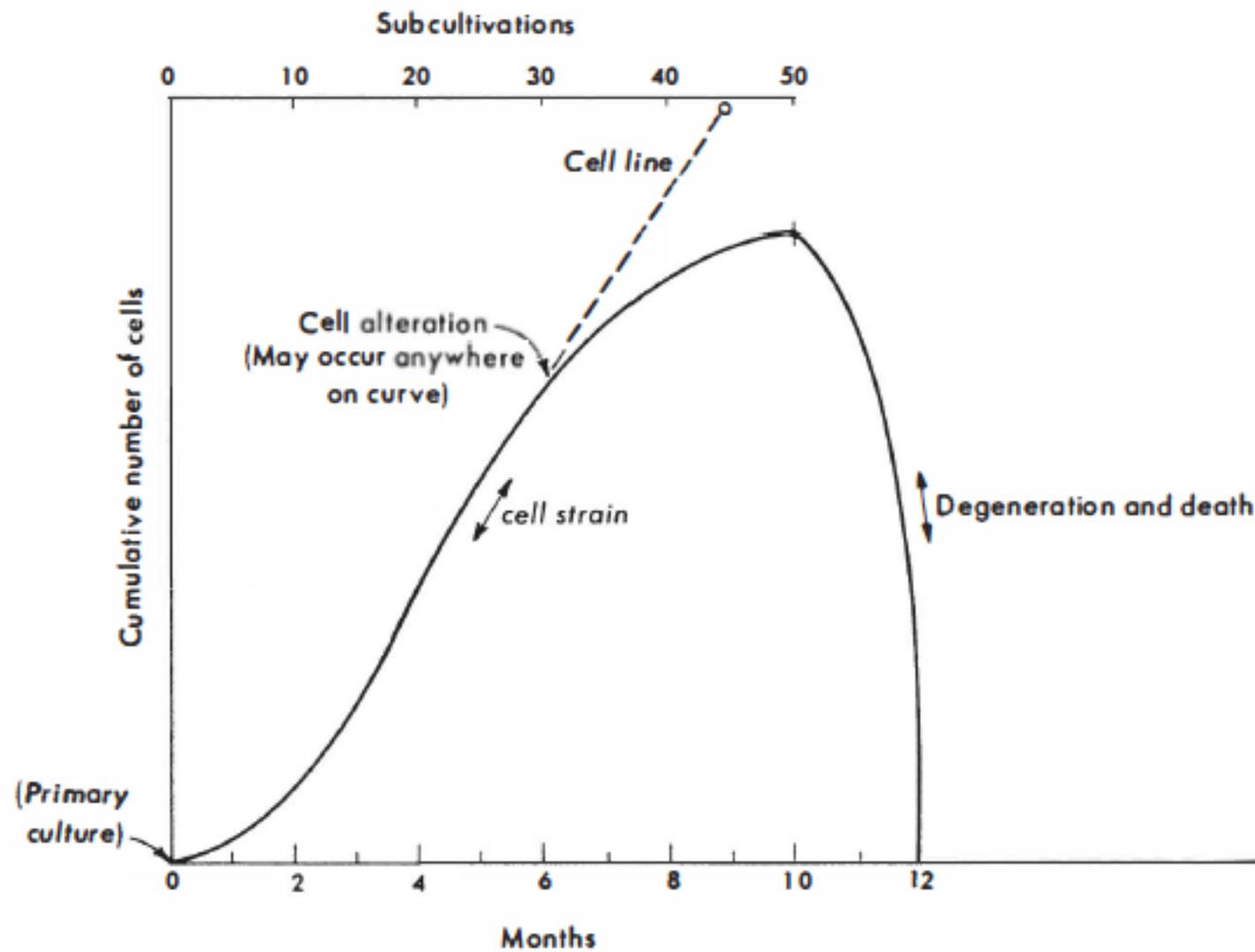
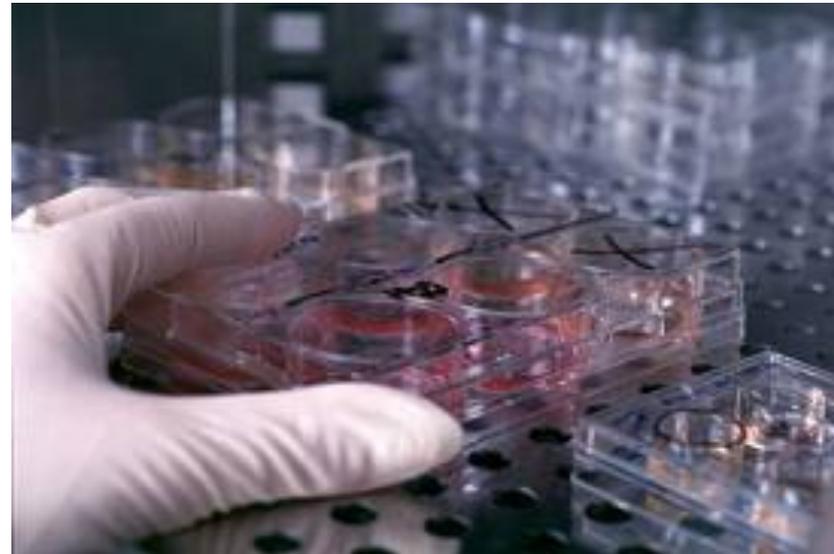


FIGURE 2.3. Growth of cells in culture.

Manutenção

- Células de mamíferos são mantidas em placas ou frascos plásticos apropriados



INCUBADORA⁷



CO₂

Meio de cultivo

- aminoácidos, vitaminas, sais minerais, proteínas e outros componentes como glicose, antibióticos, fungicidas



Armazenamento

- Nitrogênio líquido a -196°C , período muito longo
- Freezer -80°C , período curto
- Devem ser acrescentadas de Soro Fetal e DMSO!

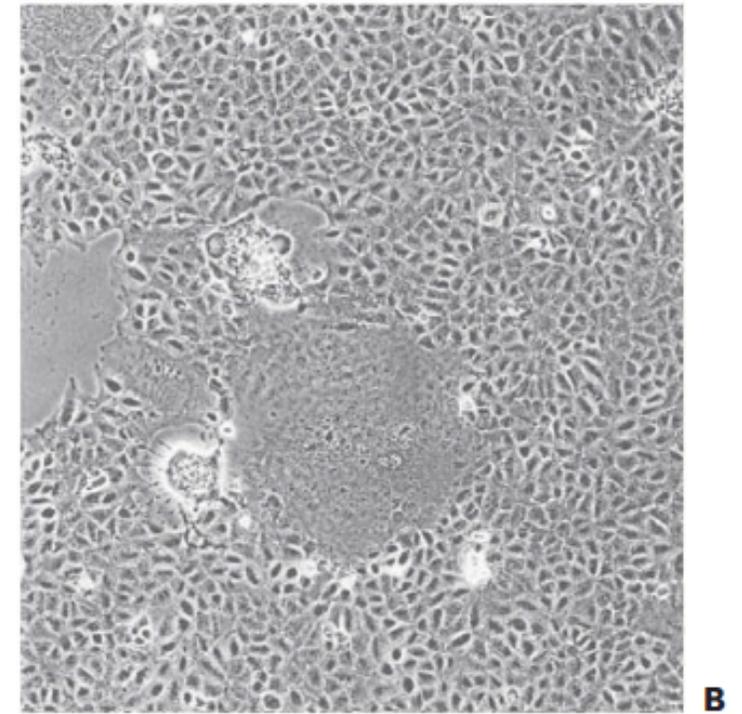
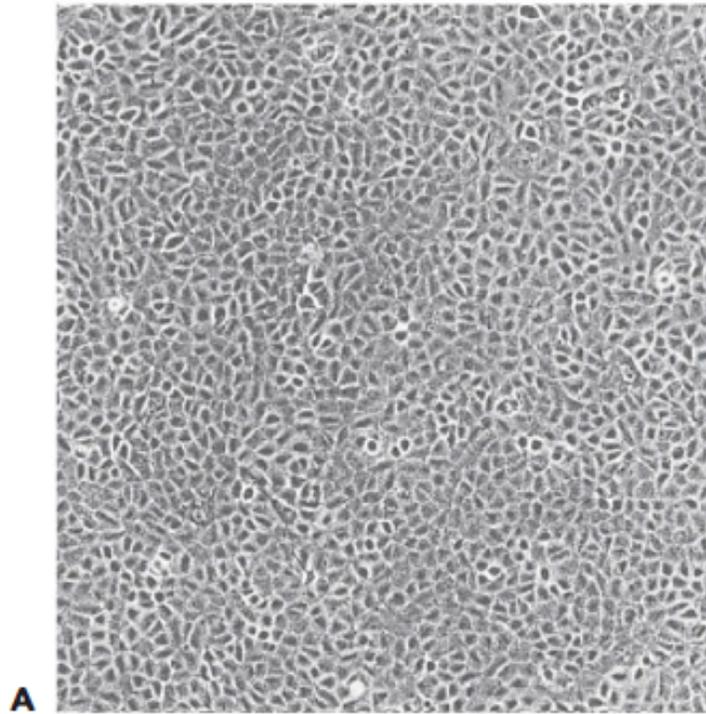


Efeito Citopático Viral

A: Células A549 não infectadas

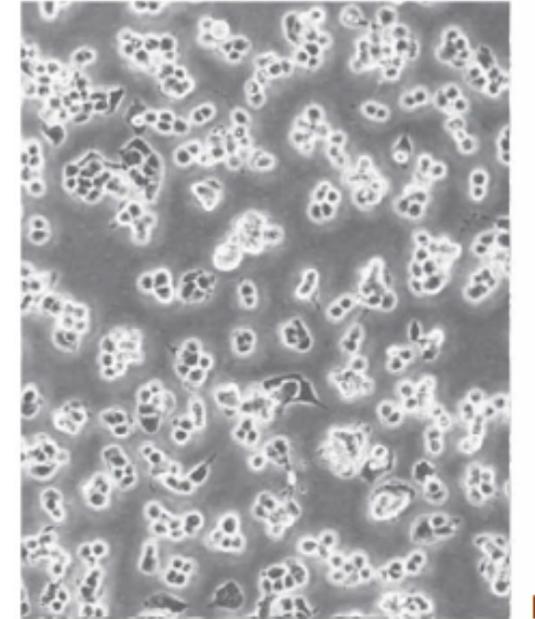
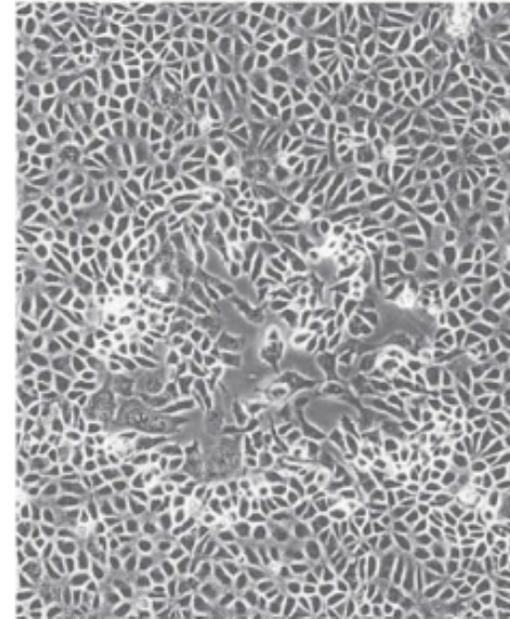
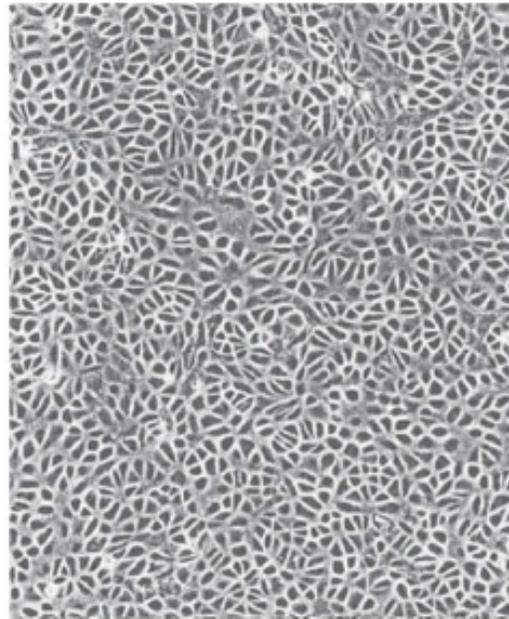
B: Sarampo

C: Células BSC40 não infectadas



D: Vaccinia vírus (gênero Orthopoxvirus da família Poxviridae), usado como vacina contra varíola humana (0,01 pfu/cell)

E: com 1000x mais vírus (10 pfu/cell)



C,D

E

