



DEPARTAMENTO DE
MICroBiologia
UNIVERSIDADE DE SÃO PAULO



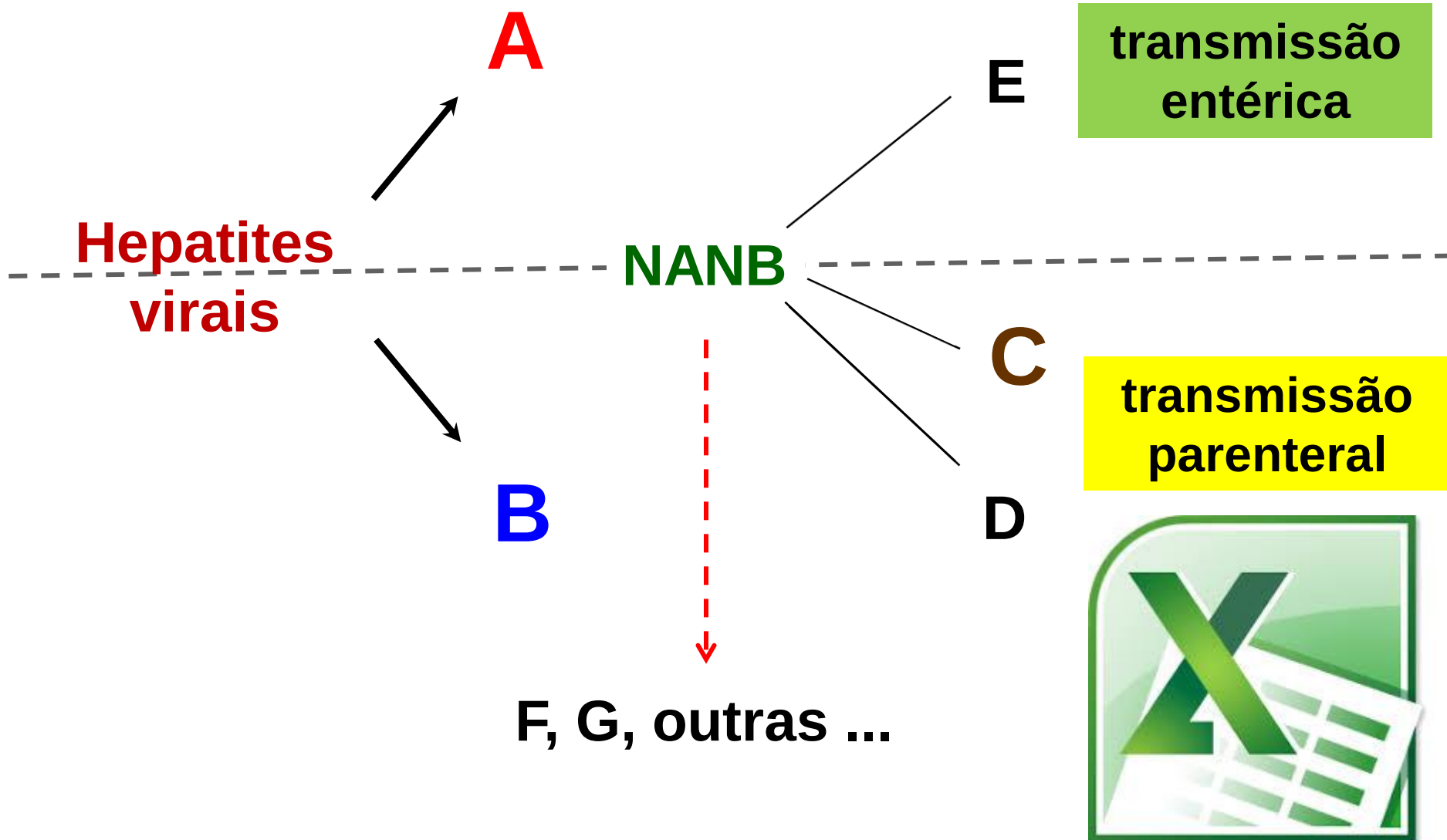
Hepatites Virais

Enrique Boccardo

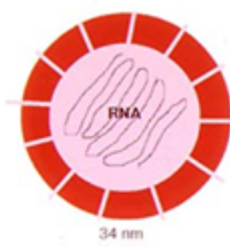
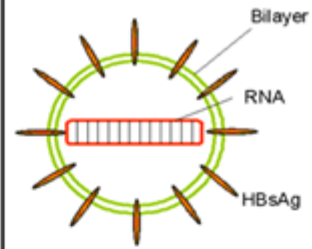
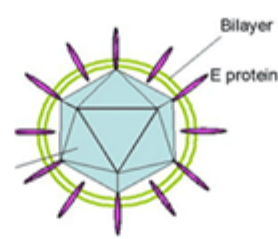
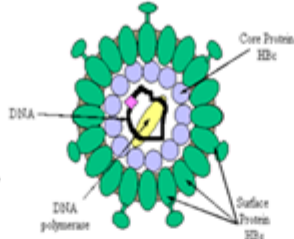
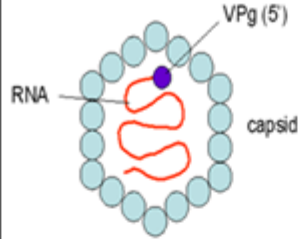
Depto. Microbiologia – ICB/USP

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É uma virose...



Hepatitis Virais



Name of Virus

Hepatitis A Virus (HAV)

Hepatitis B Virus (HBV)

Hepatitis C Virus (HCV)

Hepatitis D Virus (HDV)

Hepatitis E Virus (HEV)

Classification

Picornavirus

Hepadnavirus

Flavivirus

Deltavirus

Hepevirus

Viral genome

ssRNA

dsDNA

ssRNA

-ssRNA (-ve)

ssRNA

Transmission

Enteric

Parental

Parental

Parental

Enteric

Incubation period

15-45 days

45-160 days

15-150 days

30-60 days

15-60 days

Chronic Hepatitis

No.

Yes.
10% chance

Yes.
>50% chance

Yes.
<5% of
coinfectious
>80% of
superinfectious

No.

Cure?

No cure.
Treatments
usually tackle
the symptoms.

No cure.
Treatments
usually tackle
the symptoms.

No cure.
Treatments
usually tackle
the symptoms.

No cure.
Treatment: Alpha
interferon for 12
months.

No cure.
Treatments
usually tackle
the symptoms

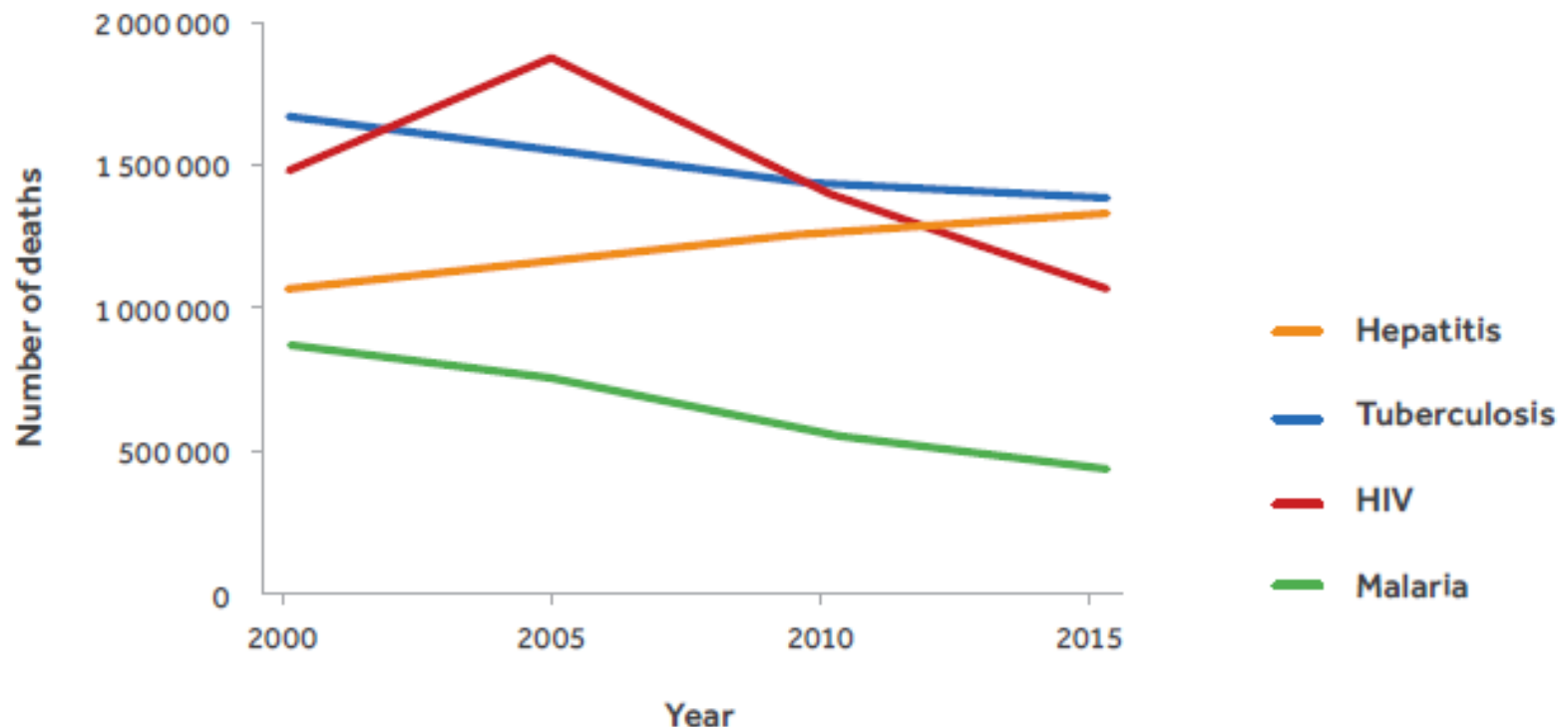
Hepatites Virais: impacto nas populações humanas

Estimativas da WHO (2015)

- 257 milhões de pessoas (3.5% da população mundial) estão infectados de maneira crônica pelo HBV.
- 5% das pessoas infectadas por HBV estão infectadas por HDV.
- 71 milhões de pessoas (1.0% da população mundial) estão infectados de maneira crônica pelo HCV.
- 20 milhões de pessoas/ano se infectam com o HEV (44.000 mortes em 2015 [3.3% das mortes por Hepatites virais]).
- Mais de 7.000 mortes por HAV em 2016 (taxa de hepatite fulminante varia de 0.14 – 0.35, portanto, 2 – 5 milhões de casos por ano).

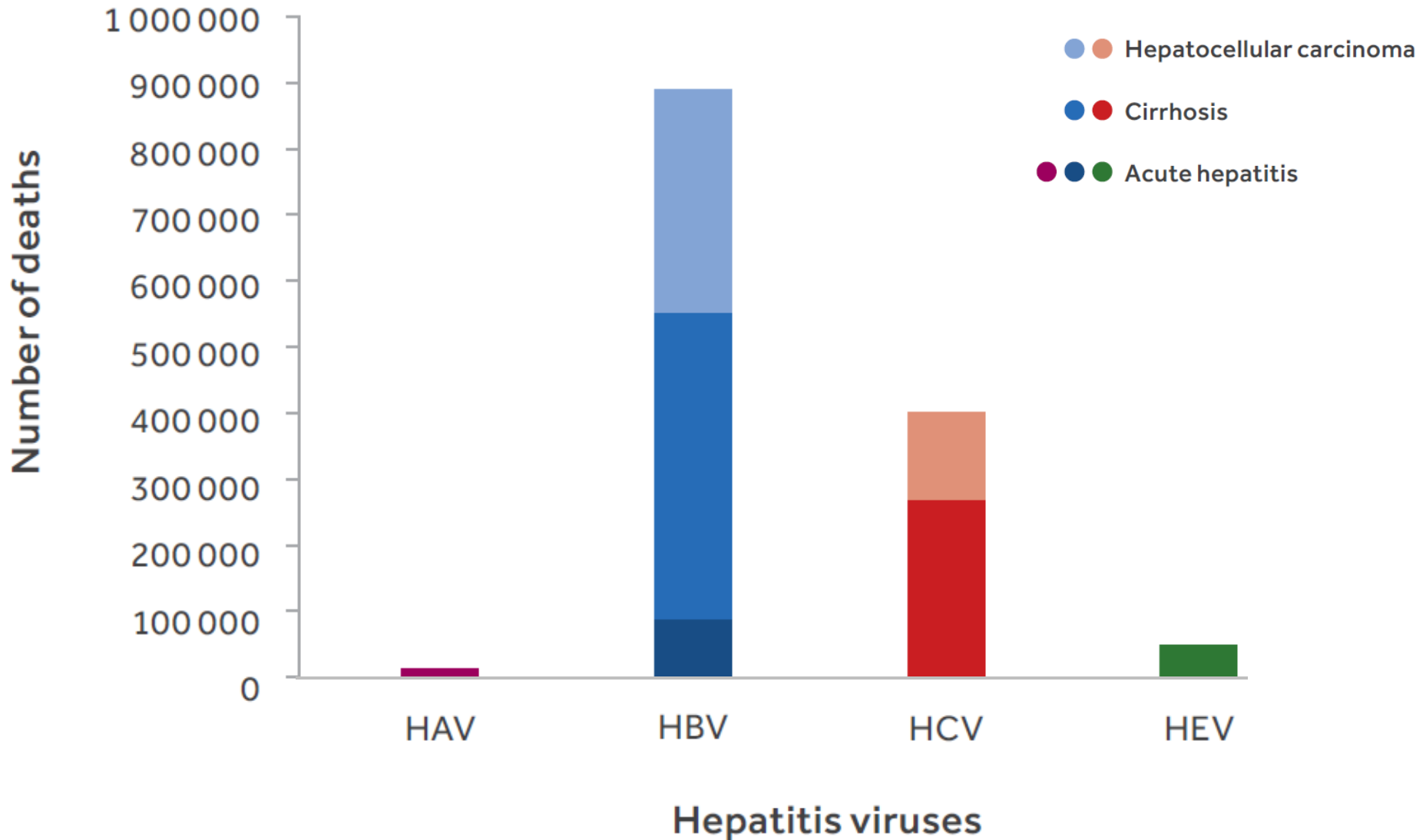
Hepatites Virais: impacto nas populações humanas

Fig. 2. Global annual mortality from hepatitis, HIV, tuberculosis and malaria, 2000–2015: unlike HIV, tuberculosis and malaria, the trend in mortality from viral hepatitis is increasing



Source: WHO global health estimates (Global Health Estimates 2015: deaths by cause, age, sex, by country and by region, 2000–2015. Geneva: World Health Organization; 2016.)

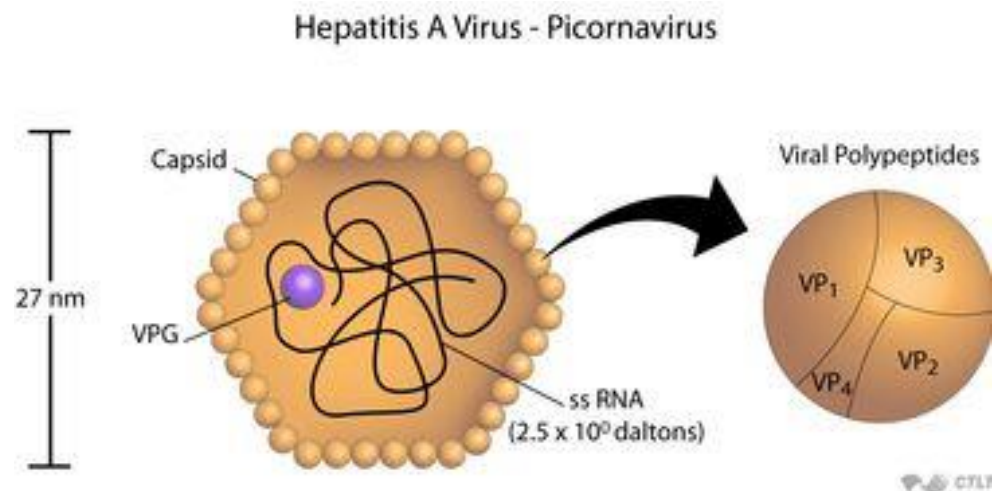
Hepatites Virais: impacto nas populações humanas



HAV: hepatitis A virus; HBV: hepatitis B virus; HCV: hepatitis C virus; HEV: hepatitis E virus Source: WHO global health estimates for 2015 published in 2016 (Global Health Estimates 2015: deaths by cause, age, sex, by country and by region, 2000–2015. Geneva: World Health Organization; 2016.)

Vírus de hepatite de transmissão entérica

Hepatitis A (HAV)



Hepatite A

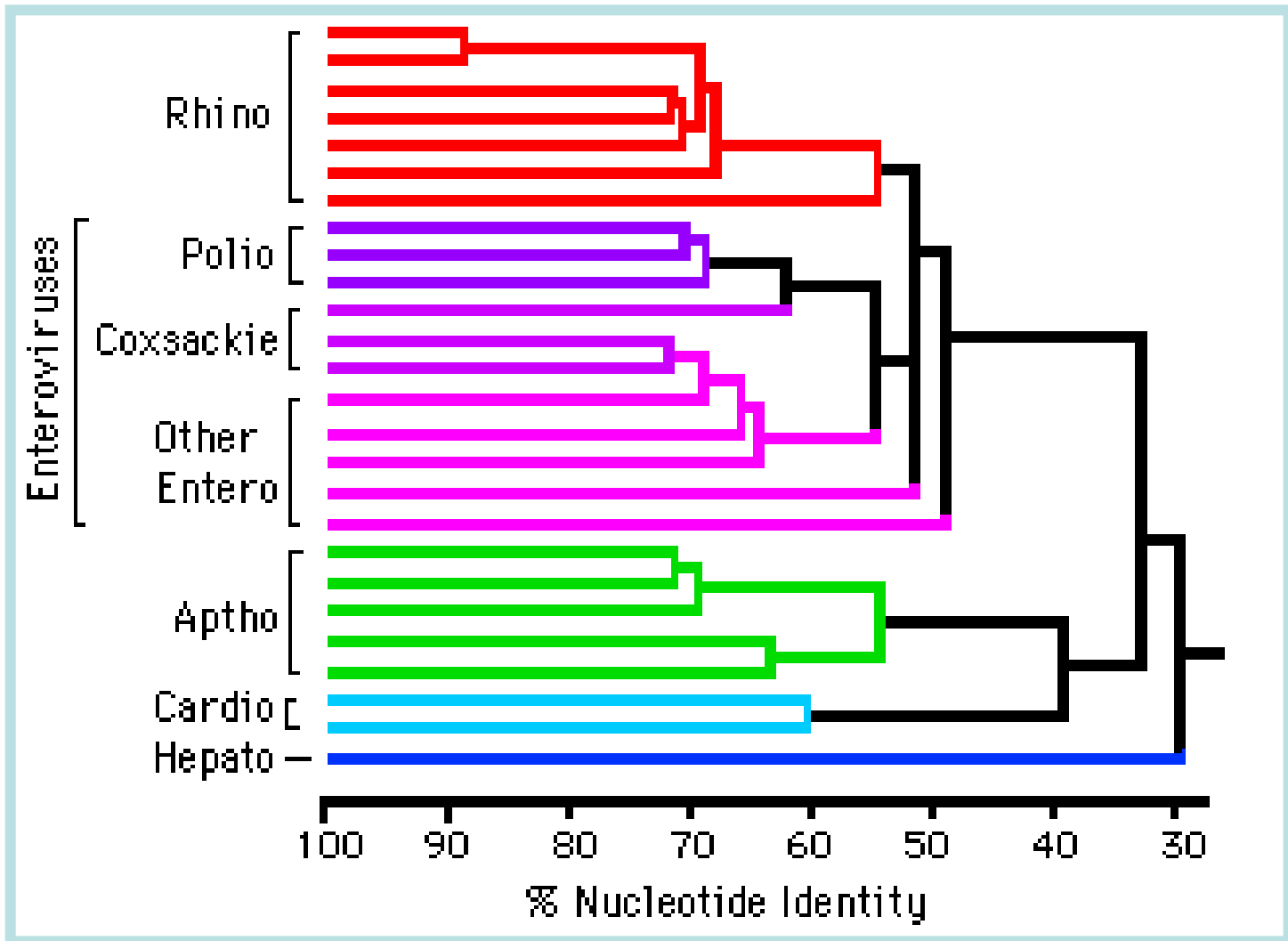
Família: *Picornavidae*

Gênero: Hepatovírus

- Capsídeo icosaédrico, não envelopado, de 27-32nm.
- Resistente ao calor, frio, condições ácidas e relativa resistência à desinfecção.
- Genoma de RNA linear, de fita simples, (+).
- Genótipos I,II e III infectam humanos
- Transmissão via oral-fecal e parentérica (Crianças = fonte de infecção)

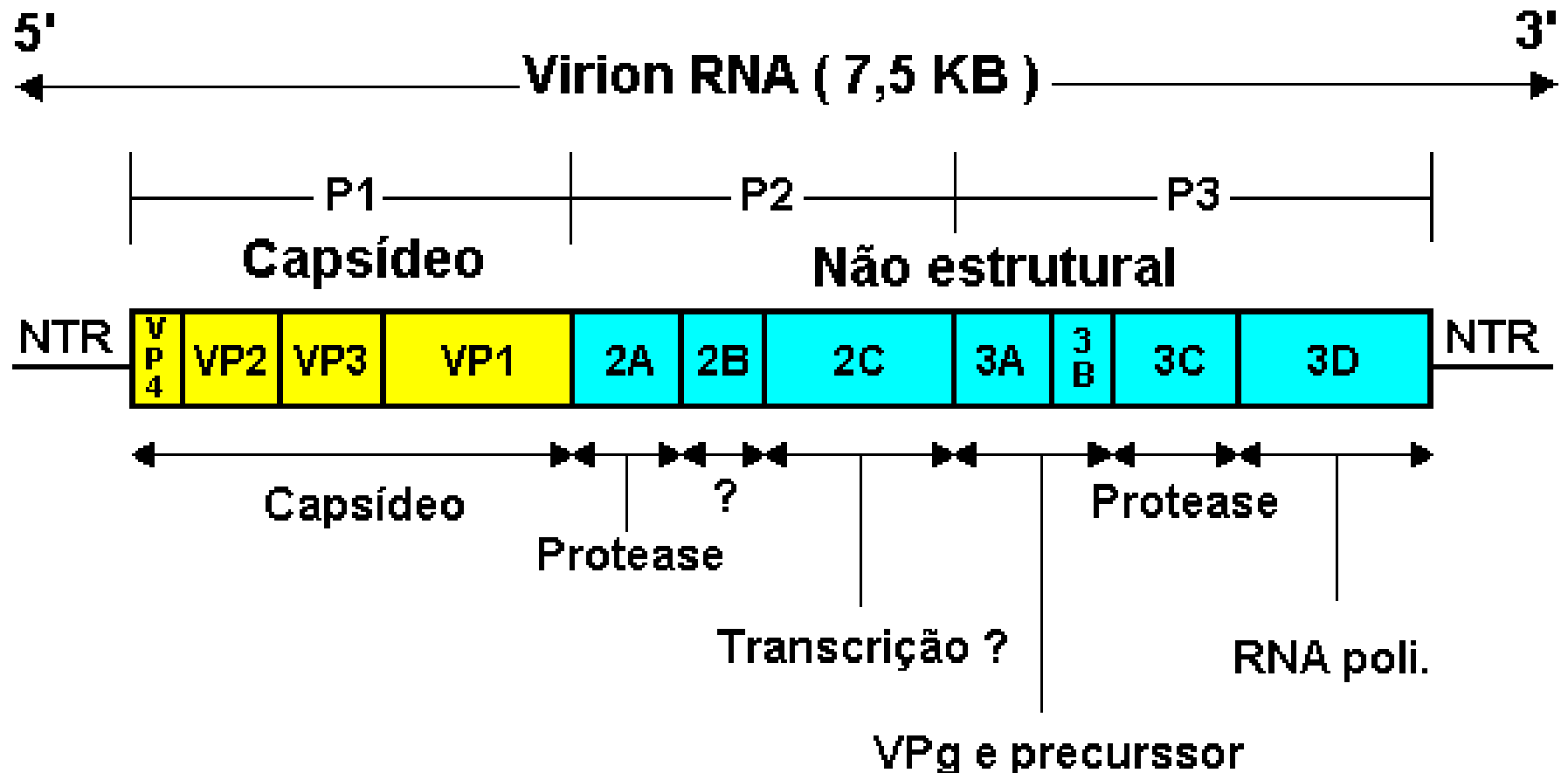
Hepatitis A

PICORNAVÍRUS



Hepatite A

Organização genômica



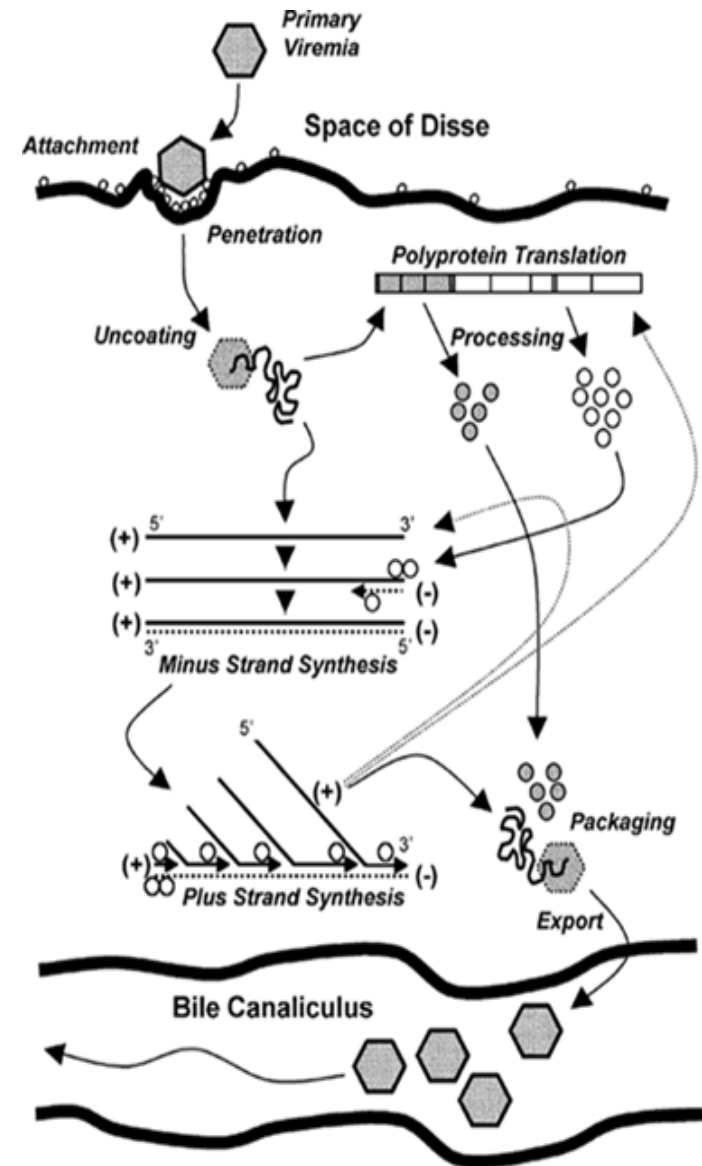
Hepatite A

Multiplicação

- Transmissão por água e alimentos.
- Porta de entrada pela via oral.
- Replicação viral primária no intestino delgado e migração por veia porta até o fígado.
- Excretado por bile e pelas fezes.

Na célula:

- entrada por endocitose
- genoma RNA+ vai para os ribossomos
- formação de poliproteína
- clivagem por protease viral
- replicação do genoma viral
- montagem no citoplasma
- liberação por lise



Hepatite A

Diagnóstico Laboratorial

- Ensaaios imunoenzimáticos
 - pesquisa de anticorpos
- Imunomicroscopia eletrônica
 - antígeno nas fezes
- Técnicas de biologia molecular
 - PCR, sequenciamento (tipagem dos vírus circulantes)

Hepatite A

Pesquisa de anticorpos

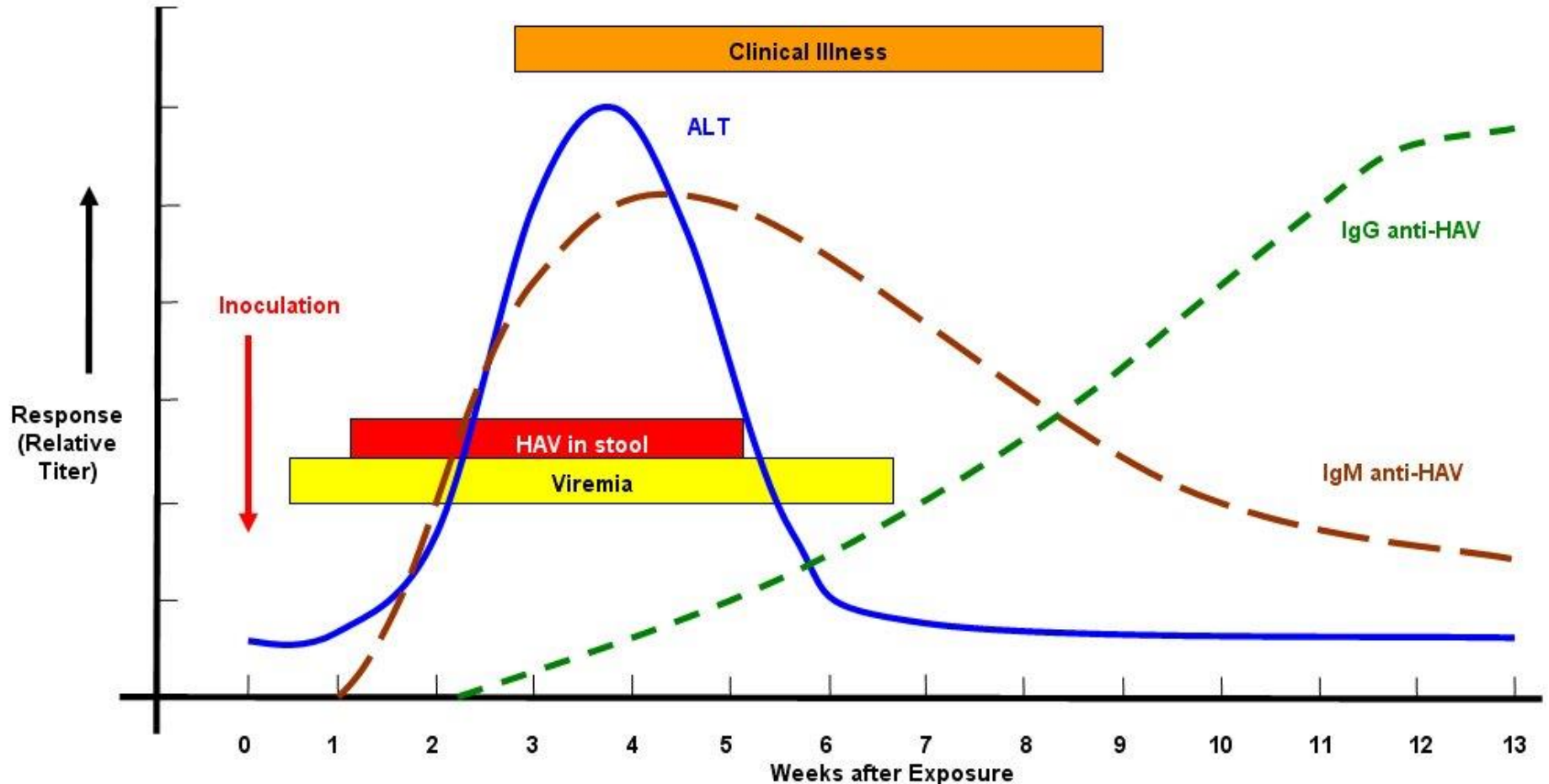
- **anti-VHA IgM**
 - marcador de infecção recente

- **anti-VHA IgG**
 - conferem imunidade duradoura
 - marcador soropidemiológico
 - indicador de contato prévio

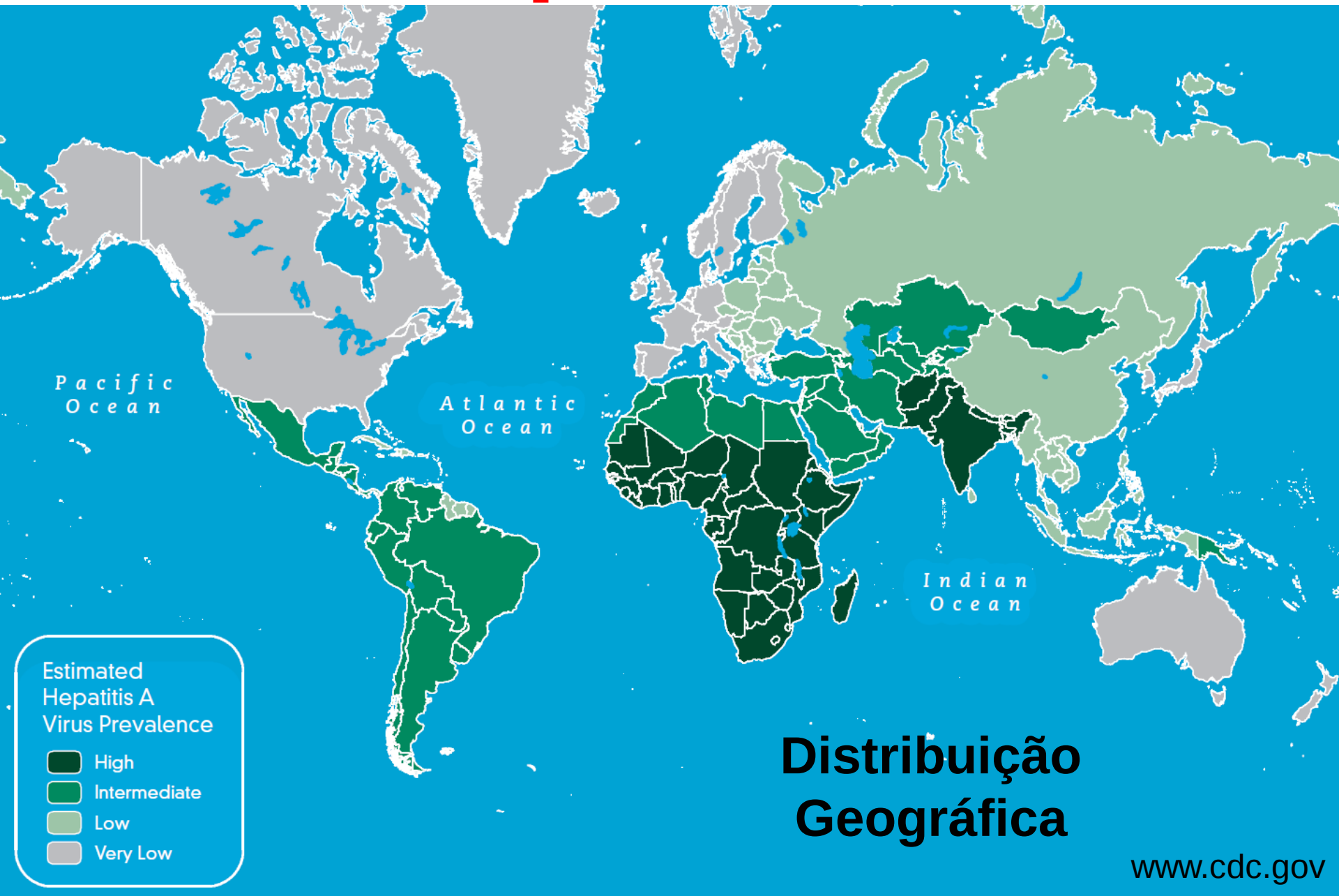
- **amostra clínica**
 - soro
 - saliva
 - sangue - polpa digital

Hepatite A

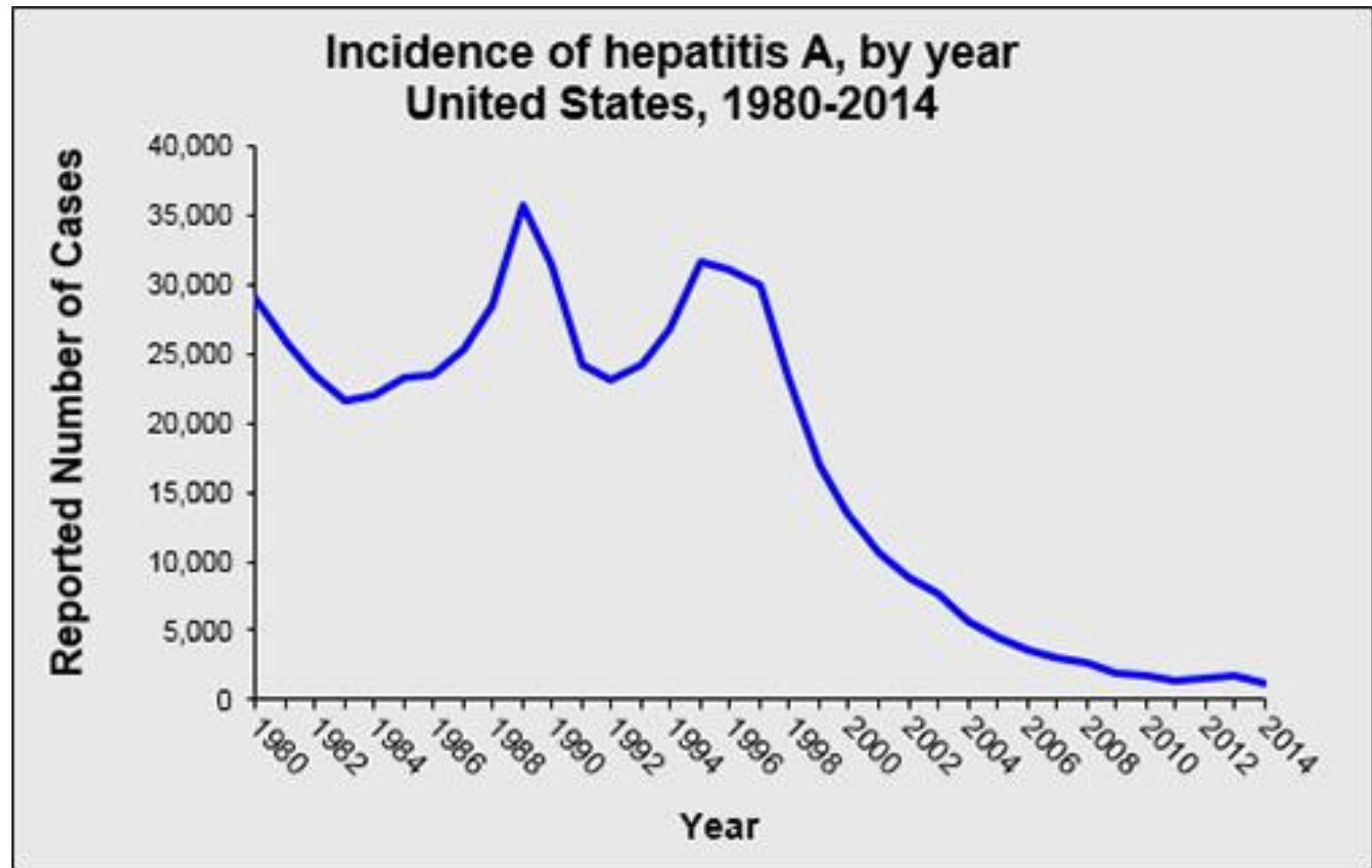
Sorologia da infecção



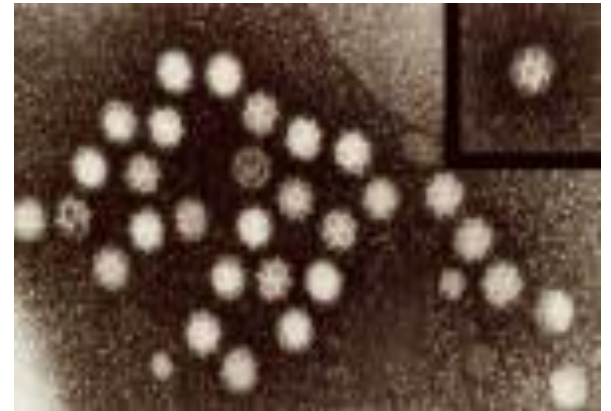
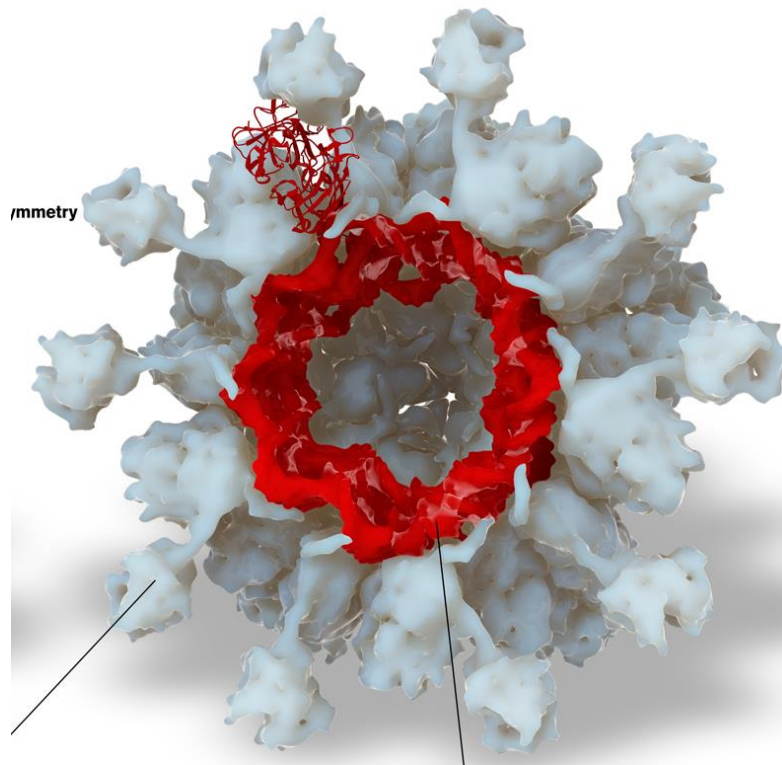
Hepatitis A



Hepatitis A



Hepatitis E (HEV)



Hepatite E

Família: Hepeviridae

Gênero: Hepevírus

- Capsídeo: icosaédrico, esférico, não envelopado, 27-34nm.
- Genoma de RNA com leitura positiva com 7,2 kb 3 ORF.
- transmissão fecal-oral e água contaminada (zoonose - suínos).
- Endêmico no Sudeste Asiático e Índia.
- Mortalidade de até 20%, especialmente em grávidas.

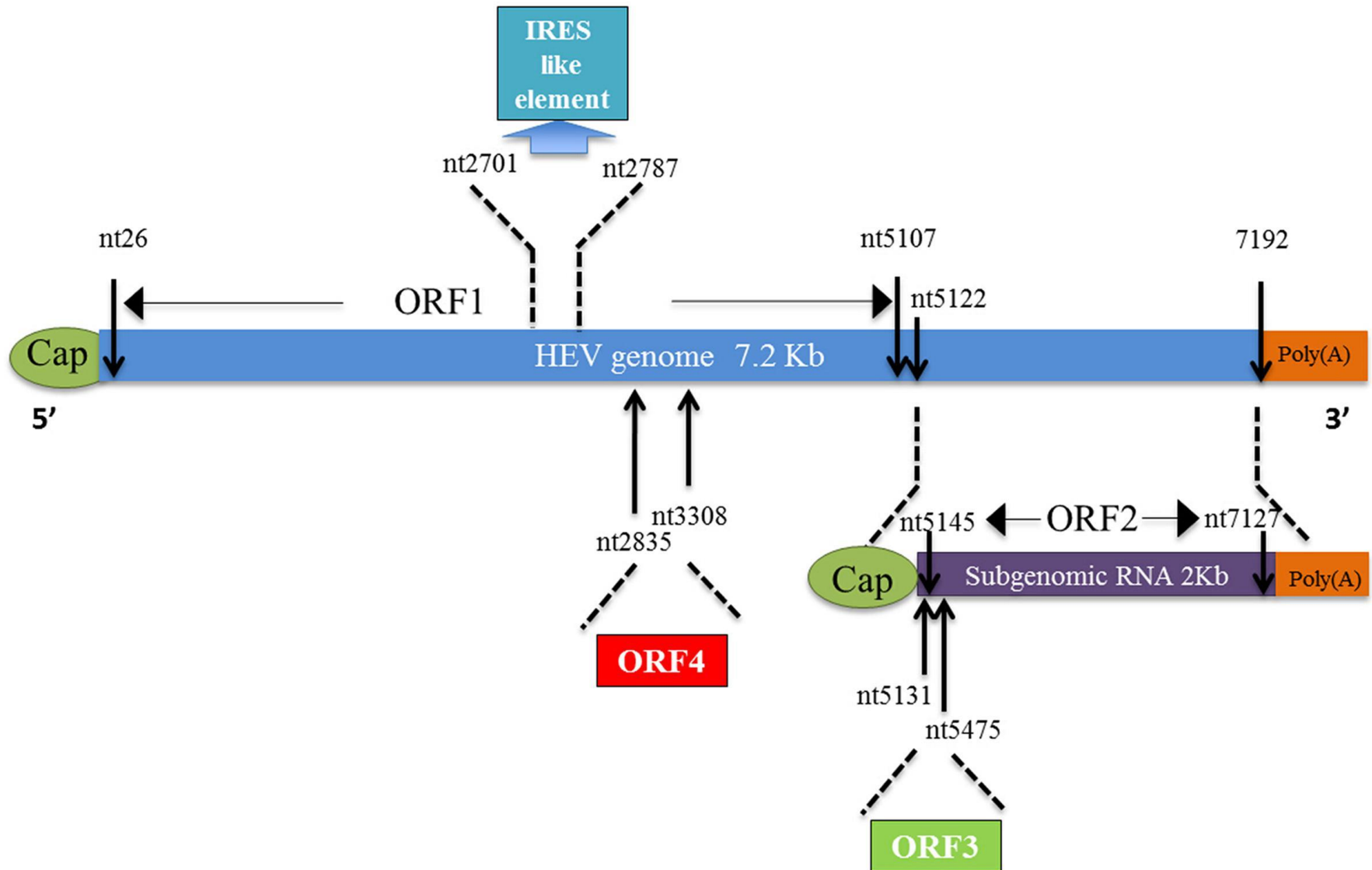
Hepatitis E

Family: <i>Hepadnaviridae</i>	(2 Genera)	History
Family: <i>Hepeviridae</i>	(2 Genera)	History
Genus: <i>Orthohepevirus</i>	(4 Species)	History
★ Species: <i>Orthohepevirus A</i>		History
Species: <i>Orthohepevirus B</i>		History
Species: <i>Orthohepevirus C</i>		History
Species: <i>Orthohepevirus D</i>		History
Genus: <i>Piscihepevirus</i>	(1 Species)	History
★ Species: <i>Piscihepevirus A</i>		History

Genus	Species	Genotype	Natural hosts	Infection to humans
<i>Orthohepevirus</i>	<i>Orthohepevirus A</i>	1	Human	Yes
		2	Human	Yes
		3	Human, pig, rabbit, deer, mongoose, wild boar	Yes
		4	Human, pig, yak, wild boar	Yes
		5	Wild boar	Unknown
		6	Wild boar	Unknown
		7	Camel	Yes
	<i>Orthohepevirus B</i>		Chicken	No
	<i>Orthohepevirus C</i>	C1	Rat	No
		C2	Ferret	No
	<i>Orthohepevirus D</i>		Bat	No
<i>Piscihepevirus</i>	<i>Piscihepevirus A</i>		Trout	No

Hepatitis E

Organização genômica



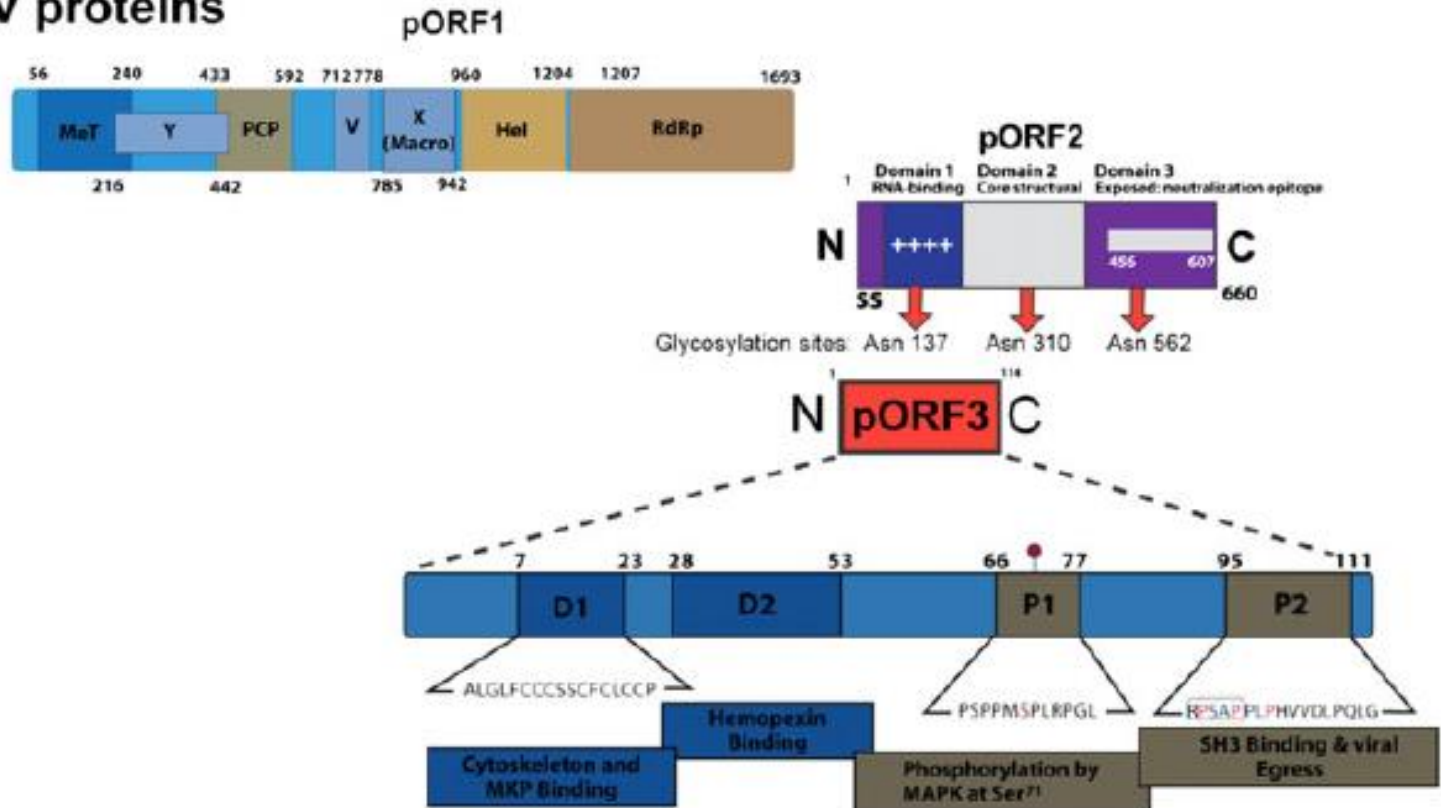
Hepatitis E

Organização genômica e expressão

(c) ORFs



(d) HEV proteins



Hepatite E

Diagnóstico Laboratorial

- ensaios imunoenzimáticos

Pesquisa de anti-VHE (IgM ou IgG) - ELISA

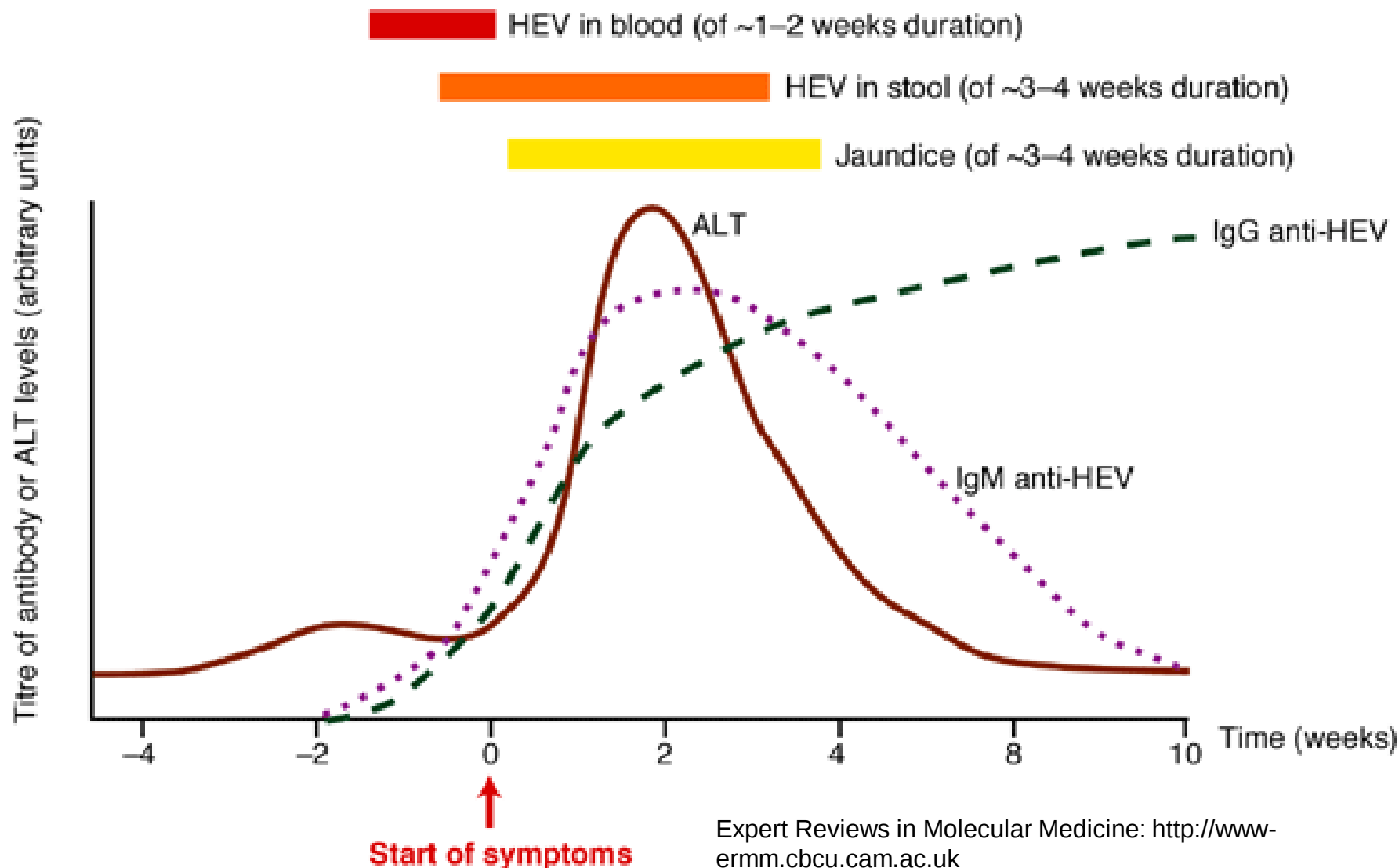
- ELISA

- IMUNOBLOT

- técnicas de biologia molecular

Hepatitis E

Sorologia da infecção

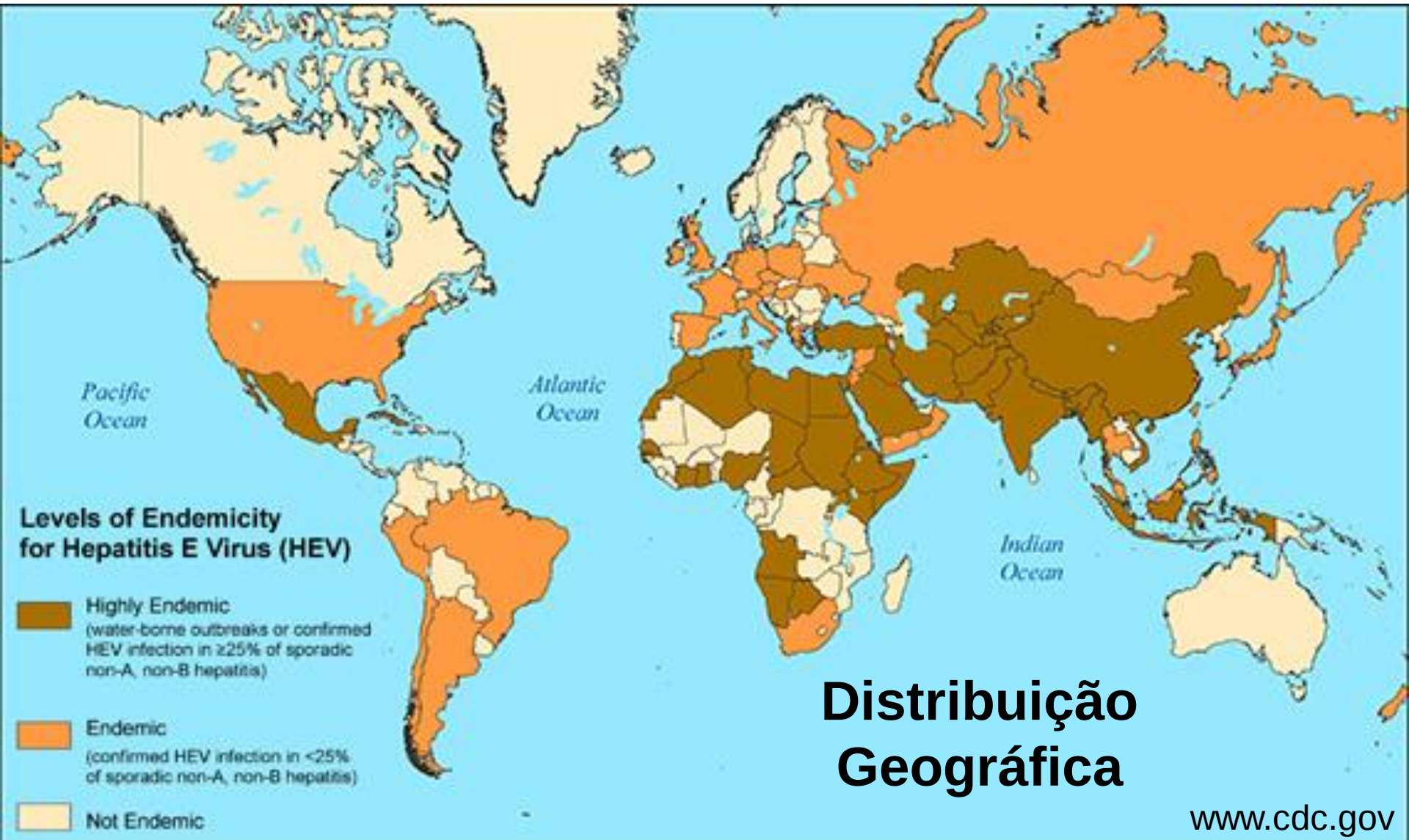


Hepatite E

Epidemiologia

- Maioria dos surtos associados a consumo de água contaminada com fezes
- Transmissão pessoa-a-pessoa mínima
- Casos descritos em áreas endêmicas ou em pessoas que viajaram para estas regiões

Hepatite E



Hepatitis E

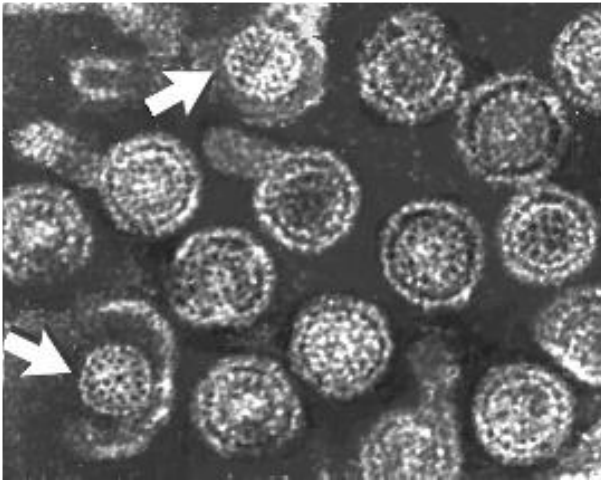
Table 1 Comparison of the four hepatitis E virus genotypes by select characteristics

Characterstics	Genotype 1	Genotype 2	Genotype3	Genotype 4
Viral discovery	1983	1986	1995	2003
Geographic distribution	Developing countries	Mexico, West Africa	Developed countries	China, Taiwan, Japan
Food-borne transmission	No	No	Yes	Yes
Fecal-oral transmission	Yes	Yes	?	No
Water-borne transmission	Yes	Yes	?	No
Person-to-person transmission	Yes	Unknown	Yes	Unknown
Zoonotic transmission	No	No	Yes	Yes
Occurrence of epidemics	Common	Smaller scale epidemics	No epidemics	Uncommon
Highest attack rate	Young adults	Young adults	Persons $\geq$ 40 yr of age	Young adults
Gender	Male preponderance	Not discriminatory	Mostly male	Not discriminatory
Mortality rate	0.5%-3%	0.5%-3%	Not determined	0.5%-3%
Mortality among pregnant women	High	High	Not determined	High
Chronic infection	None	None	Yes	None
Severe disease among immuno-compromised	Not reported	Not reported	Yes	Not reported
Interspecies transmission	Only humans and non-human primates	Only humans and non-human primates	Humans Pigs	Humans Pigs
Subtypes	5	2	10	7

**Vírus de hepatite
de transmissão
parenteral
(sexual?)**

Hepatitis B (HBV)

Hepatite B



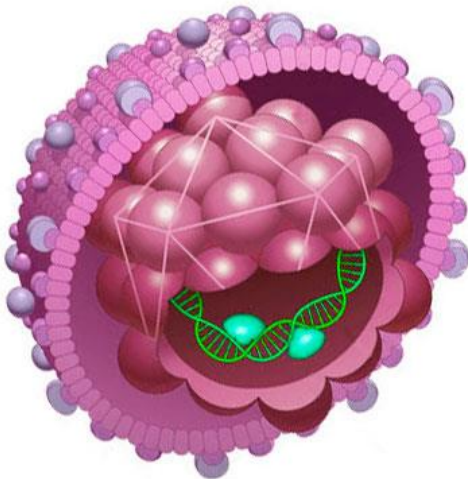
Familia : *Hepadnaviridae*
Gênero: *Orthohepadnavírus*

- Pararetrovírus

Vírus pequenos de DNA envelopados (~45 nm)

DNA circular dupla-fita parcial (~3,5 kpb)

Célula alvo: hepatócitos

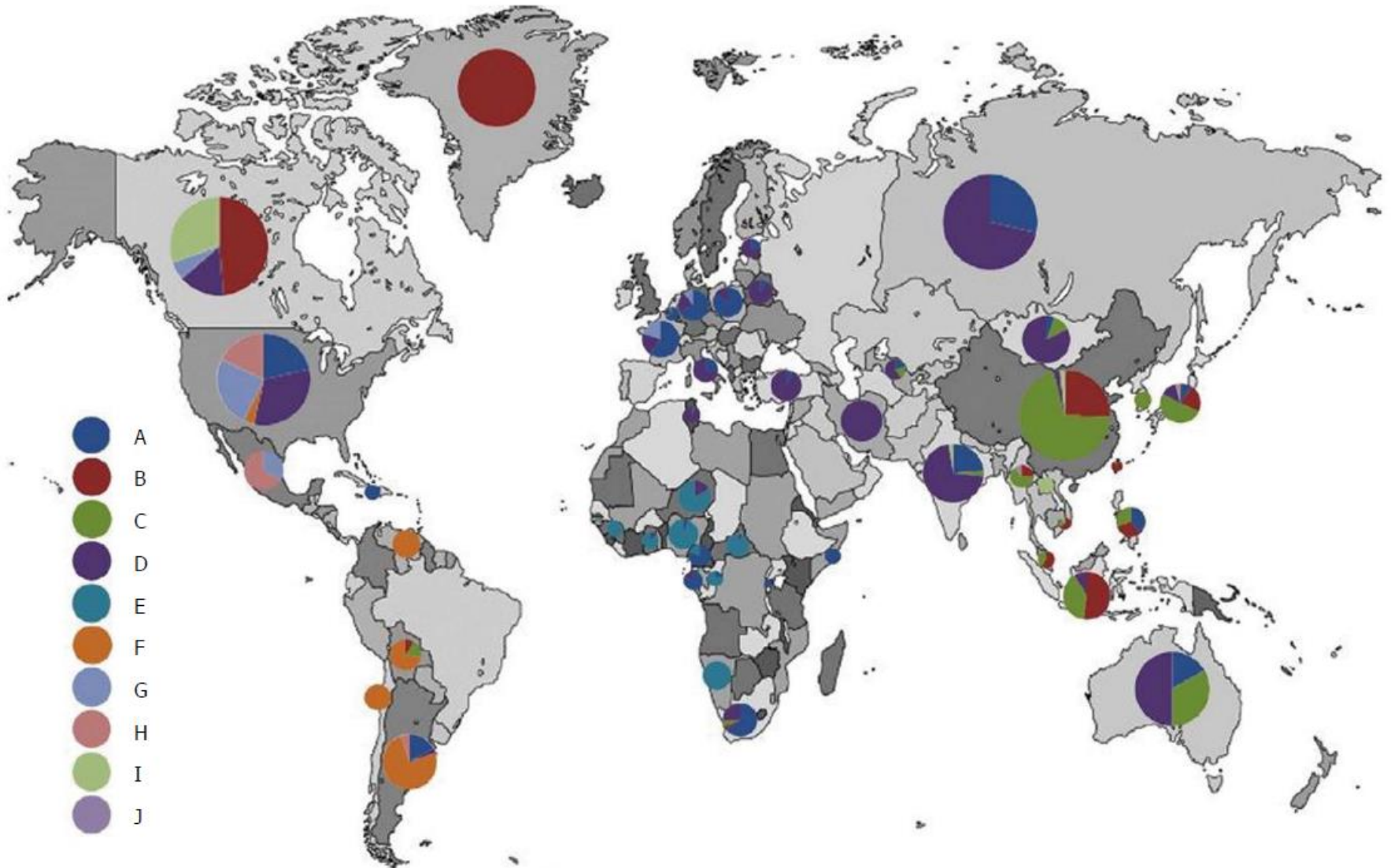


Etiologicamente associado com:

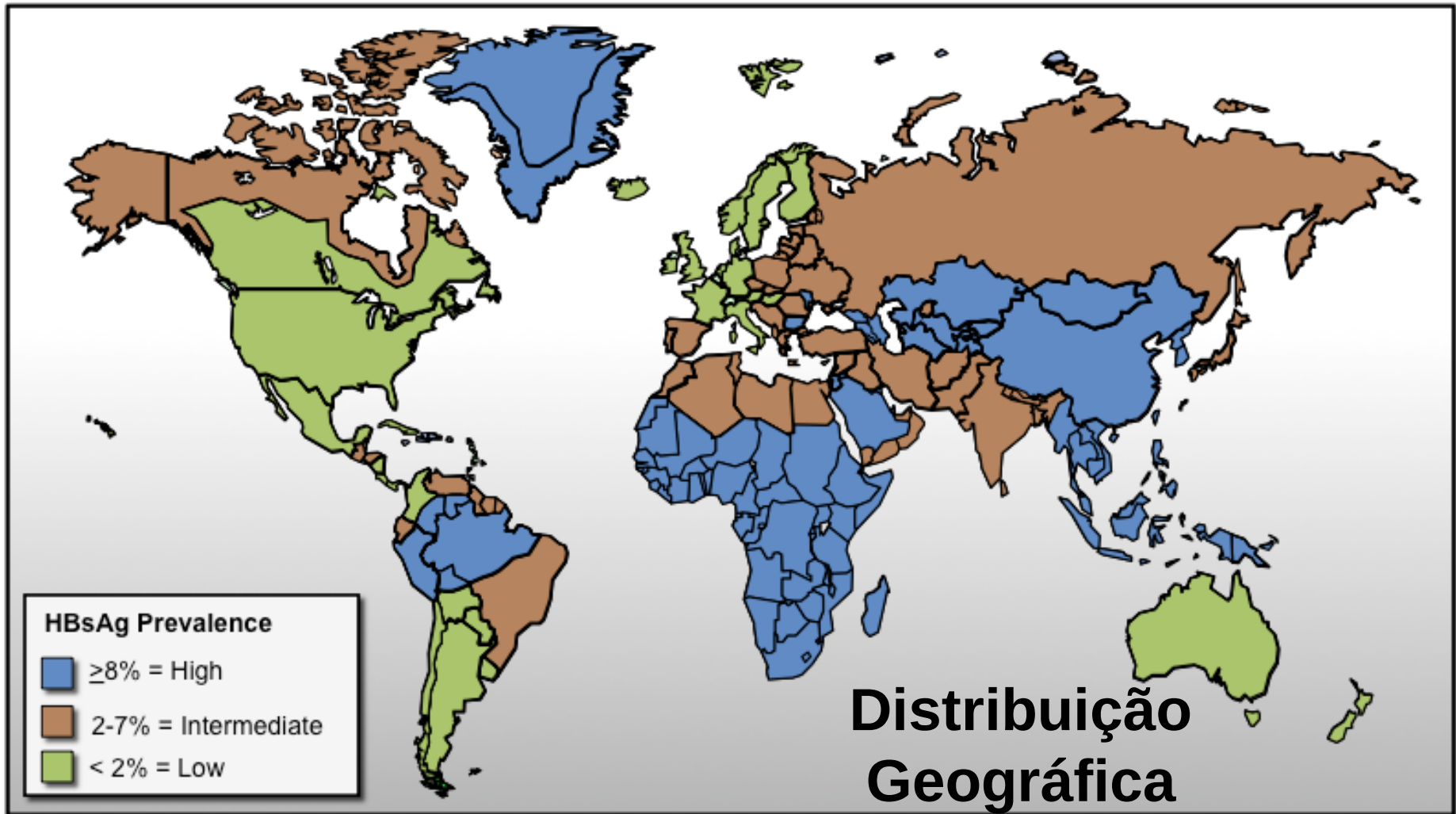
- Hepatite
- Cirrose
- Carcinoma hepatocelular

Hepatite B

Dez genótipos

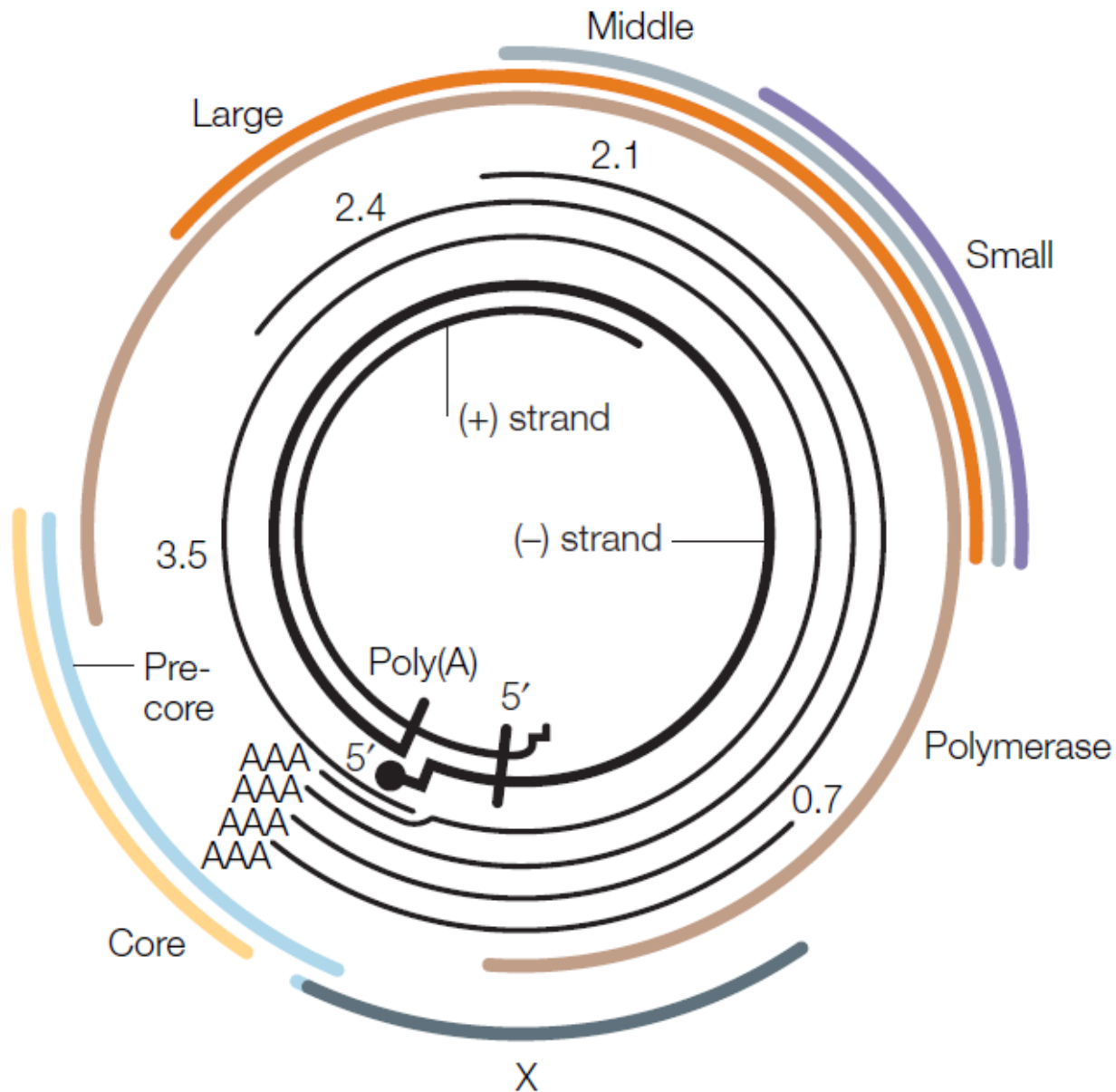


Hepatite B



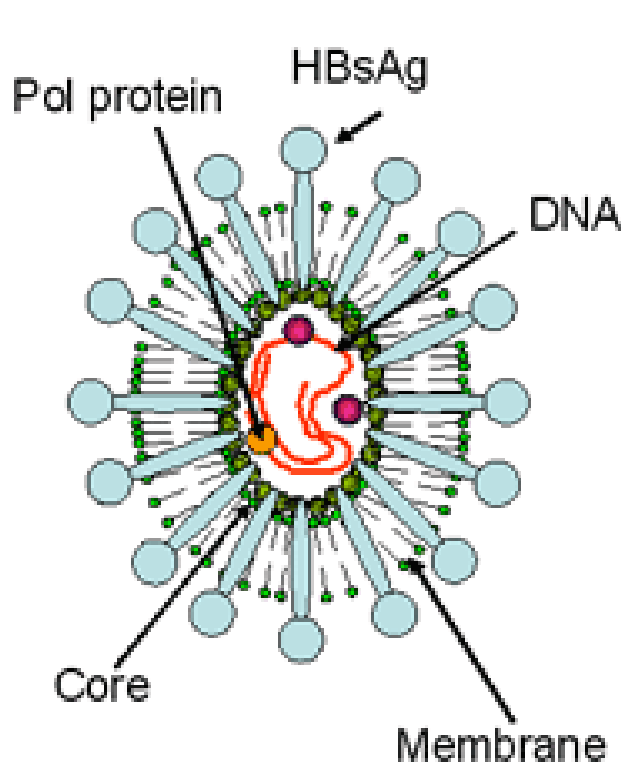
Hepatitis B

Organização genômica

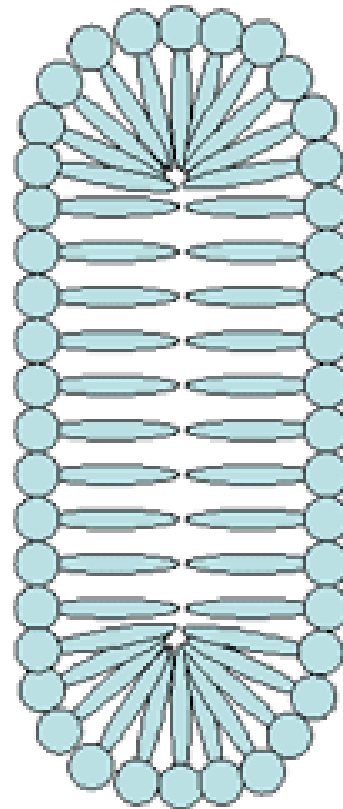


Hepatitis B

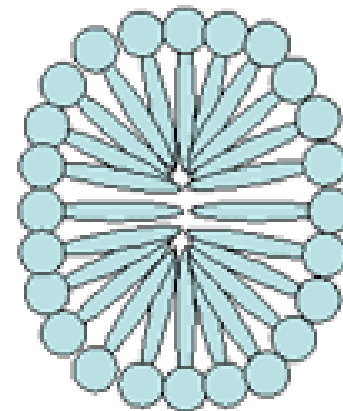
Antígenos virais



Virus
Dane particle
40nm
diameter



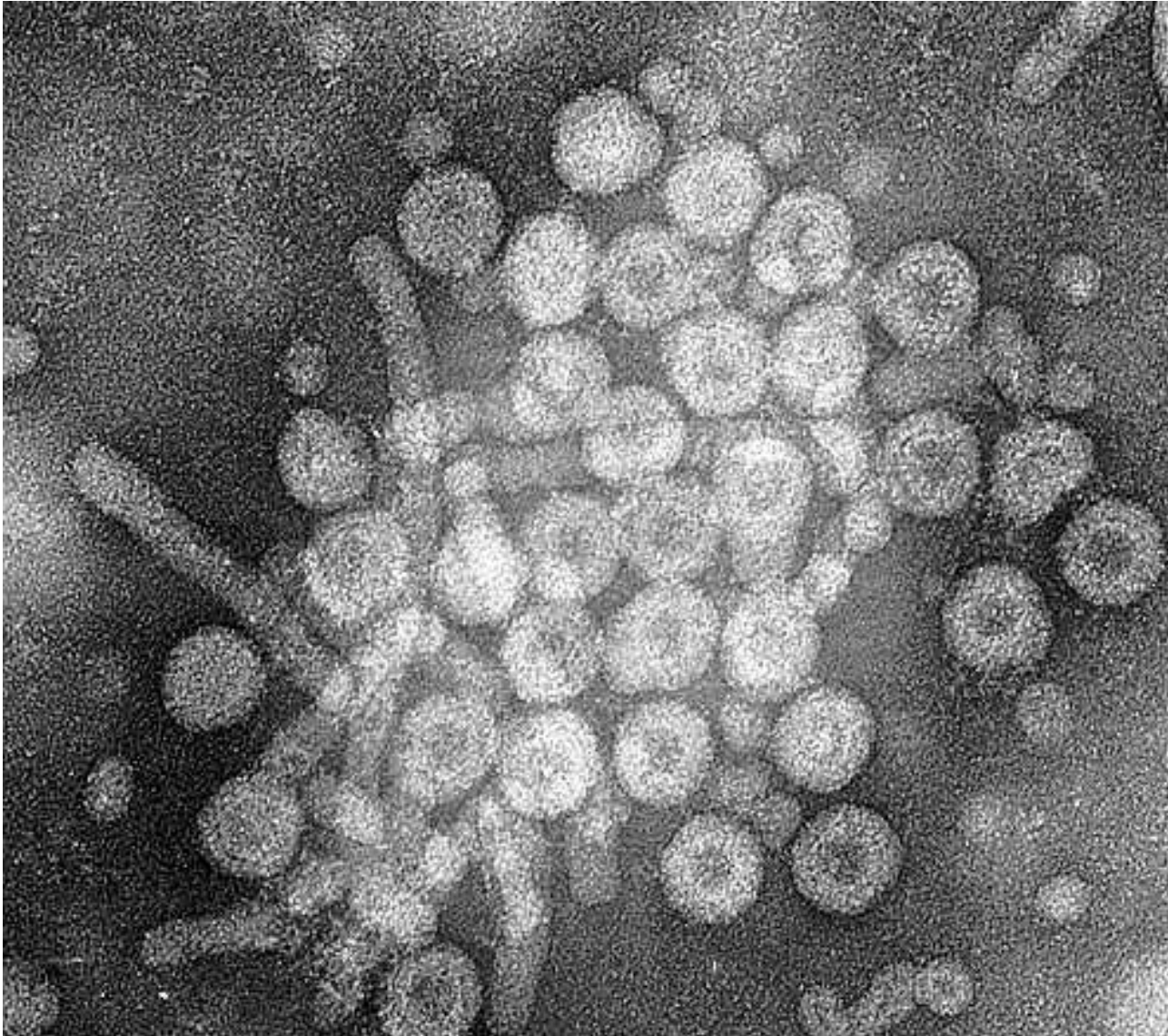
Filamentous particle
Up to 200nm long



Spherical particle
~20nm diameter

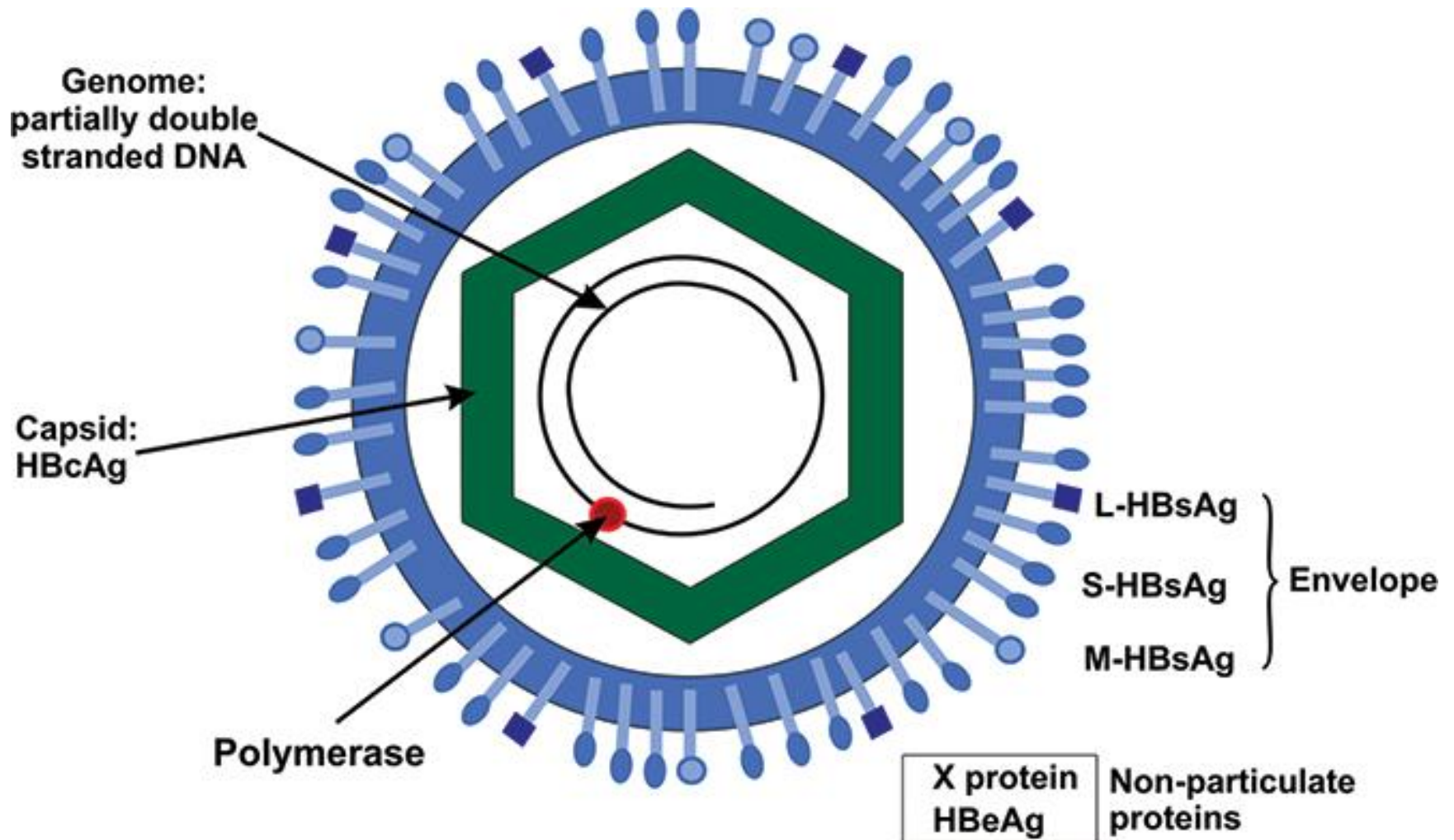
Hepatite B

Antígenos virais



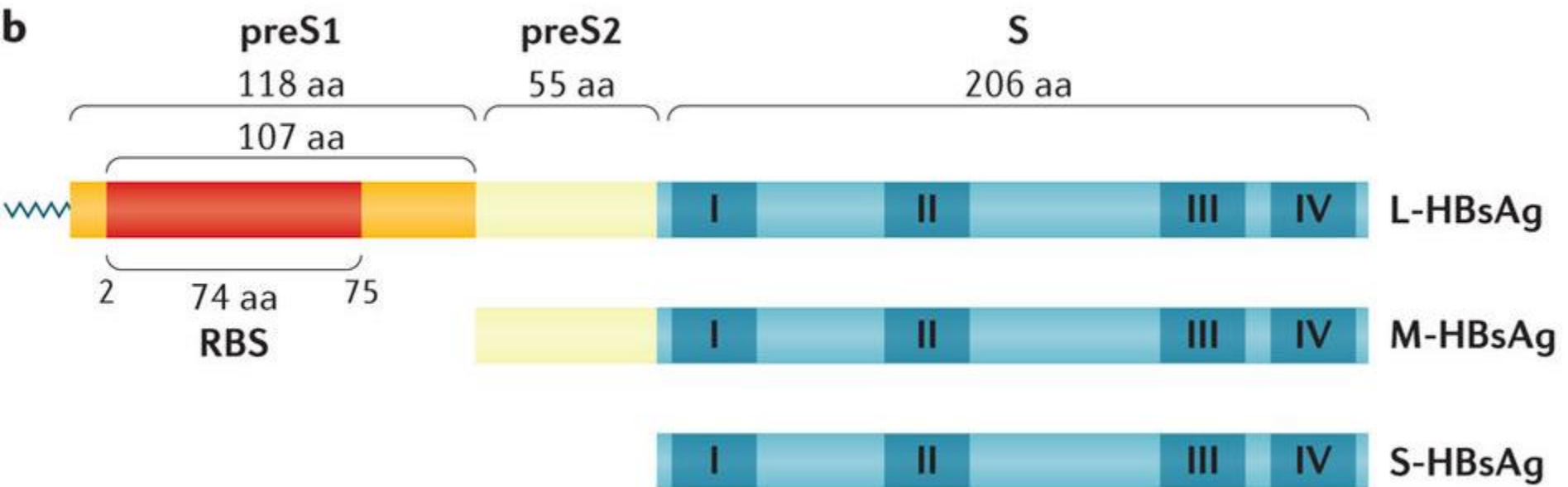
Hepatitis B

Antígenos virais



Hepatitis B

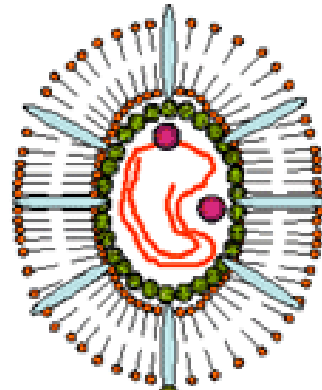
Antígenos virais



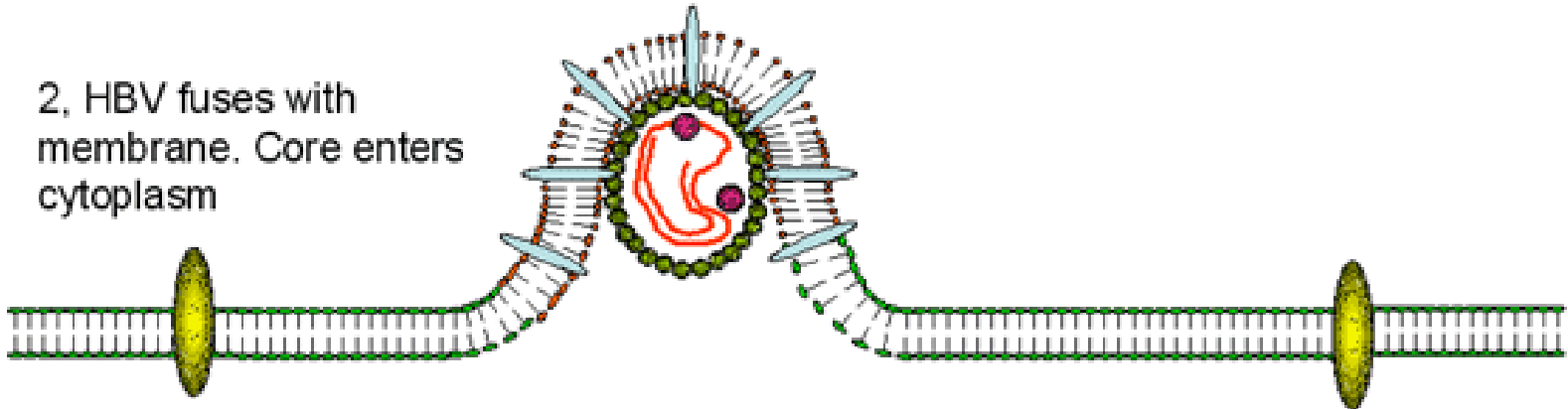
Hepatitis B

Ciclo Viral

1. HBV fuses binds to host cell membrane



2. HBV fuses with membrane. Core enters cytoplasm

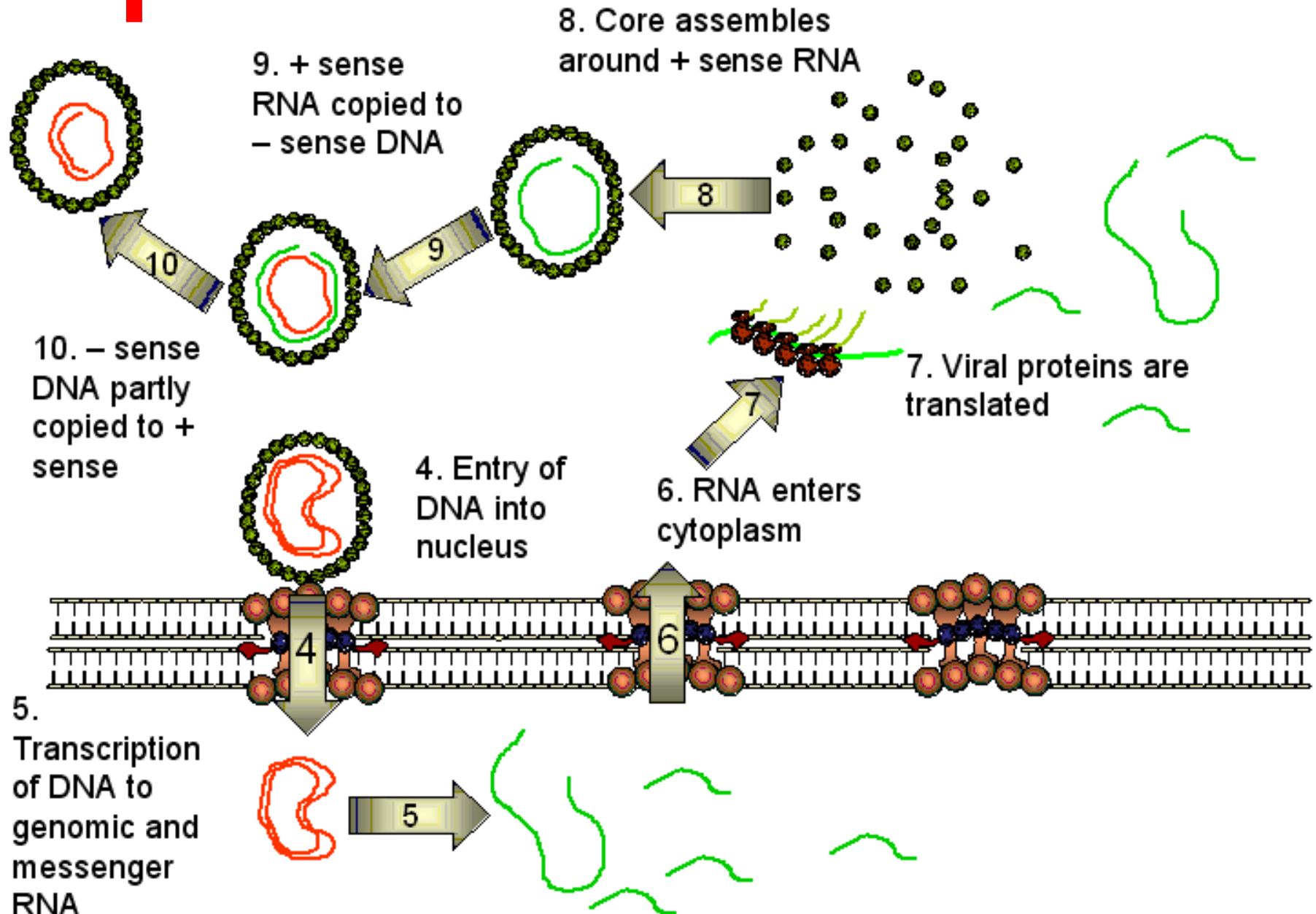


3. Double stranded genome is completed by enzymes contained with the core



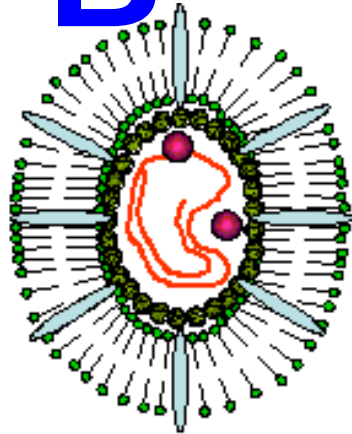
Hepatitis B

Ciclo Viral

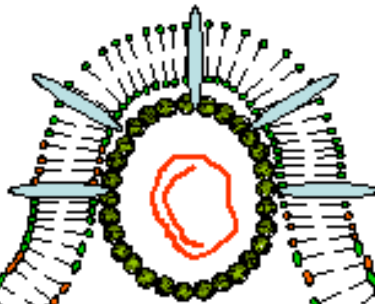


Hepatitis B

Ciclo Viral



13. Virus with partly double strand DNA leaves the cell



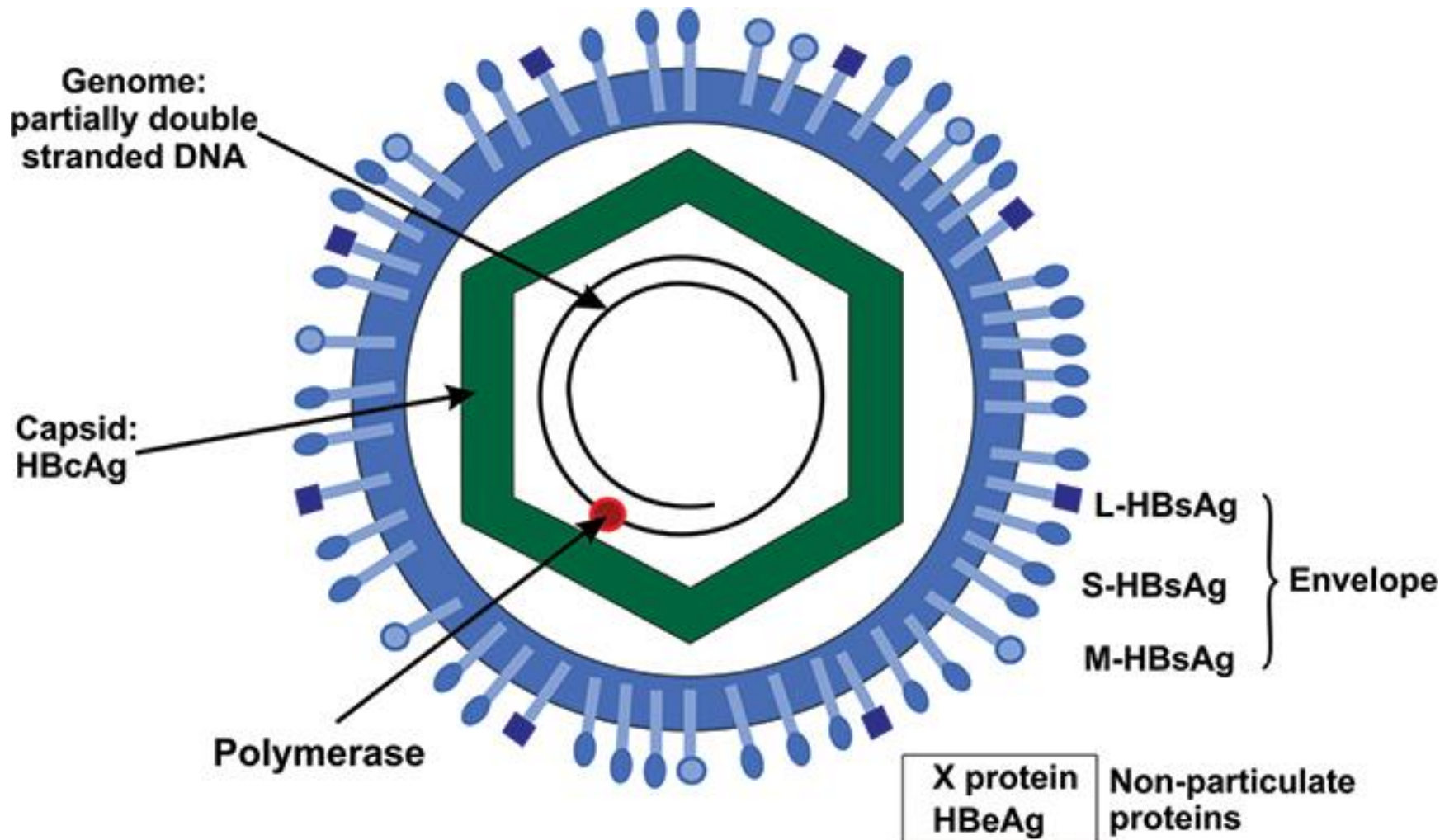
12. Virus buds through host cell membrane



11. Core associates with HbS antigen in cell membrane

Hepatitis B

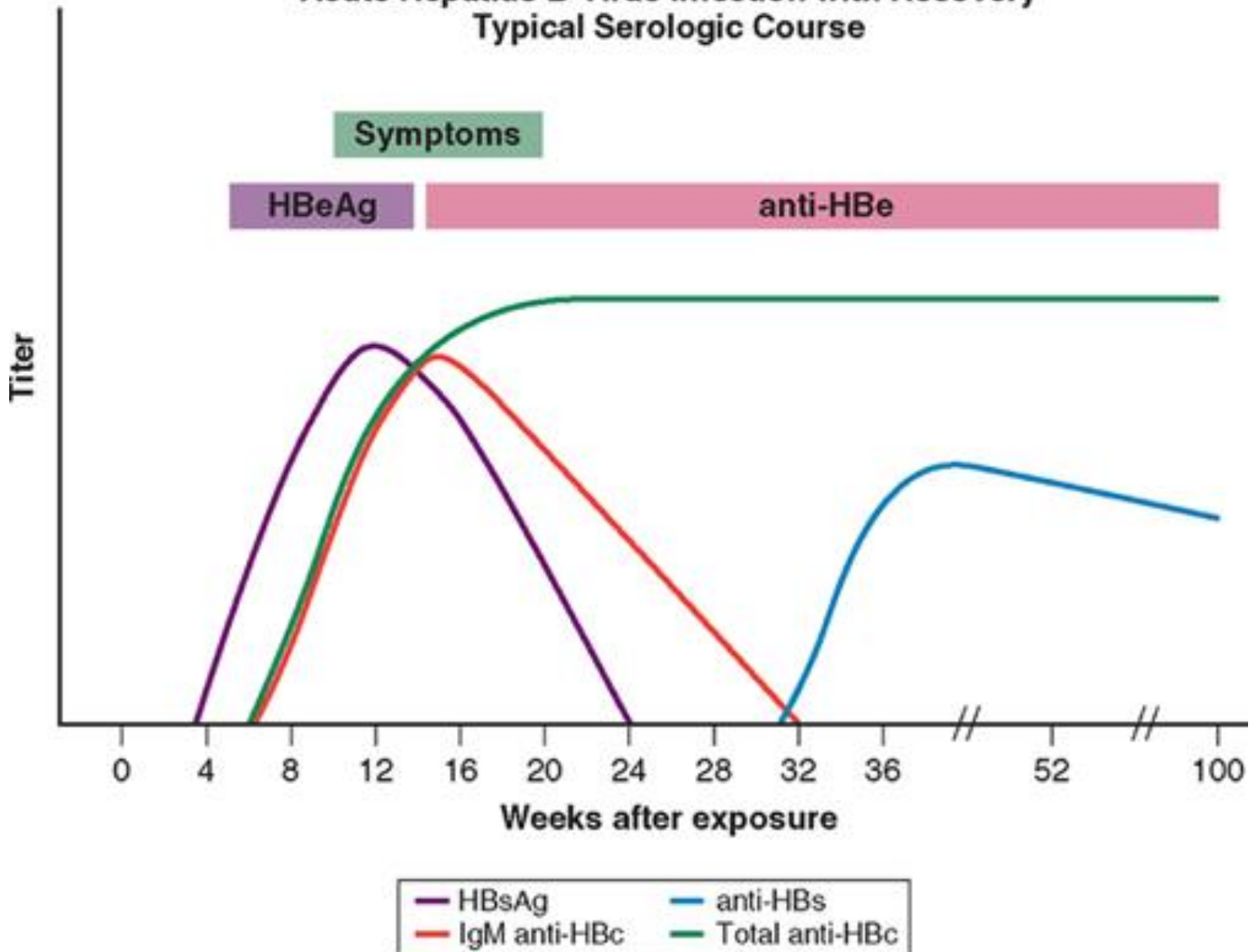
Antígenos virais



Hepatitis B

Antígenos virais

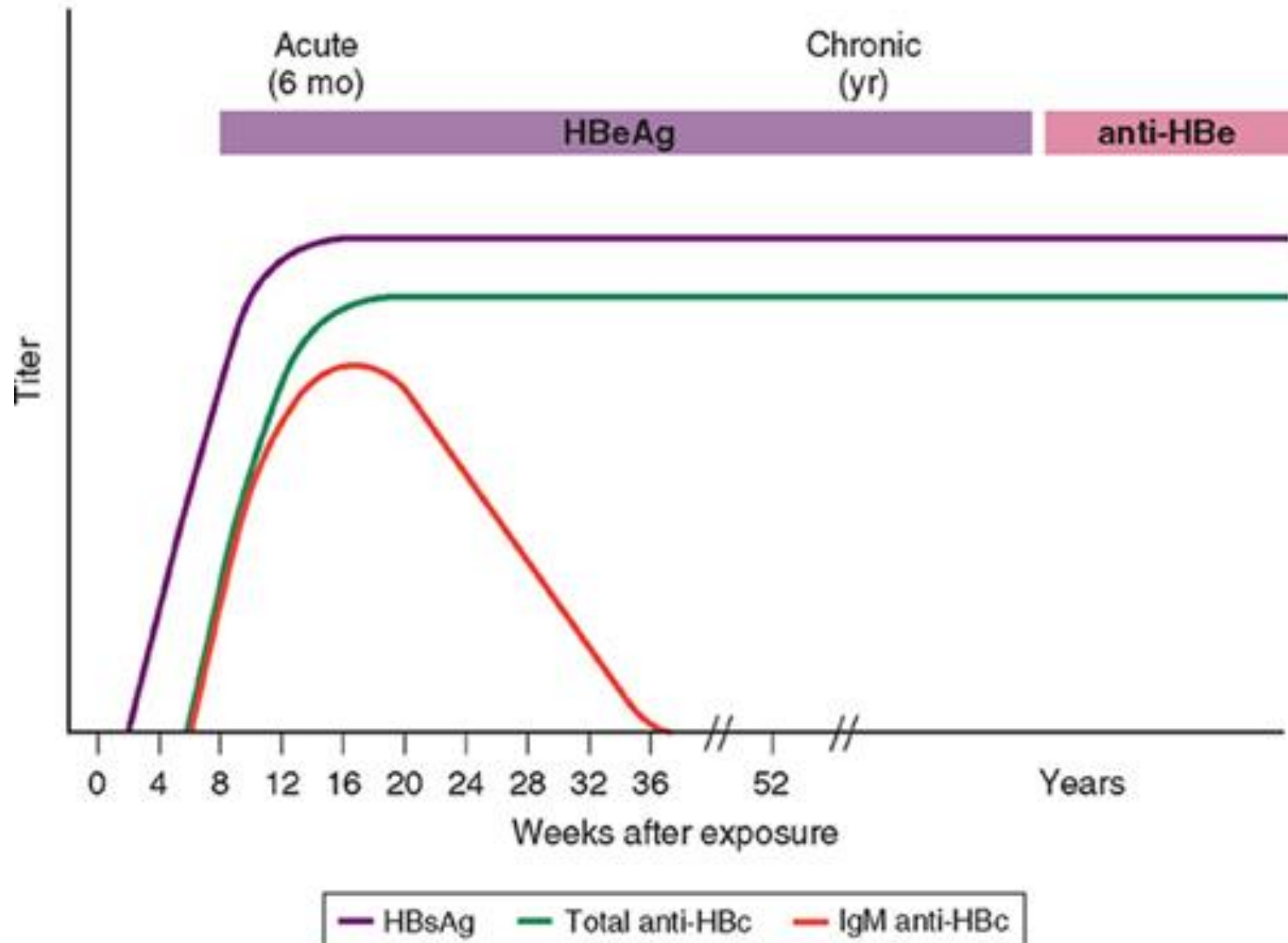
Acute Hepatitis B Virus Infection with Recovery
Typical Serologic Course



Hepatitis B

Antígenos virais

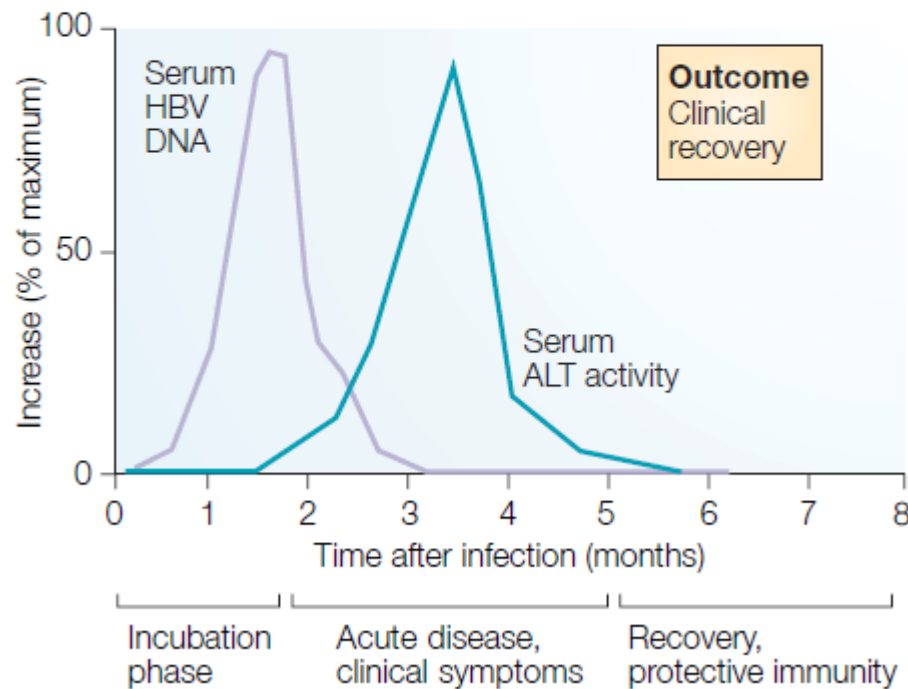
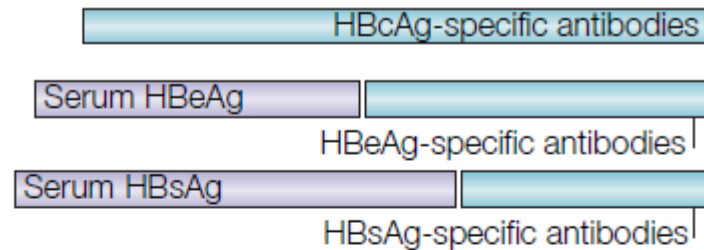
Progression to Chronic Hepatitis B Virus Infection
Typical Serologic Course



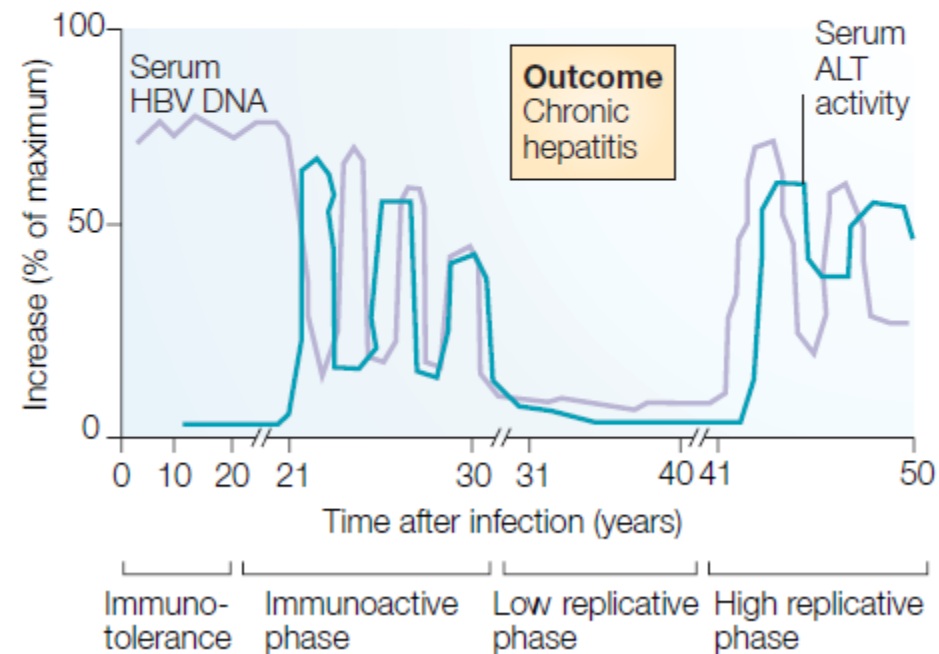
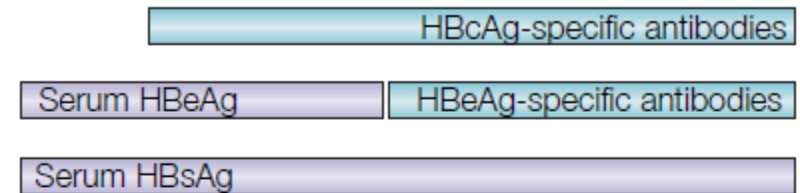
Hepatitis B

Antígenos virais

a Hepatitis B (acute)



b Hepatitis B (chronically evolving)



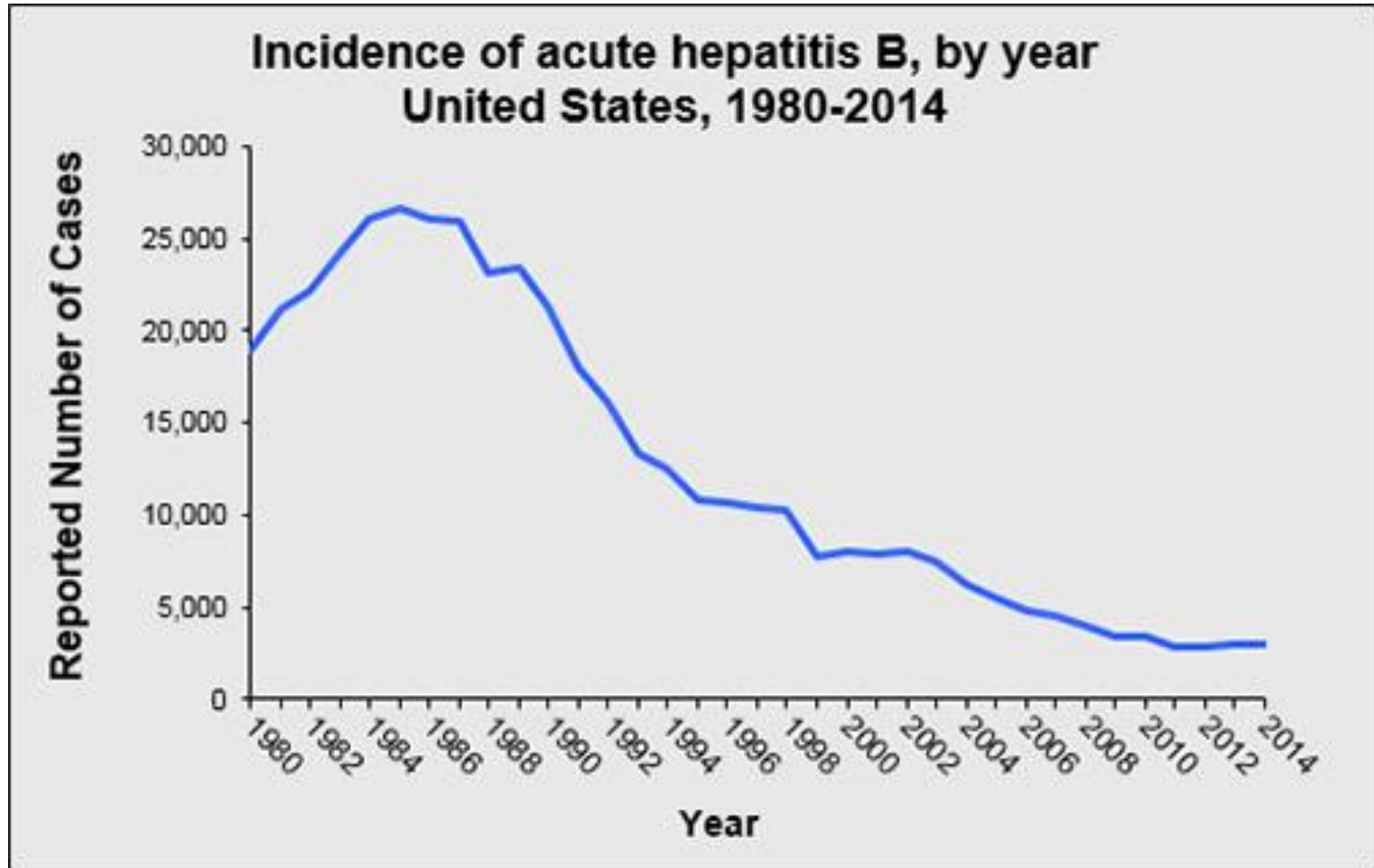
Interpretation of Hepatitis B Serologic Tests

Tests	Results	Interpretation
HBsAg	Negative	Susceptible
anti-HBc	Negative	
anti-HBs	Negative	
HBsAg	Negative	Immune due to vaccination
anti-HBc	Negative	
anti-HBs	Positive with $\geq 10\text{mIU/mL}^*$	
HBsAg	Negative	Immune due to natural infection
anti-HBc	Positive	
anti-HBs	Positive	
HBsAg	Positive	Acutely infected
anti-HBc	Positive	
IgM anti-HBc	Positive	
anti-HBs	Negative	
HBsAg	Positive	Chronically infected
anti-HBc	Positive	
IgM anti-HBc	Negative	
anti-HBs	Negative	
HBsAg	Negative	Four interpretations possible [†]
anti-HBc	Positive	
anti-HBs	Negative	

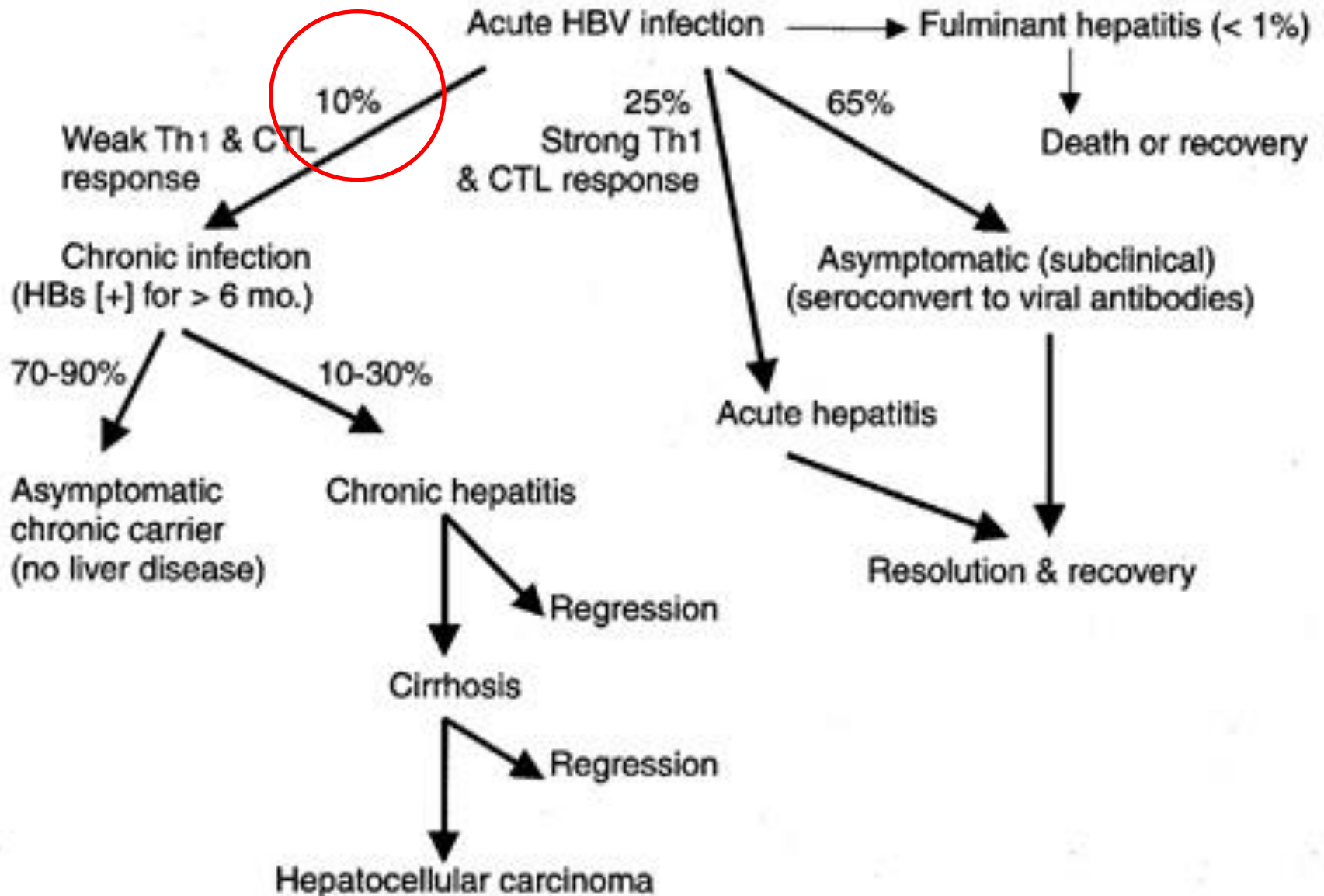
*Postvaccination testing, when it is recommended, should be performed 1-2 months following dose #3.

- †
1. May be recovering from acute HBV infection.
 2. May be distantly immune and the test is not sensitive enough to detect a very low level of anti-HBs in serum.
 3. May be susceptible with a false positive anti-HBc.
 4. May be chronically infected and have an undetectable level of HBsAg present in the serum.

Hepatitis B



Hepatitis B



Hepatitis C (HCV)

Hepatite C

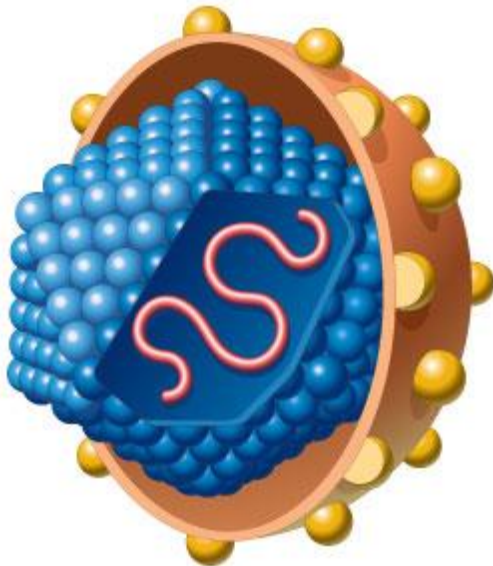


Familia : *Flaviviridae*
Gênero: *Hepacivírus*

Vírus pequenos de RNA envelopados (~45 nm)

RNA linear simples-fita (~9,4 kpb)

Célular alvo: hepatócitos

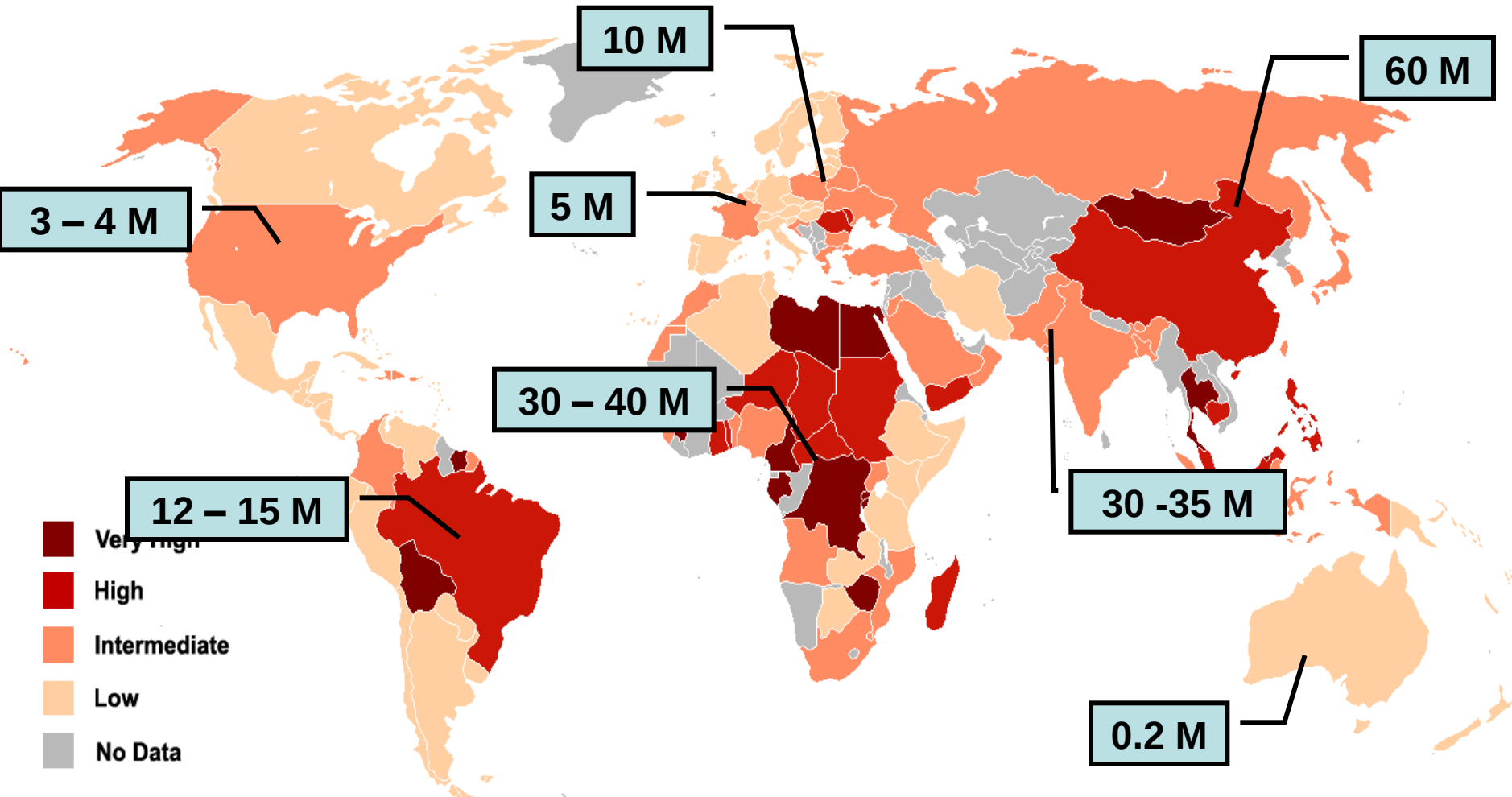


Etiologicamente associado com:

- Hepatite
- Cirrose
- Carcinoma hepatocelular

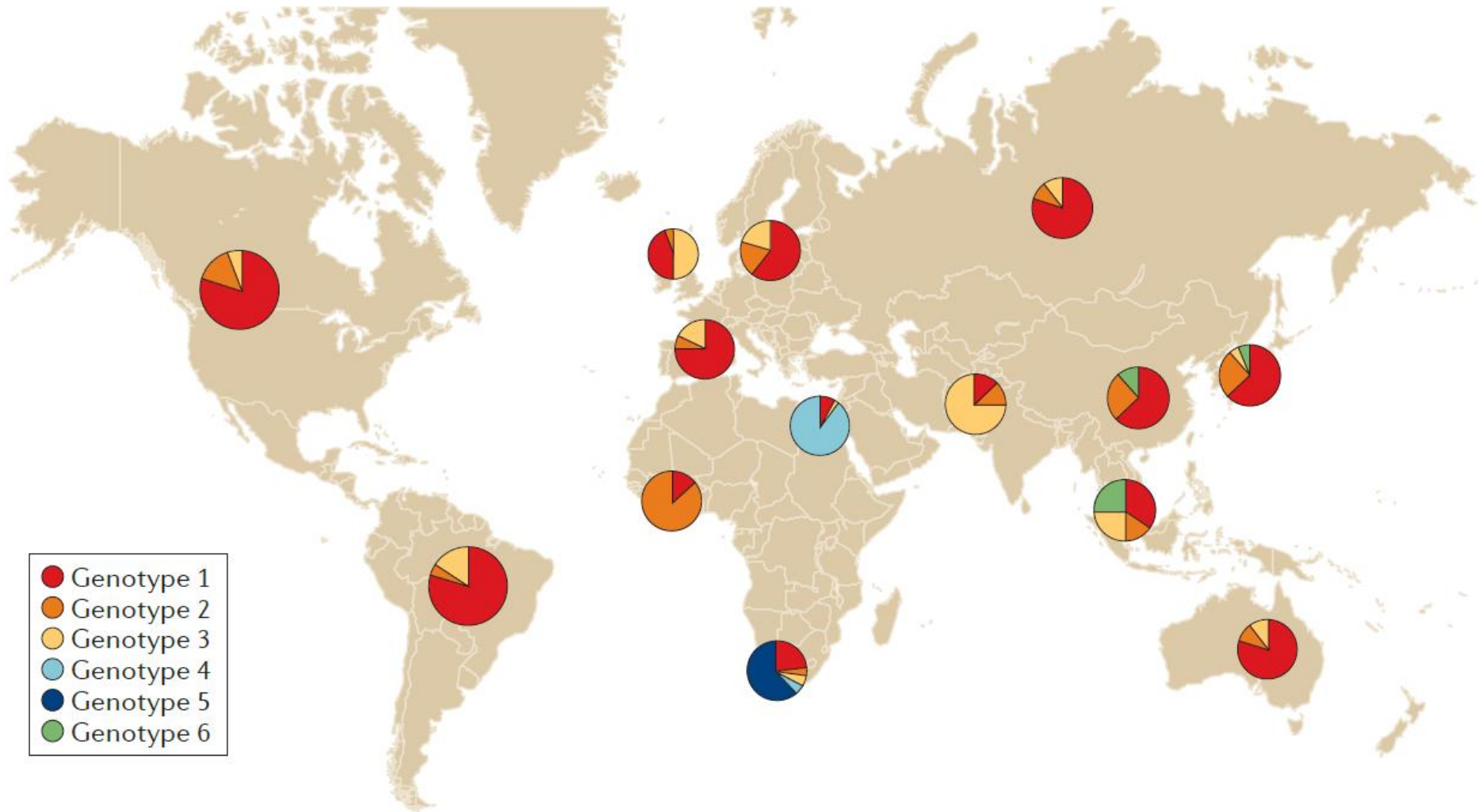
Hepatite C

170 – 200 milhões de portadores no Mundo todo



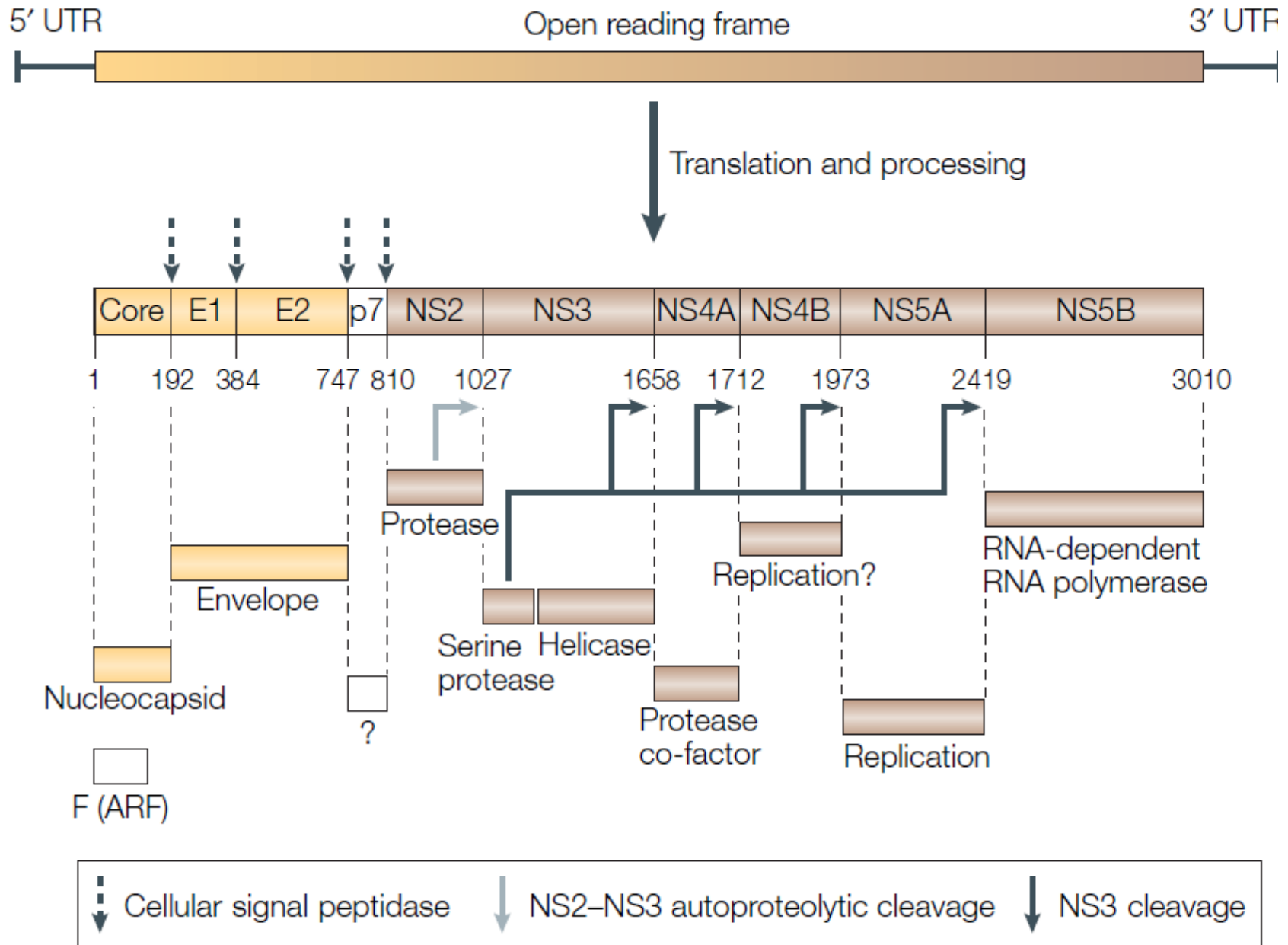
Hepatite C

170 – 200 milhões de portadores no Mundo todo



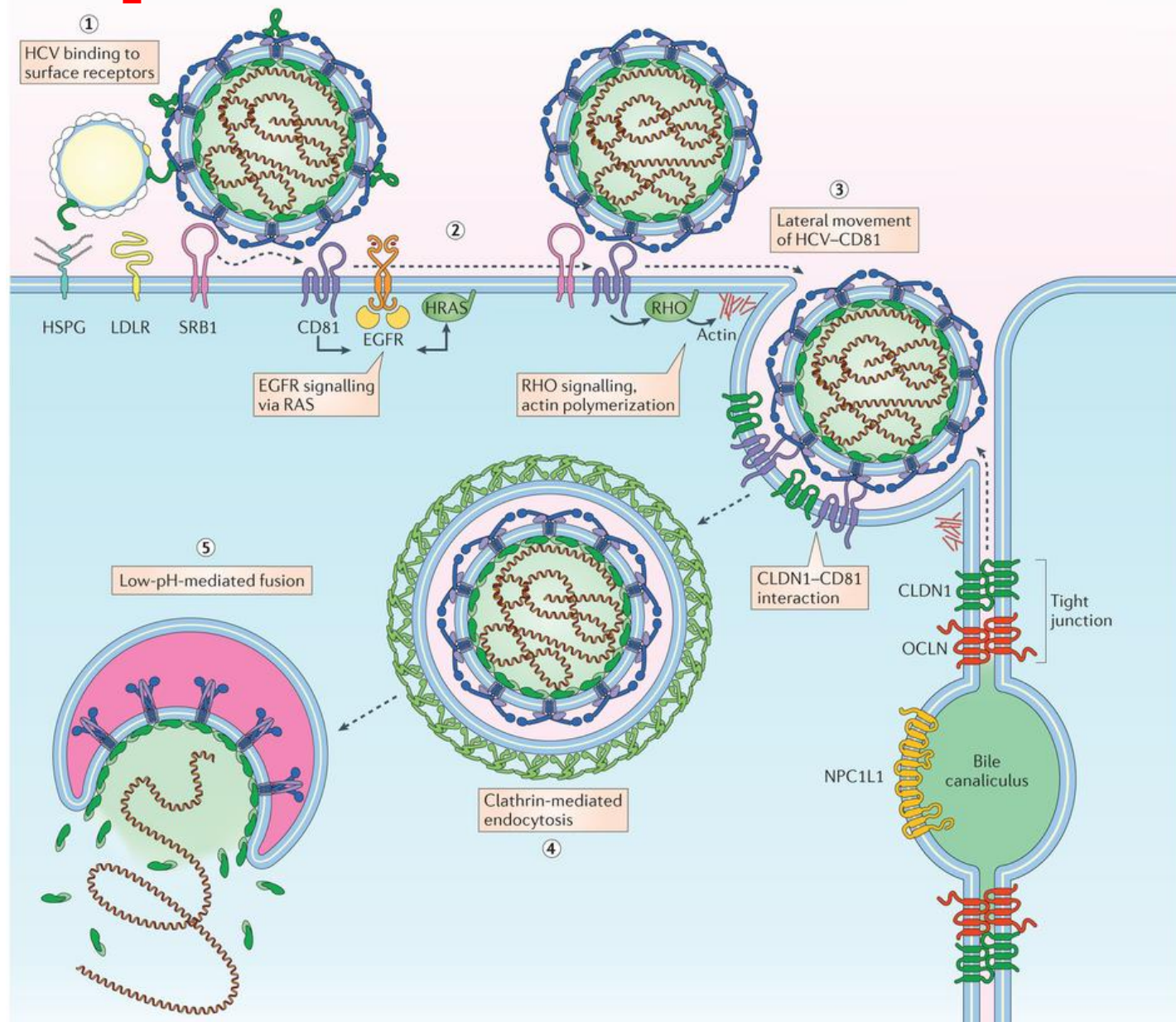
Hepatitis C

b Genomic structure of HCV



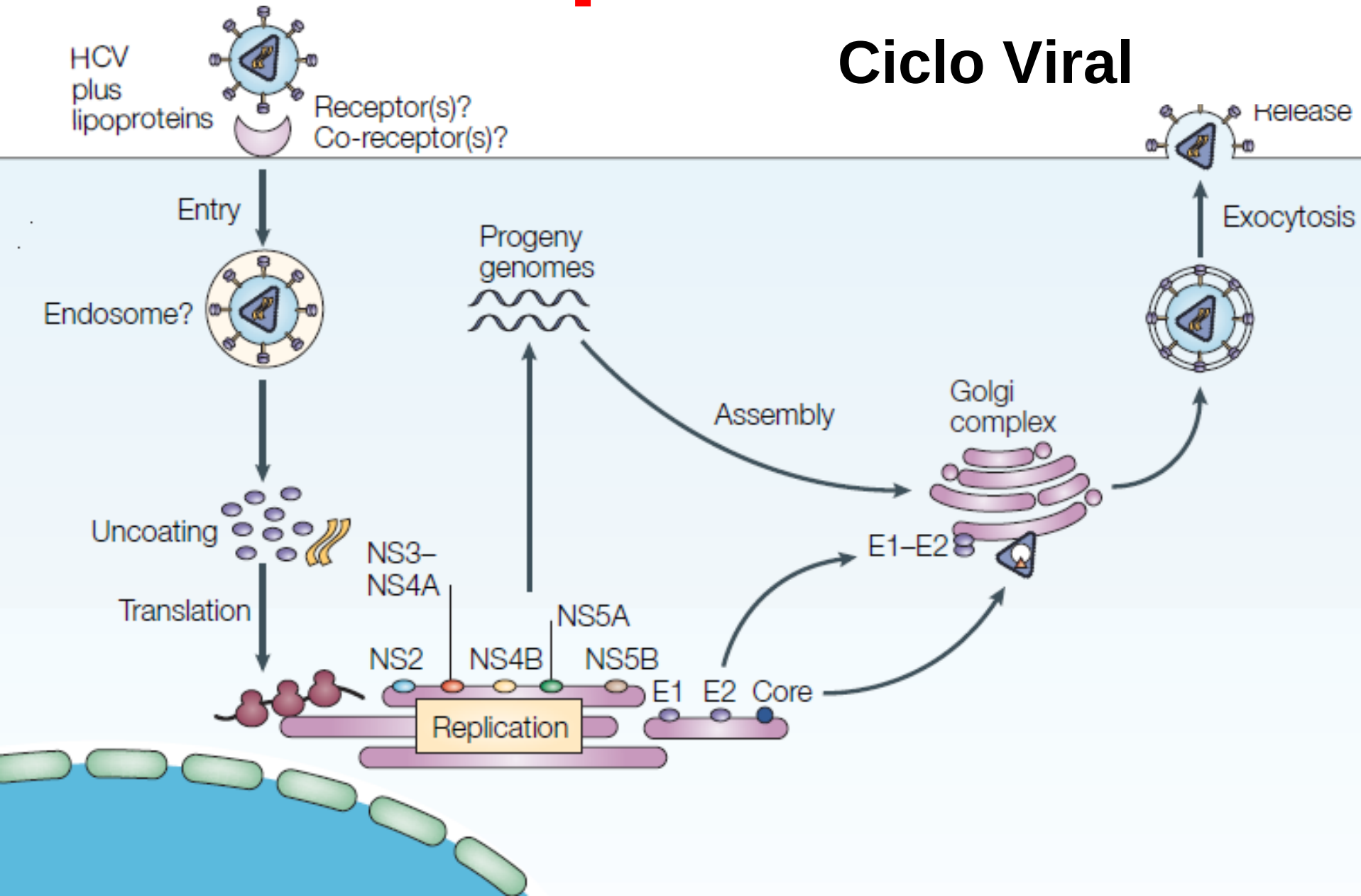
Hepatitis C

Ciclo Viral



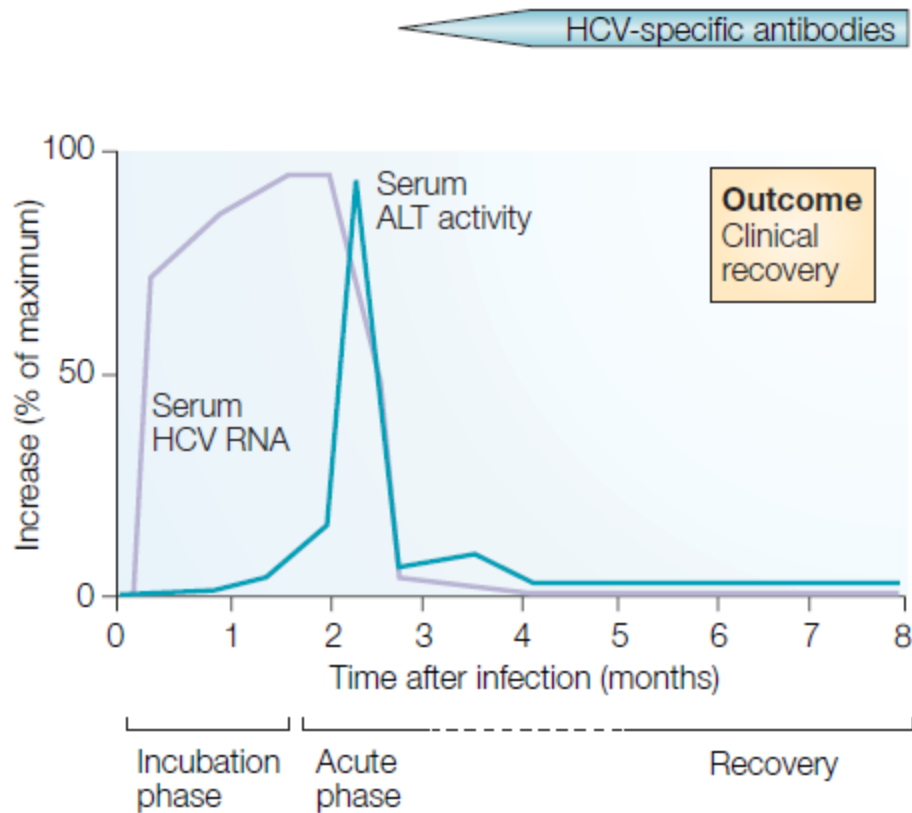
Hepatitis C

Ciclo Viral



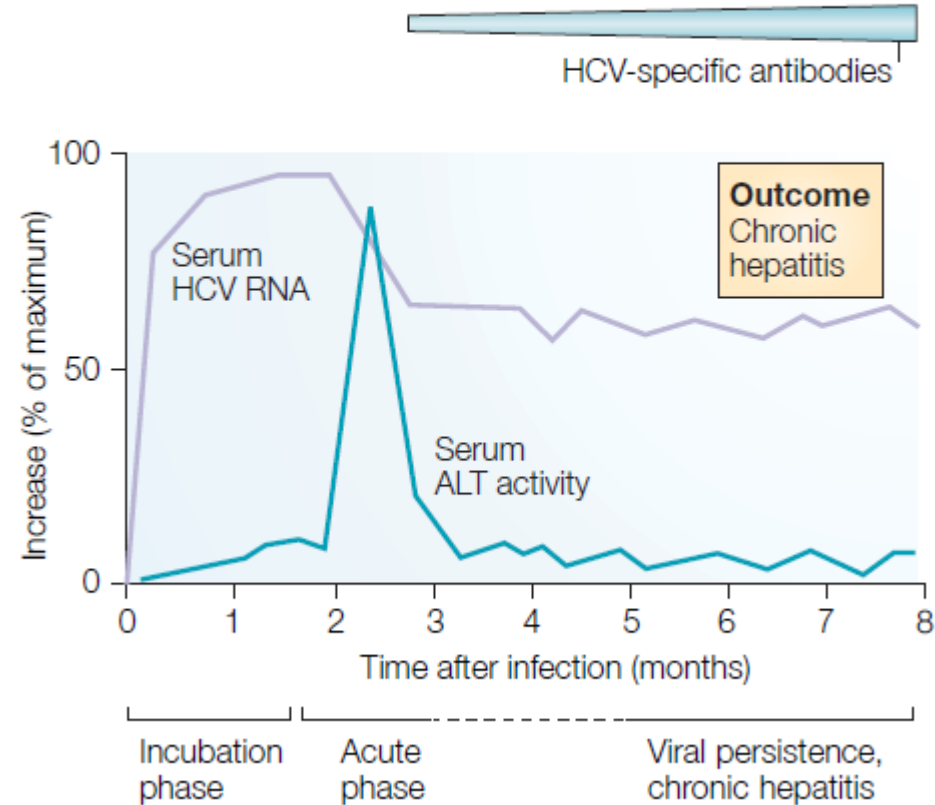
Hepatitis C

c Hepatitis C (acute)



Antígenos virais

d Hepatitis C (chronically evolving)



Hepatitis C

Acute HCV infection

Weak CTL response
Strong Th2 response

85%

15%

Strong CTL response
Strong Th1 response

Chronic HCV infection

Recovery

Mild and moderate
immune responses

Strong immune responses

Chronic hepatitis

Cirrhosis

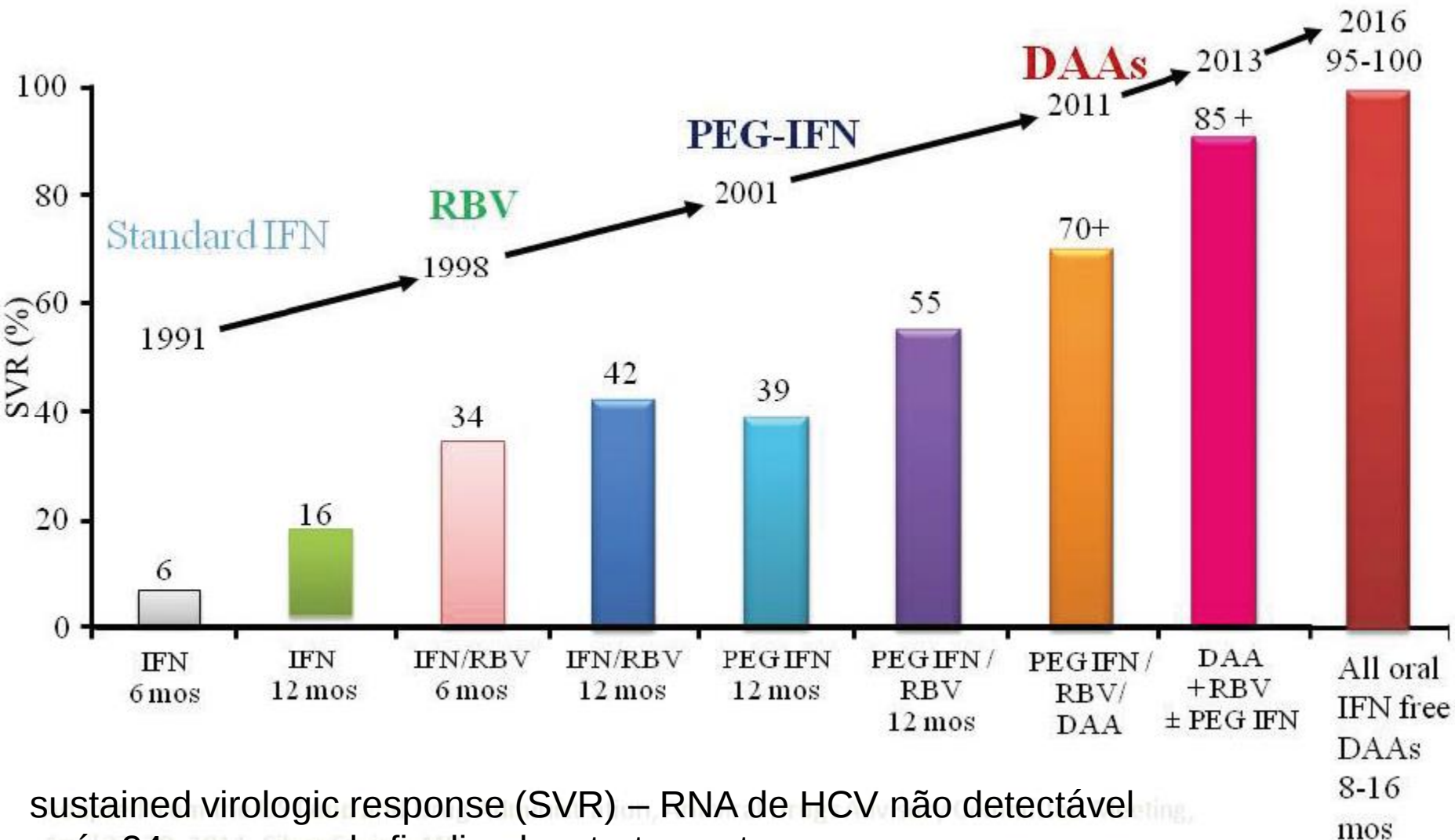
End-stage
liver disease

HCC

**É por isso que precisamos da
ajuda dos... Universitários**

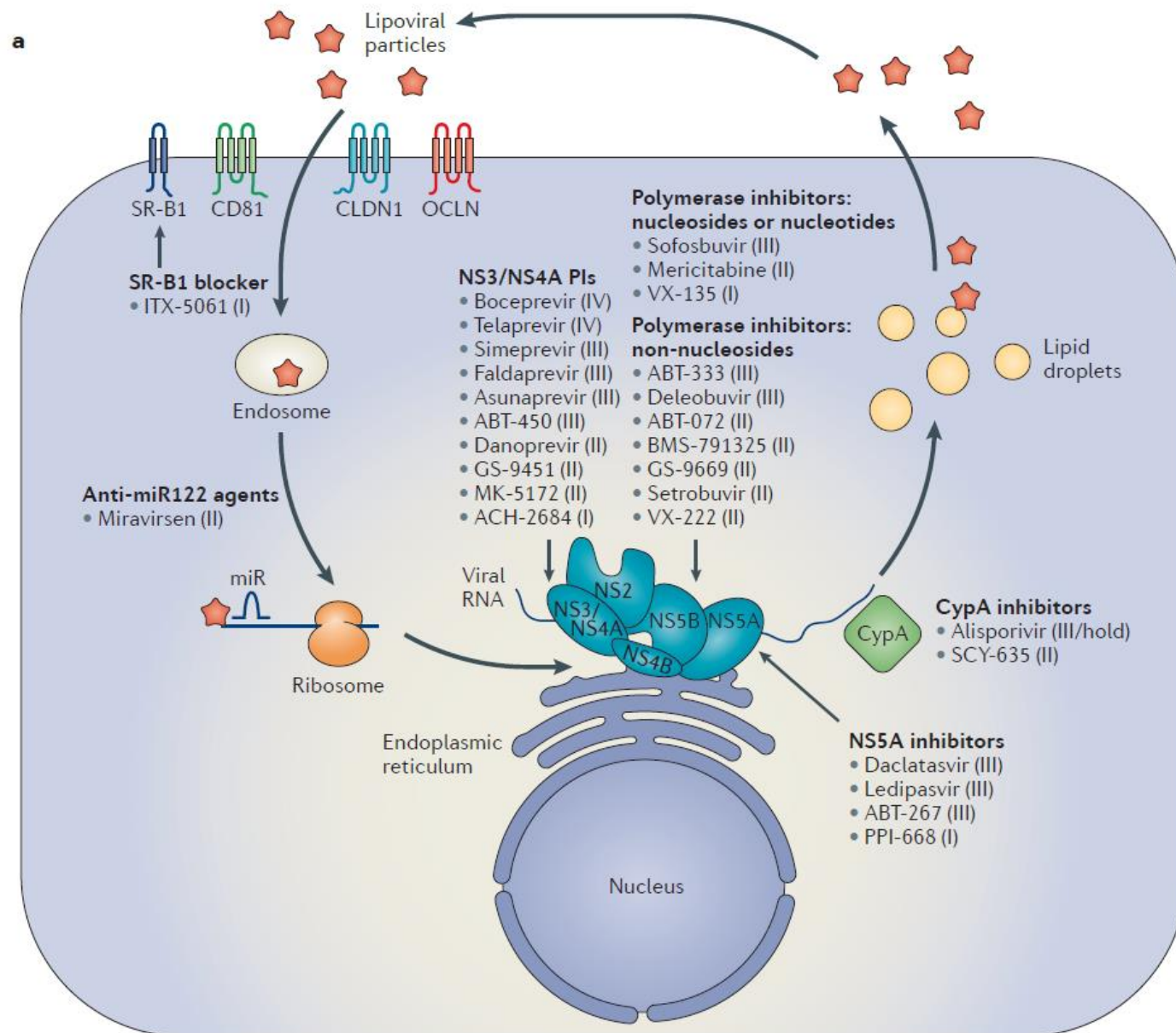
Hepatite C

Evolução das terapias contra HCV

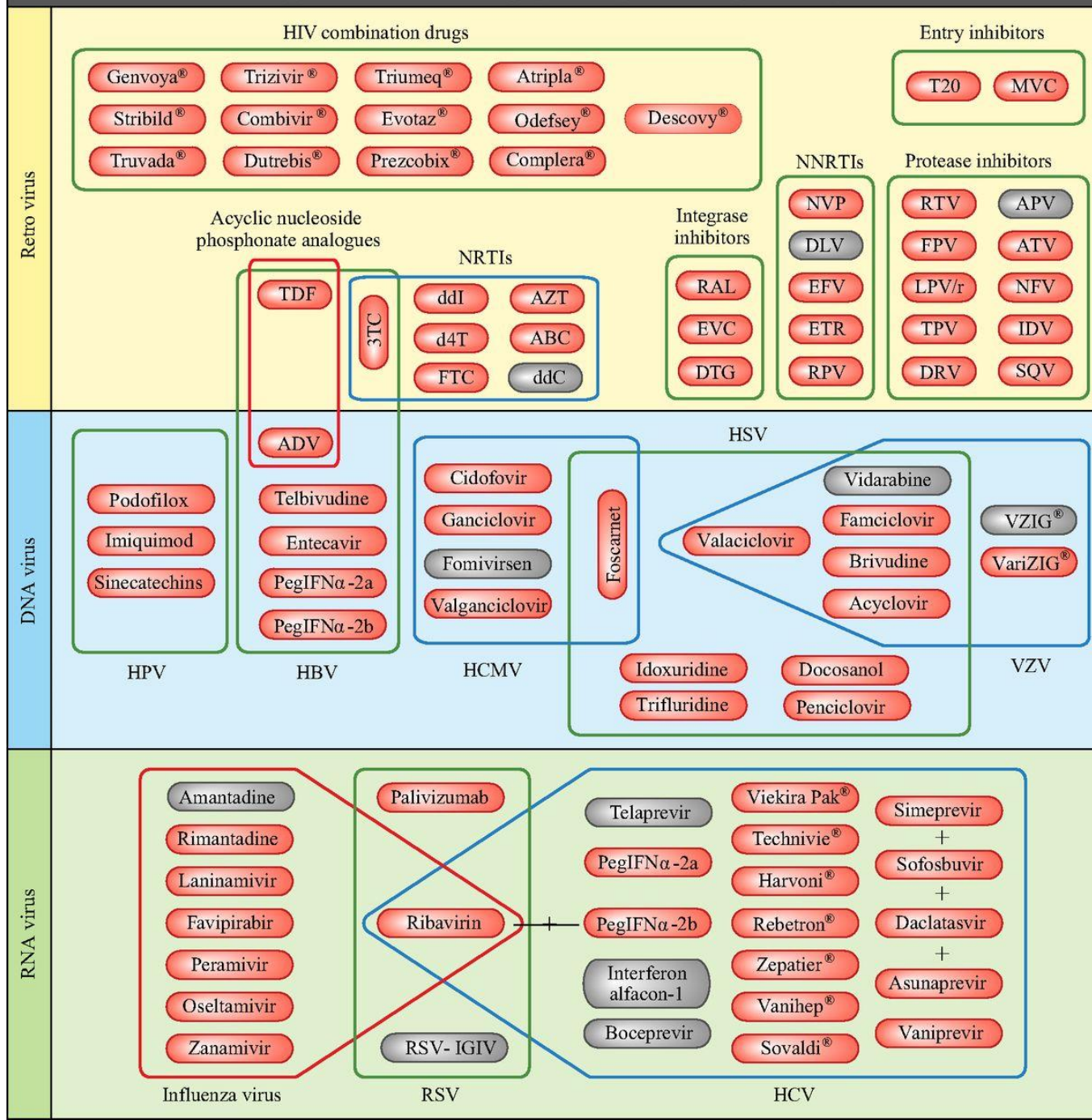


sustained virologic response (SVR) – RNA de HCV não detectável após 24 semanas de finalizado o tratamento.

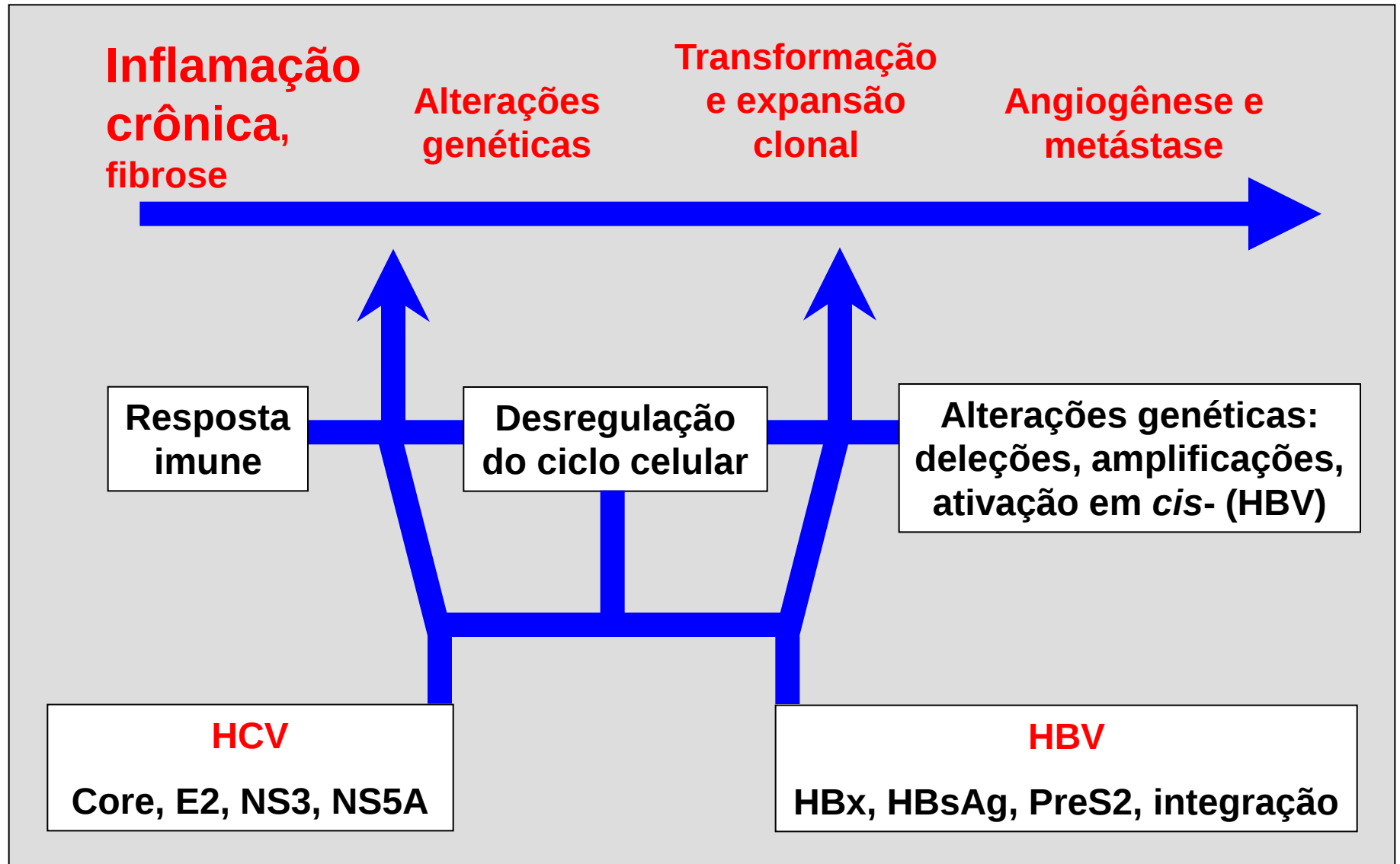
Hepatitis C Profilaxia



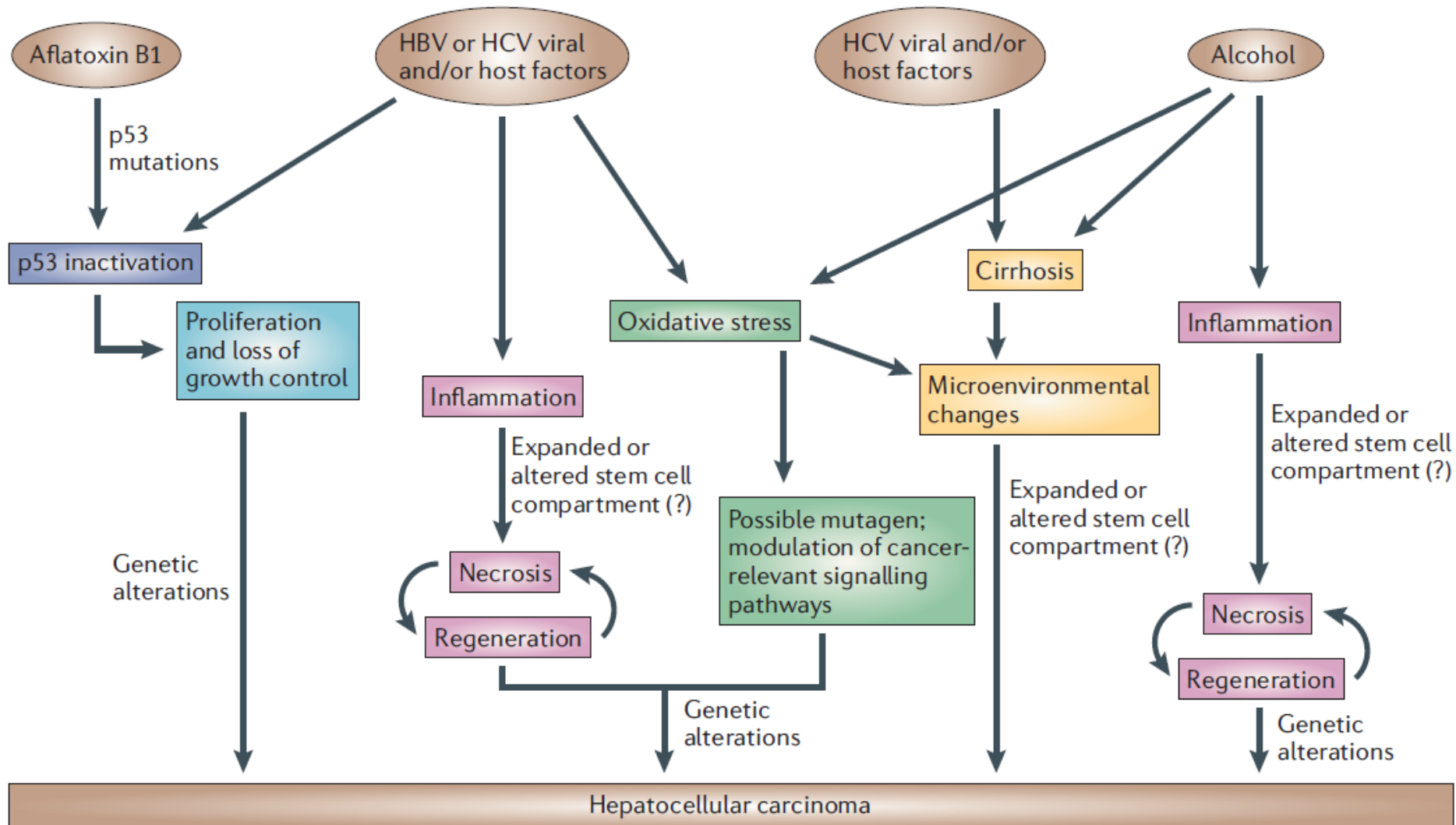
Approved antiviral drugs (January 1959 - April 2016)



Transformação celular por HBV e HCV



HCC: Co-fatores

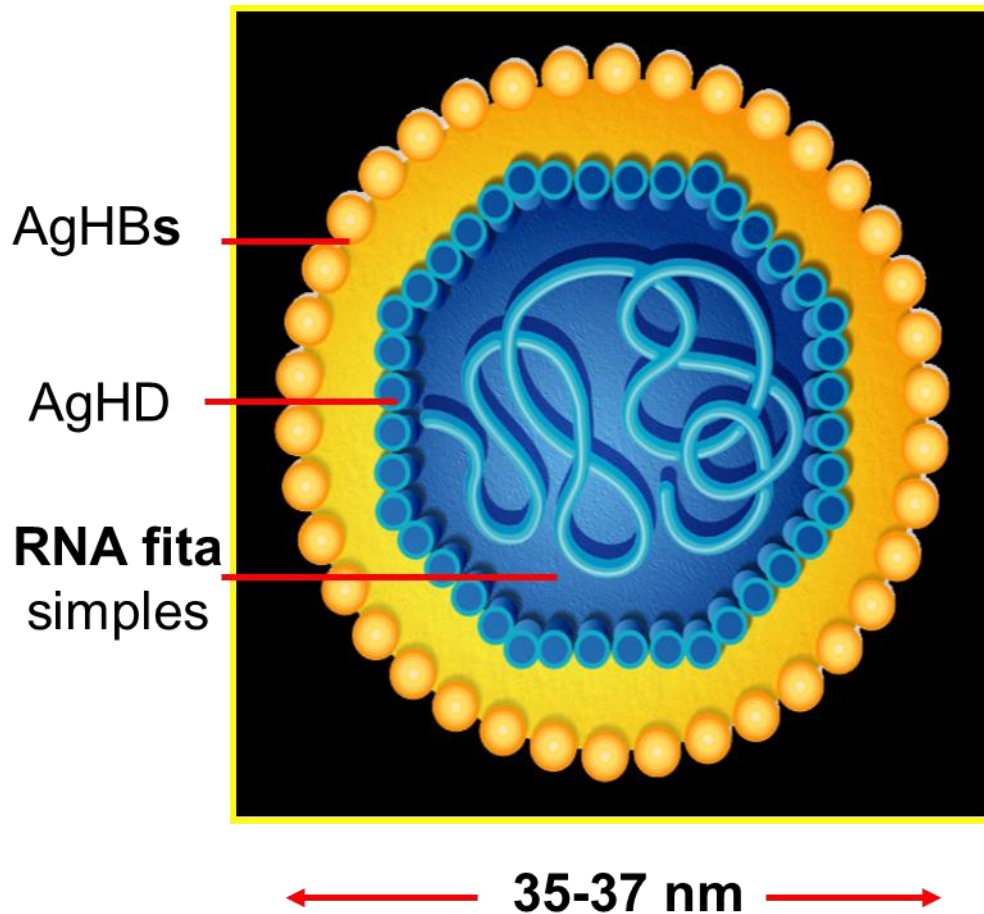


Hepatitis Delta (HDV)

Hepatite D

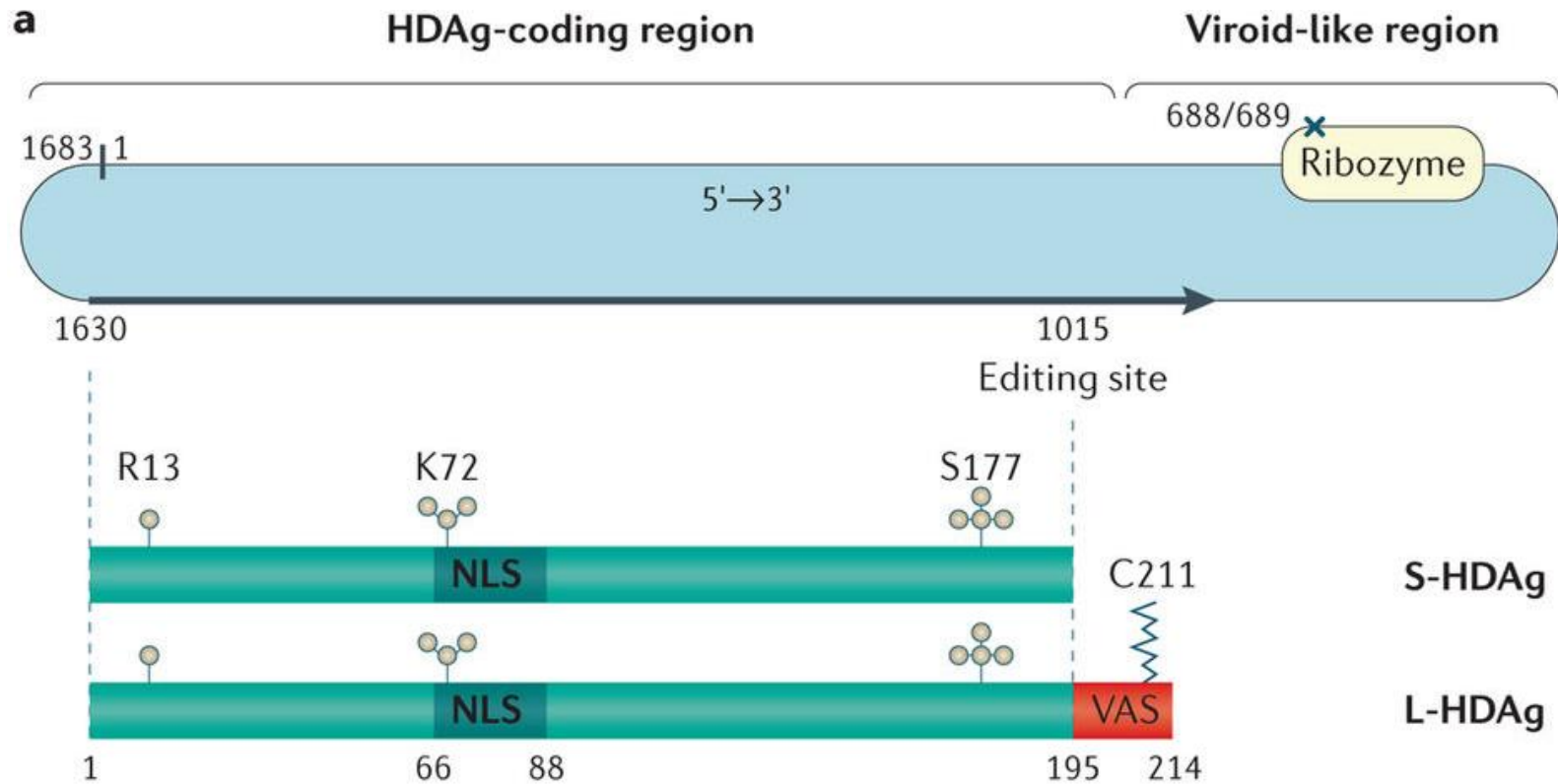
- Similar a viróides de plantas
- codifica proteínas (S-HDAg/replicação e L-HDAg/montagem)
- RNA com 1,7Kb, circular, negativo, dobrado sobre si mesmo
- Infecção concomitante com HBV, mas com multiplicação independente
- Citopático, 3 genótipos
- Antígeno + RNA envelopado por capsídeo codificada por HBsAg

Hepatitis D

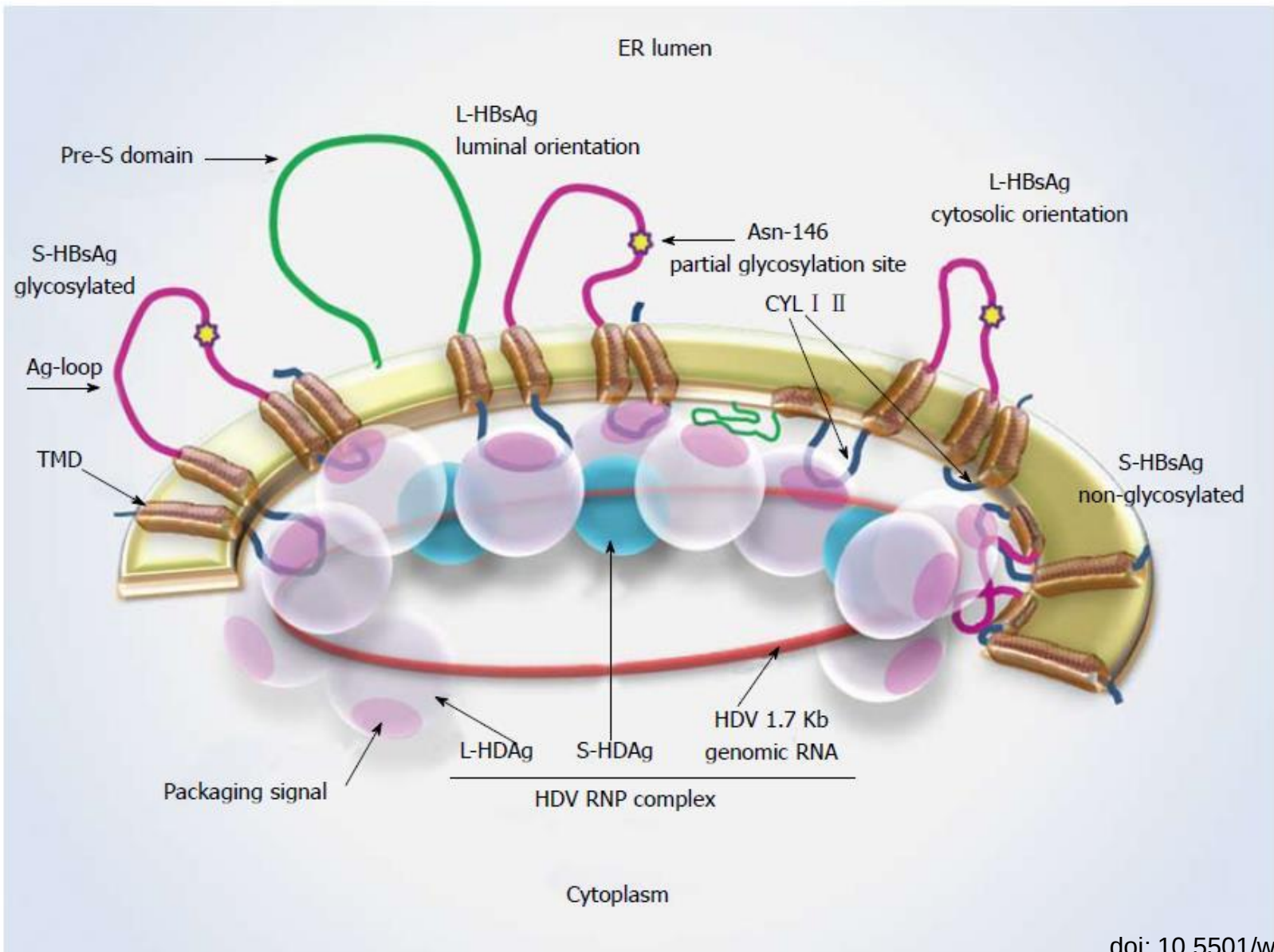


Hepatitis D

Organização genômica

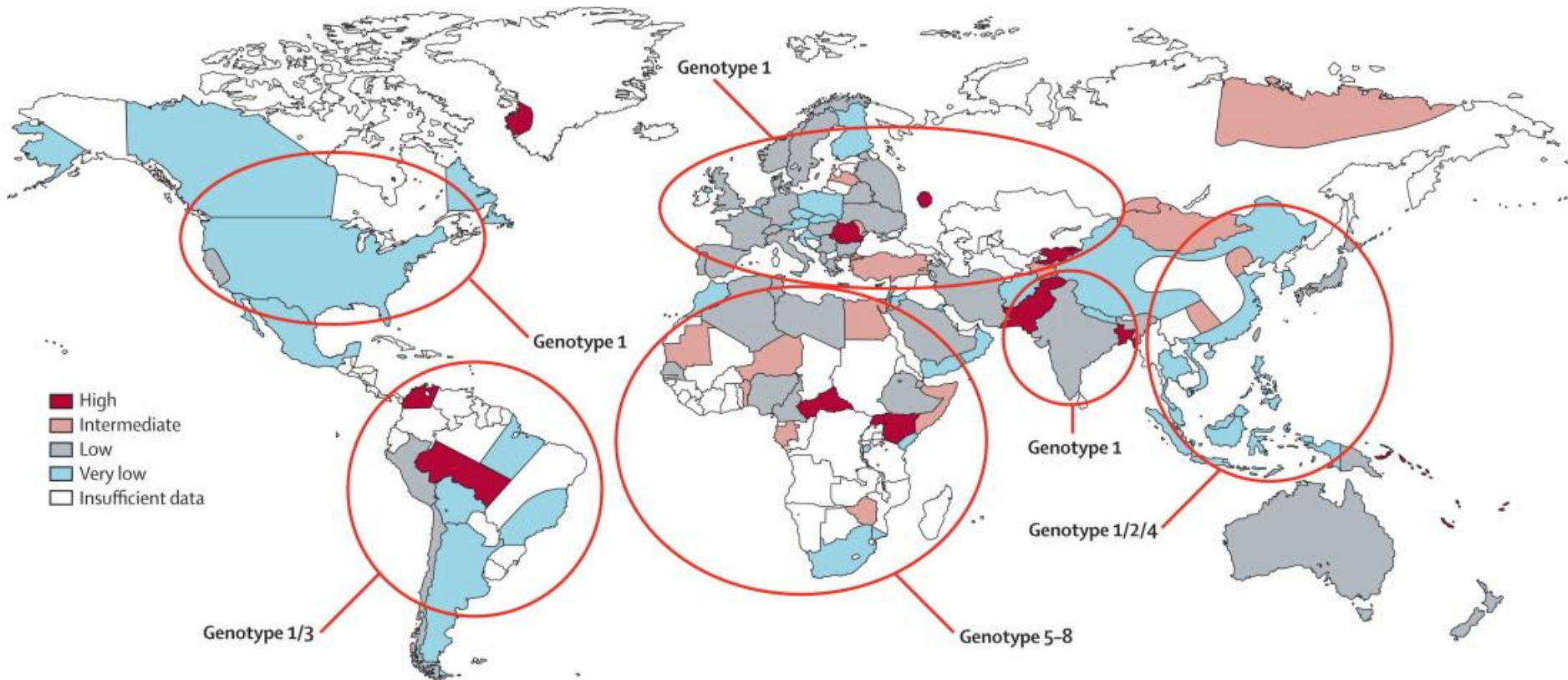


Hepatitis D



Hepatitis D

Oito genótipos



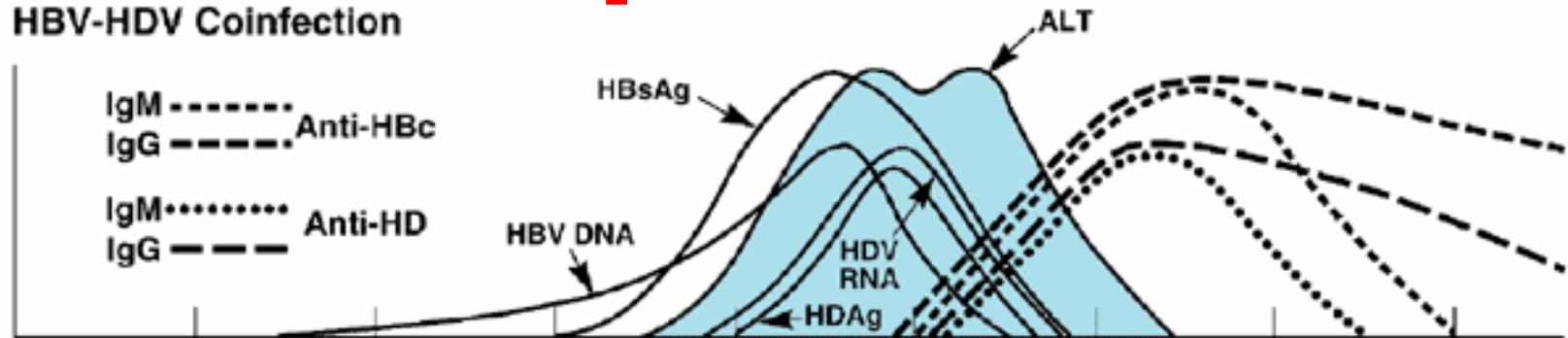
Hepatite D

Características clínicas

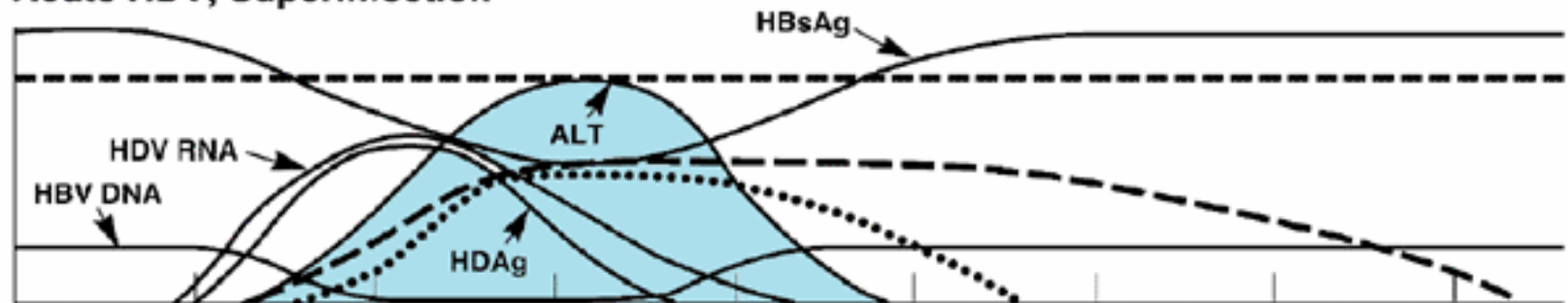
- Co-infecção
 - Doença aguda severa - hepatite fulminante
 - Baixo risco de infecção crônica
- Super-infecção
 - Geralmente desenvolve infecção crônica por HDV
 - Alto risco de doença crônica grave

Hepatitis D

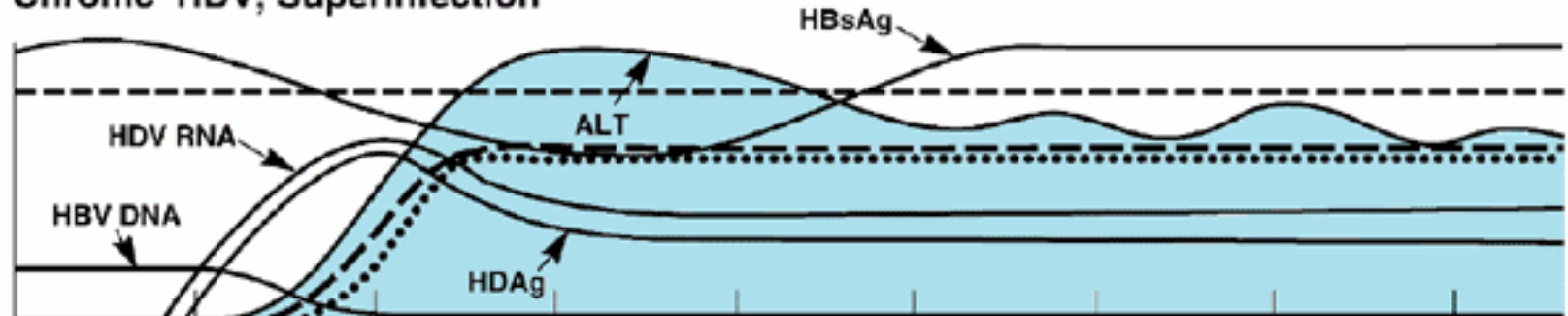
HBV-HDV Coinfection



Acute HDV, Superinfection



Chronic HDV, Superinfection



0

2

4

6

8

10

12

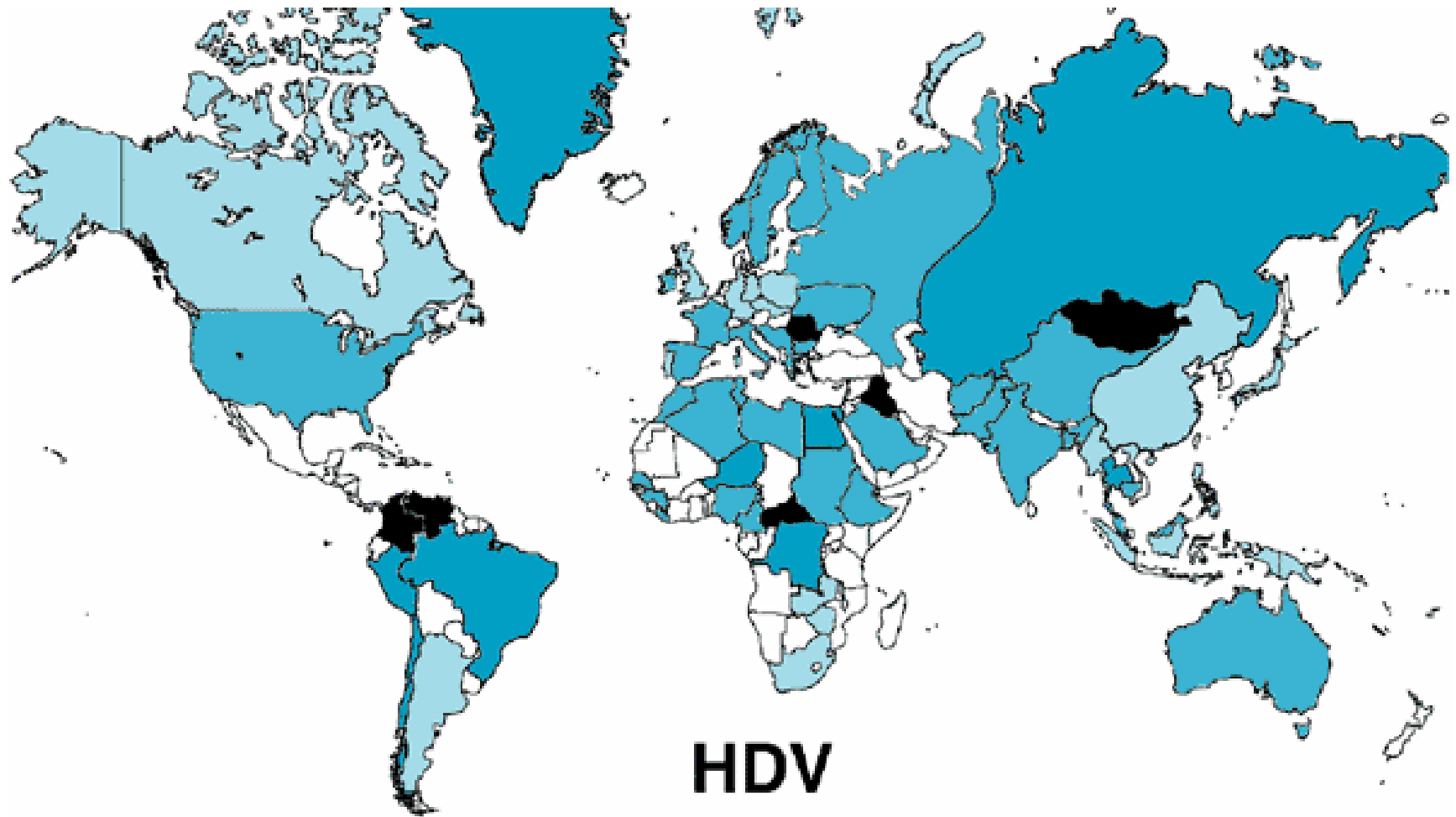
24

32

WEEKS AFTER EXPOSURE

Hepatite D

Prevalência de anti-HD em indivíduos positivos para HBsAg



HDV

Anti-HD(HBsAg (+)) ☐ ? ☐ 0-5% ☐ 6-20% ☐ 21-60% ☐ >60%

Hepatite D

Diagnóstico Laboratorial

- Marcador Sorológico:
 - VHB (Ag e Ac)
 - Anti-VHD IgM
 - Anti-VHD IgG

- Marcador Molecular: RNA
 - RT-PCR / “nested” PCR

Table 1 | A summary of diagnostic tests for the evaluation of patients with HDV infection

Test	Purpose	Remarks
Anti-HDV IgG antibody	Detects IgG antibodies against HDV, indicates previous or ongoing contact with HDV	Should be the first diagnostic screening test employed and should be performed in all HBsAg-positive patients
Anti-HDV IgM antibody	Detects IgM antibodies against HDV, indicates acute HDV infection or chronic HDV infection with disease activity	Can be used to determine disease activity in patients who test positive for anti-HDV IgG antibodies. Available tests are not standardized
HDV RNA qualitative	Detects HDV RNA Indicates HDV replication and active infection	Gold standard to determine HDV infection. Test can be false-negative if primers are not optimized for all HDV genotypes
HDV RNA quantitative	Determines the level of HDV RNA in the blood	Can be useful in the context of antiviral treatment to predict treatment response. There is no association between serum HDV RNA levels and the grade or stage of liver disease
HDV genotyping	Determines the HDV genotype	Different HDV genotypes may be associated with distinct clinical courses
HBsAg quantitative	Determines the level of HBsAg in the blood	HBsAg is associated with HDV RNA levels. HBsAg clearance is associated with HDV eradication and thus HBsAg monitoring can be useful during antiviral treatment
HBeAg/anti-HBe antibody	Determines the presence of the HBeAg and the presence of anti-HBe antibodies	About 15–20% of patients with HDV infection test positive for HBeAg, which can be associated with HBV replication. Treatment with HBV polymerase inhibitors might be indicated if IFN- α treatment is not possible
HBV DNA quantitative	Determines the level of HBV DNA in the blood	Indication for treatment with HBV polymerase inhibitors depends on the amount of HBV DNA detectable in the blood
Anti-HCV antibody/HCV RNA	Determines the presence of the antibodies against HCV and the presence of HCV RNA	Up to one third of patients in Europe with HDV infection are coinfectd with HCV. Screening for HCV should be performed at least once. HCV RNA is frequently suppressed by coinfection
Liver biopsy	Histological evaluation of and grading or staging of liver disease	Should be performed in all patients with hepatitis D as noninvasive markers of liver fibrosis are not proven to be able to accurately predict the stage of liver disease in patients with HDV infection

Abbreviations: HBsAg, hepatitis B surface antigen; HBe; hepatitis B early antigen; HBeAg, hepatitis B e antigen; HDV, hepatitis D virus.

OBRIGADO!!!