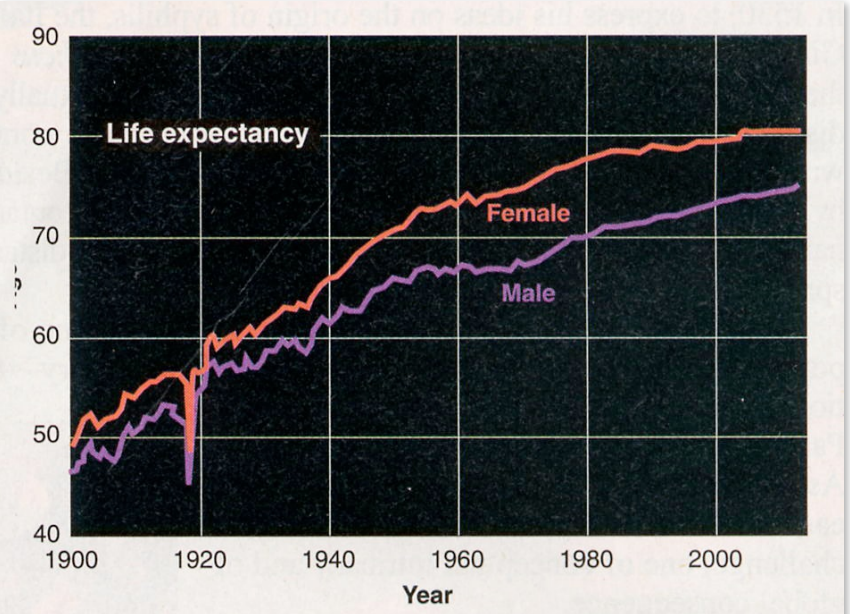
Vaccines

Lecture 19
Biology 3310/4310
Virology
Spring 2017

Nothing shocks me. I'm a scientist.
INDIANA JONES

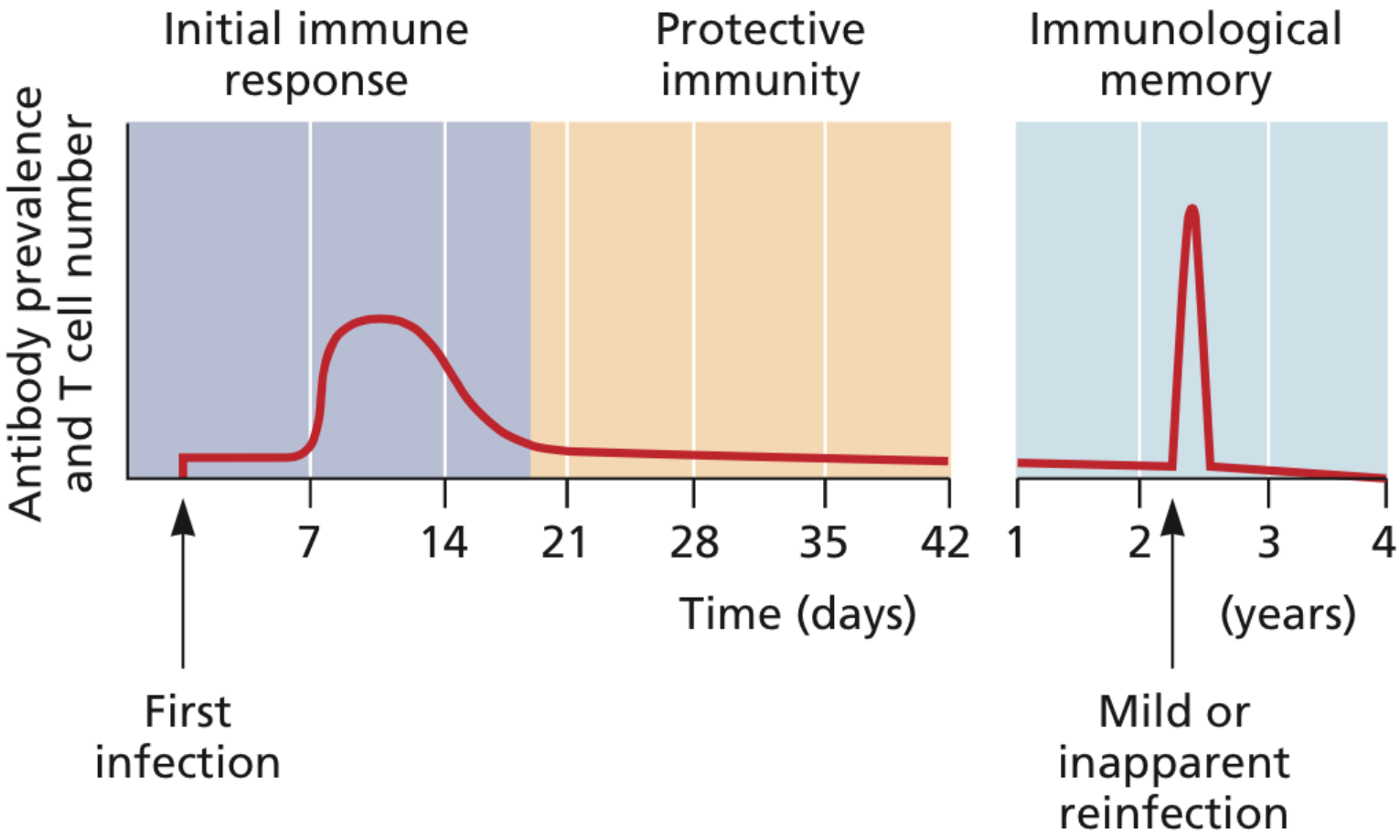
Vaccines are our proven best defense against viruses

- Vaccination mobilizes the host immune system to prevent virus infections
  - *Immune memory*
- Vaccination breaks the chain of transmission

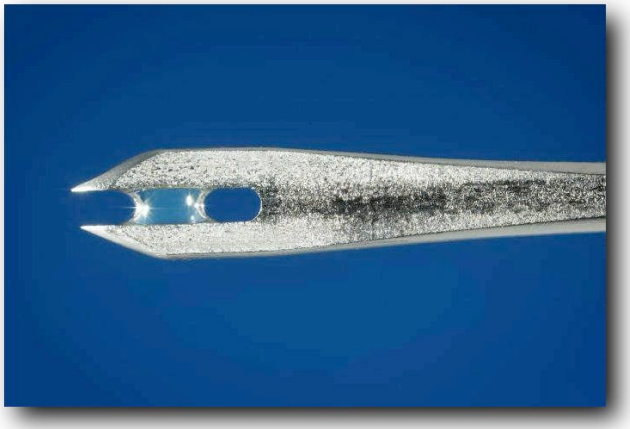


Longer life. Anti-infection medicine and other factors have helped dramatically lengthen the average life expectancy in the U.S.

Vaccines stimulate a protective immune response



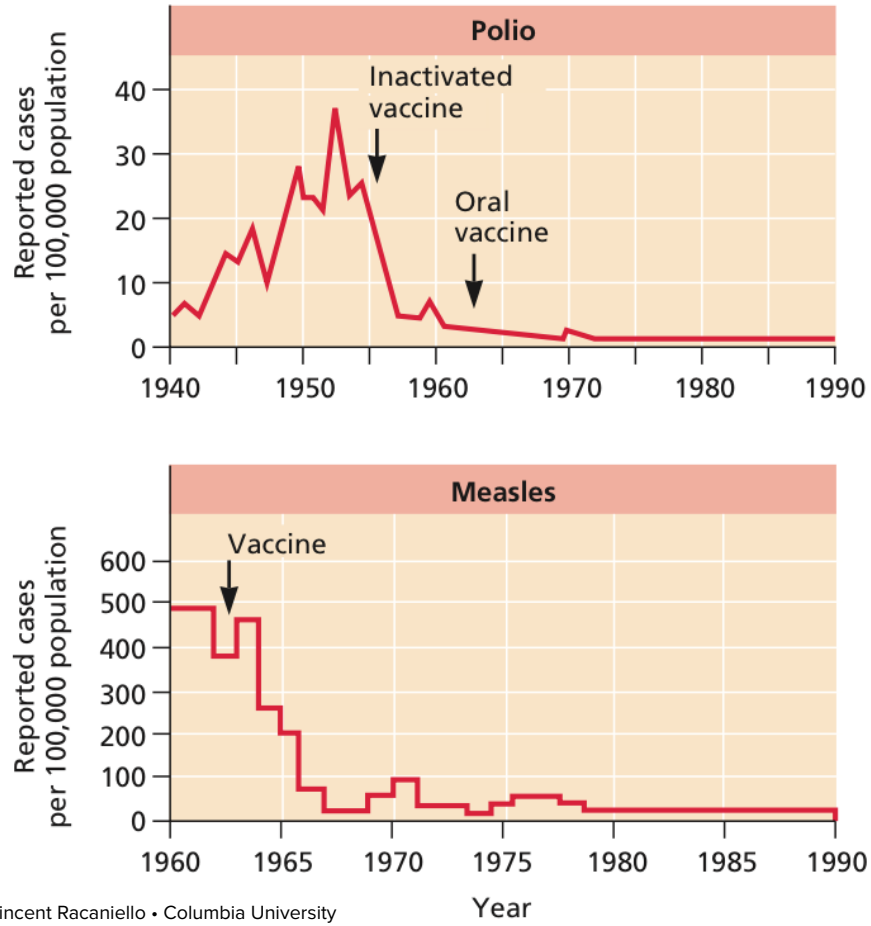
Principles of Virology, ASM Press

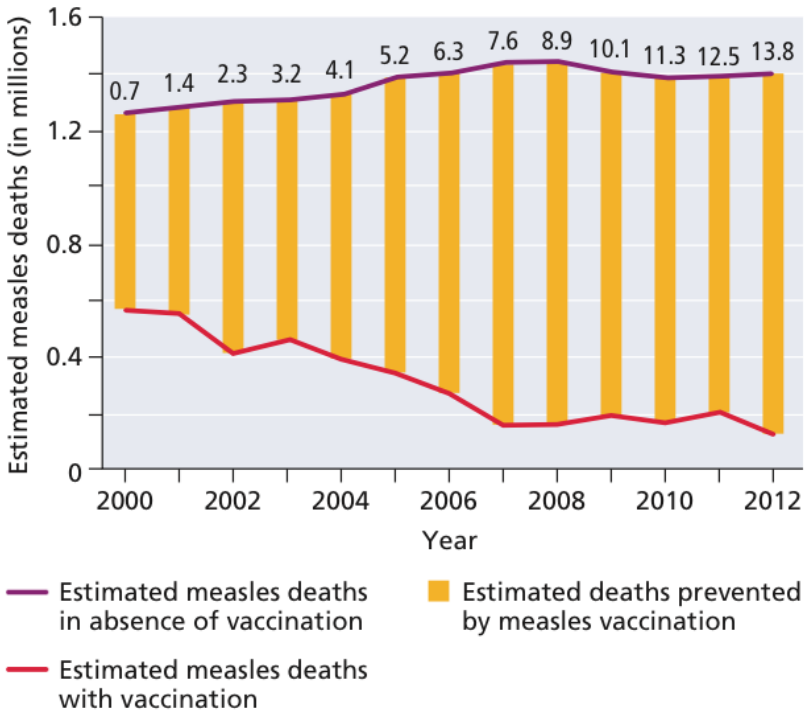


- Jenner, 1796
- Pasteur, 1885 - rabies vaccine; introduced the term vaccination from *vacca* (Latin, cow) in honor of Jenner
- Yellow fever, influenza vaccines - 1930s


OPENING MIXTURE
DRINK POCK
Cow
The Cow-Pock — or — the Wonderful Effects of the New Inoculation! — Vide the Publications of ye Anti-Vaccine Society.
9904

Large-scale vaccination campaigns can be successful





Vaccines are now an integral part of our existence

- We immunize children, adults of all ages, domesticated and wild animals
- Because of immunization, many childhood diseases are rare
- Vaccines are a major part of the First World's public health measures, **but not the Third World** (e.g. rubella, measles)

How vaccines work in the real world

- Maintenance of a critical level of immunity
- Herd immunity

Herd Immunity

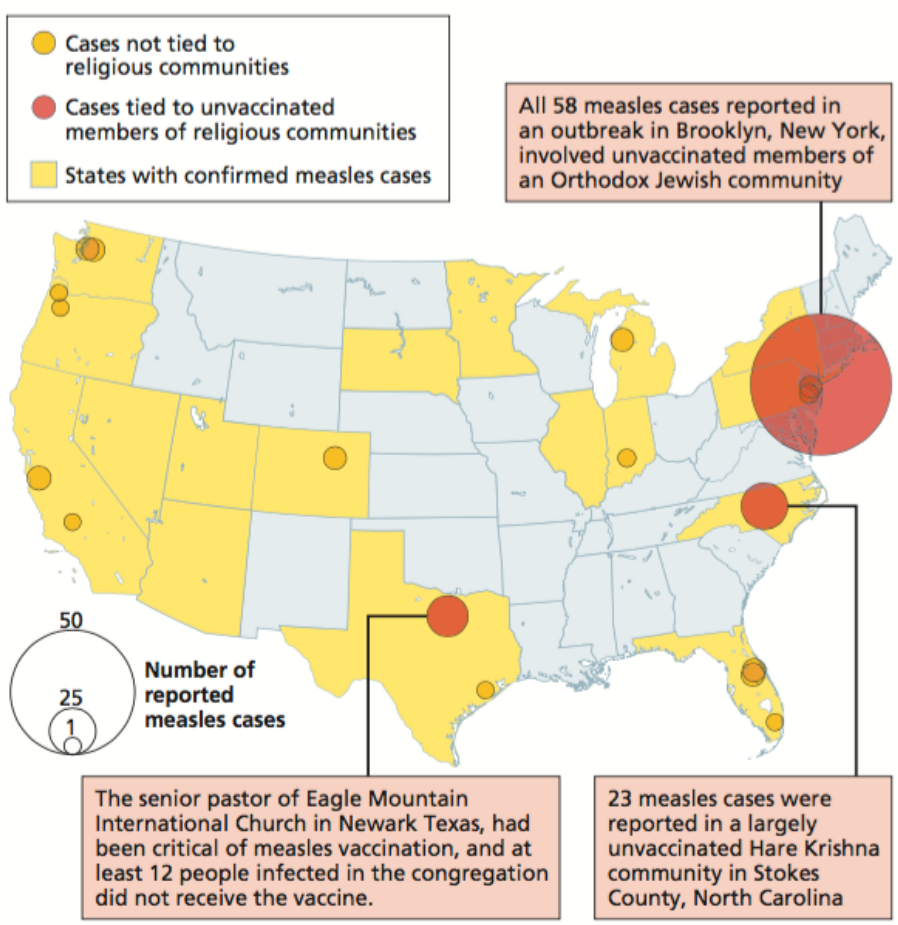
- Virus spread stops when the probability of infection drops below a critical threshold
- The threshold is virus and population specific
- Smallpox: 80 - 85%
- Measles: 93 - 95%
- No vaccine is 100% effective
- When 80% of population is immunized with measles, 76% of population is immune

Public complacency is dangerous to any vaccine program

- “Viral diseases are a thing of the past”
- “Polio is long gone”
- “I never get the flu”
- “Measles is just a trivial kid’s disease”
- “Chicken pox only affects kids”
- “Kids should get infected naturally”
- “I’m not injecting anything into *my* body”
- “Vaccines make you sick, they cause autism, they cause multiple sclerosis, etc etc”
- “I know a guy who got the flu shot and then got the flu”
- “I can’t afford to immunize my kids”
- “I don’t have time this year”

When these attitudes prevail, society has serious problems with large-scale vaccination programs

Vaccine programs depend on public acceptance of their value



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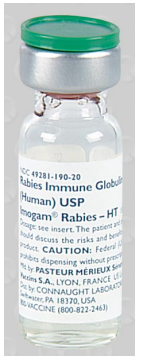
room number: virus

Herd immunity:

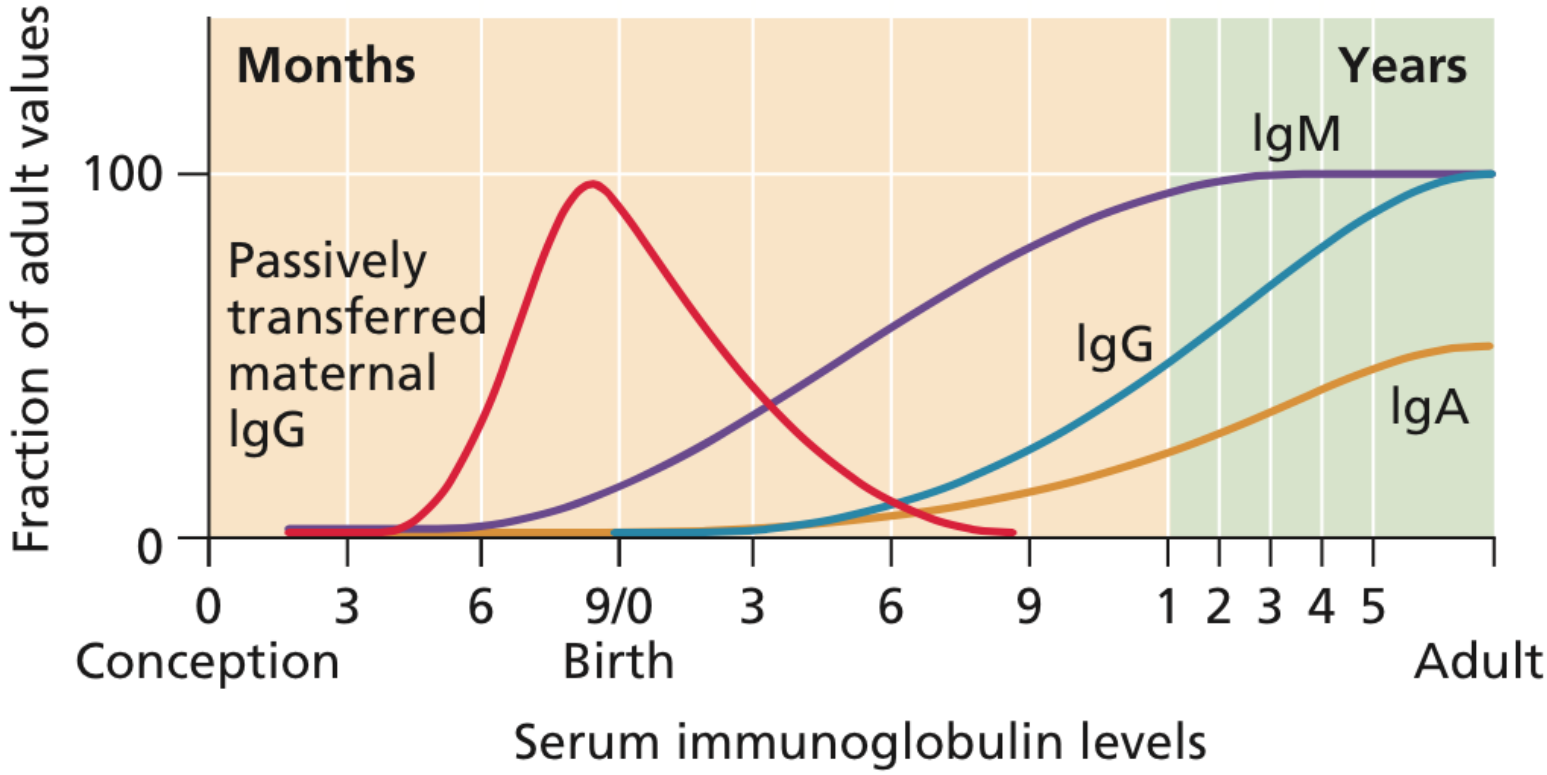
A. Demonstrates the importance of immunizing livestock
B. Emphasizes that not everyone must be immune to protect a population
C. Emphasizes that everyone must be immune to protect a population
D. Describes how group-think can dominate anti-vaccine choices
E. All of the above

Vaccines can be *active* or *passive*

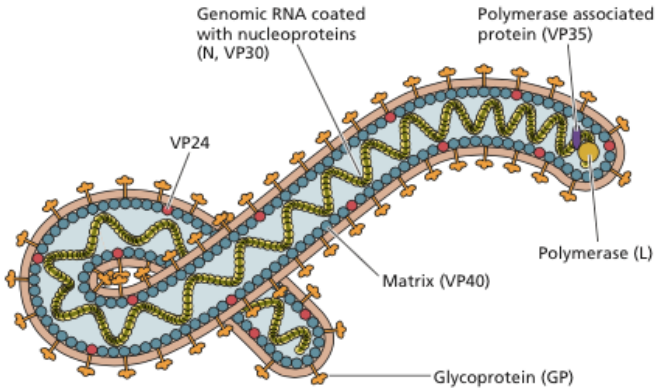
- Active - instilling into the recipient a modified form of the pathogen or material derived from it that induces immunity to disease
  - *Long term protection*
- Passive - instilling the products of the immune response (antibodies or immune cells) into the recipient
  - *Short term protection*

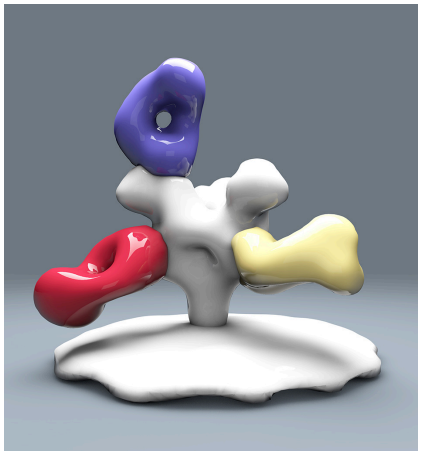


A natural passive vaccine



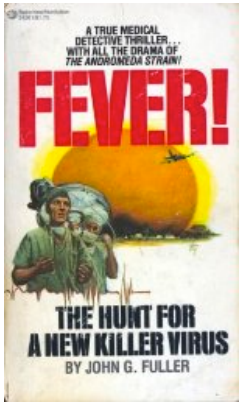
Zmapp, the best known passive vaccine





- Raised in mice immunized with virus-like particles
- Chimerized into human IgG1 scaffold
- Produced in tobacco plants

Passive therapy with convalescent serum



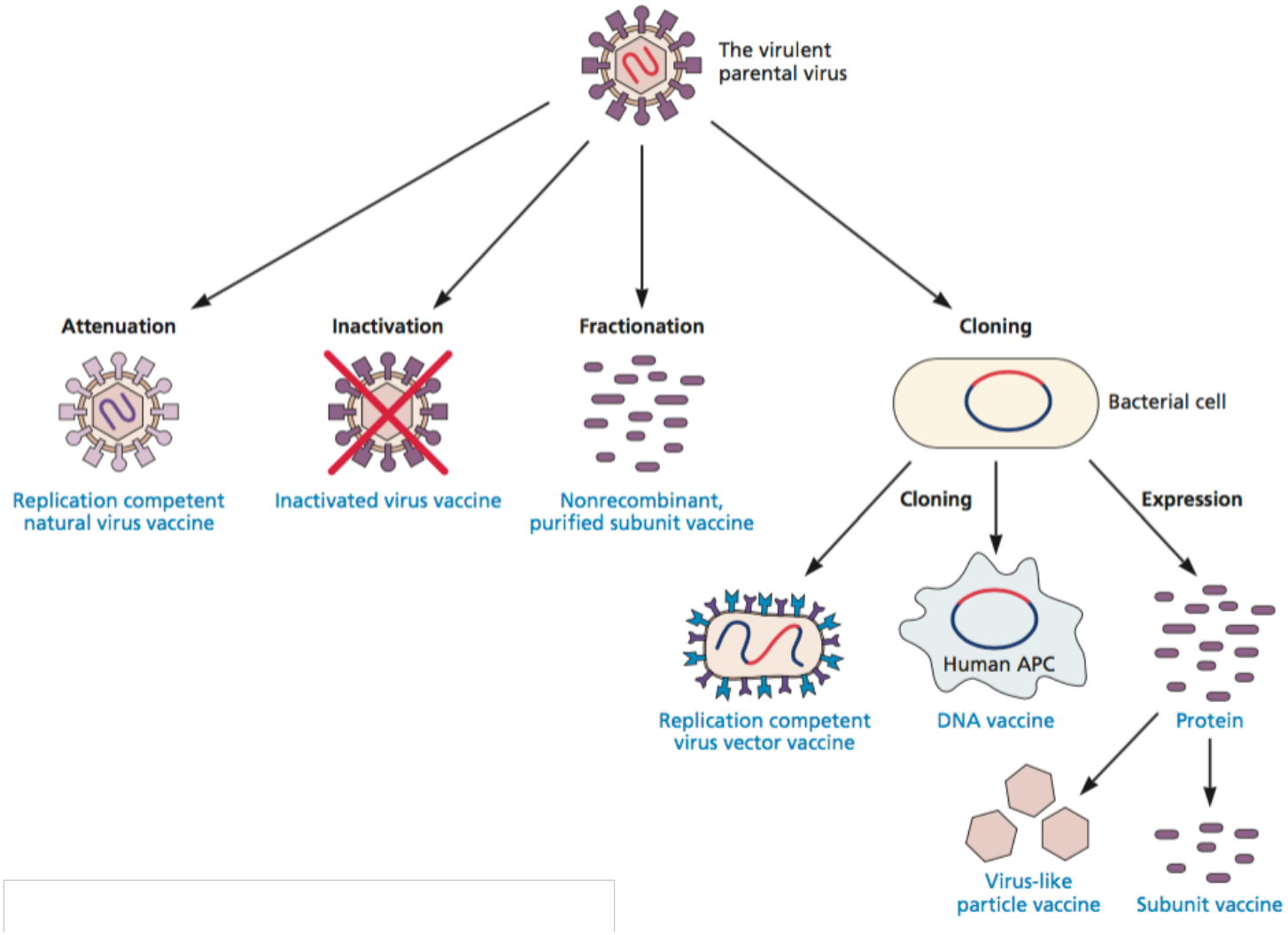
- Jordi Casals infected himself with Lassa virus at Yale in 1969
- Transfused with blood from nurse (Penny Pinneo) who had survived Lassa fever

Requirements of an effective vaccine

- Induction of an *appropriate immune response*
  - *Th1 vs Th2 response*
- Vaccinated individual must be *protected against disease* caused by a virulent form of the specific pathogen
  - *Just getting 'a response' is not enough (e.g. producing antibodies)*

Requirements of an effective vaccine

- Safety: no disease, minimal side effects
- Induce protective immunity in the population
- Protection must be long-lasting
- Low cost (<$1, WHO); genetic stability; storage considerations; delivery (oral vs. needle)

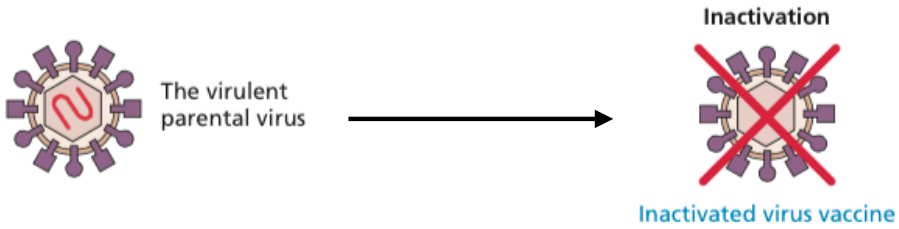

The virulent parental virus
Attenuation
Inactivation
Fractionation
Cloning
Bacterial cell
Cloning
Expression
Replication competent natural virus vaccine
Inactivated virus vaccine
Nonrecombinant, purified subunit vaccine
Human APC
Replication competent virus vector vaccine
DNA vaccine
Protein
Virus-like particle vaccine
Subunit vaccine

Viral vaccines licensed in the US

Disease or virus	Type of vaccine	Indications for use	Schedule
Adenovirus	Attenuated, oral	Military recruits	One dose
Hepatitis A	Inactivated whole virus	Travelers, other high-risk groups	0, 1, and 6 mo
Hepatitis B	Yeast-produced recombinant surface protein	Universal in children, exposure to blood, sexual promiscuity	0, 1, 6, and 12 mo
Influenza	Inactivated viral subunits	Elderly and other high-risk groups	One dose seasonally
	Recombinant proteins	Elderly; those with egg allergies	One dose seasonally
Influenza	Attenuated	Children 2–8 yr old, not previously vaccinated with influenza vaccine	Two doses at least 1 mo apart
		Children 2–8 yr old, previously vaccinated with influenza vaccine	One dose
		Children, adolescents, and adults 9–49 yr old (e.g., FluMist, FluBlo)	One dose
Japanese encephalitis	Inactivated whole virus	Travelers to or inhabitants of high-risk areas in Asia	0, 7, and 30 days
Measles	Attenuated	Universal vaccination of infants	12 mo of age; 2nd dose, 6 to 12 yr of age
Mumps	Attenuated	Universal vaccination of infants	Same as measles, given as MMR
Papilloma (human)	Yeast- or SF9-produced virus-like particles	Females 9–26 yr old Males 11-21 yr old	Three doses
Rotavirus	Reassortant	Healthy infants	2, 3, and 6 mo or 2 and 4 mo of age depending on vaccine
Rubella	Attenuated	Universal vaccination of infants	Same as measles, given as MMR
Polio (inactivated)	Inactivated whole viruses of types 1, 2, and 3	Changing: commonly used for immunosuppressed where live vaccine cannot be used	2, 4, and 12–18 mo of age, then 4 to 6 yr of age
Polio (attenuated)	Attenuated, oral mixture of types 1, 2, and 3	Universal vaccination; no longer used in United States	2, 4, and 6–18 mo of age
Rabies	Inactivated whole virus	Exposure to rabies, actual or prospective	0, 3, 7, 14, and 28 days postexposure
Smallpox	Vaccinia virus	Certain laboratory workers	One dose
Varicella	Attenuated	Universal vaccination of infants	12 to 18 mo of age
Varicella-zoster	Attenuated	Adults 60 yr old and older	One dose
Yellow fever	Attenuated	Travel to areas where infection is common	One dose every 10 yr

Principles of Virology, ASM Press

Inactivated vaccines



- Chemical procedures (e.g. formalin, β-propriolactone, nonionic detergents)
- Infectivity is eliminated, antigenicity not compromised

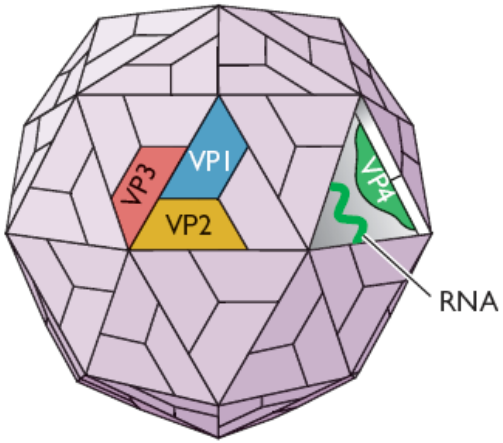
Poliomyelitis

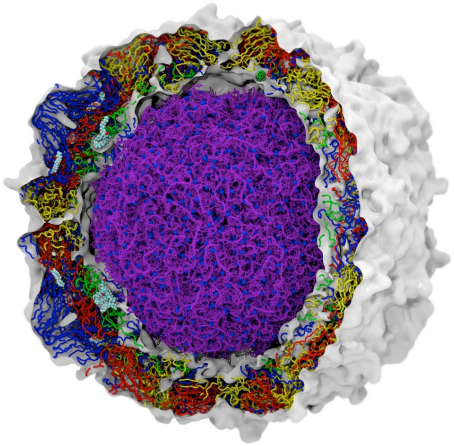
- Polio (grey), myelon (marrow) = Greek
- itis (inflammation of) = Latin

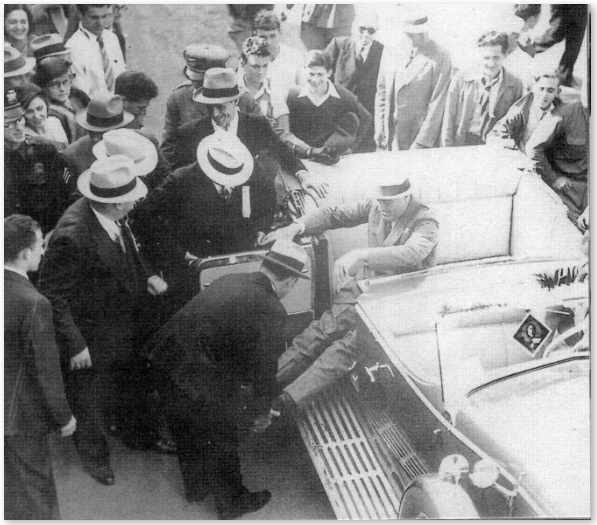
- "A common, acute viral disease characterized clinically by a brief febrile illness with sore throat, headache and vomiting, and often with stiffness of the neck and back. In many cases a lower neuron paralysis develops in the early days of illness"

  —*J.R. Paul, "Poliomyelitis (Infantile Paralysis)", in A Textbook of Medicine, 1959.*

Poliomyelitis



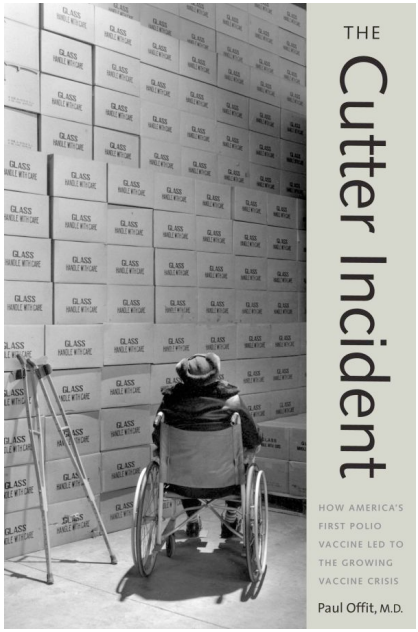


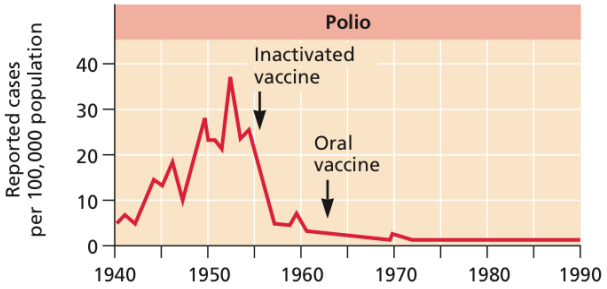


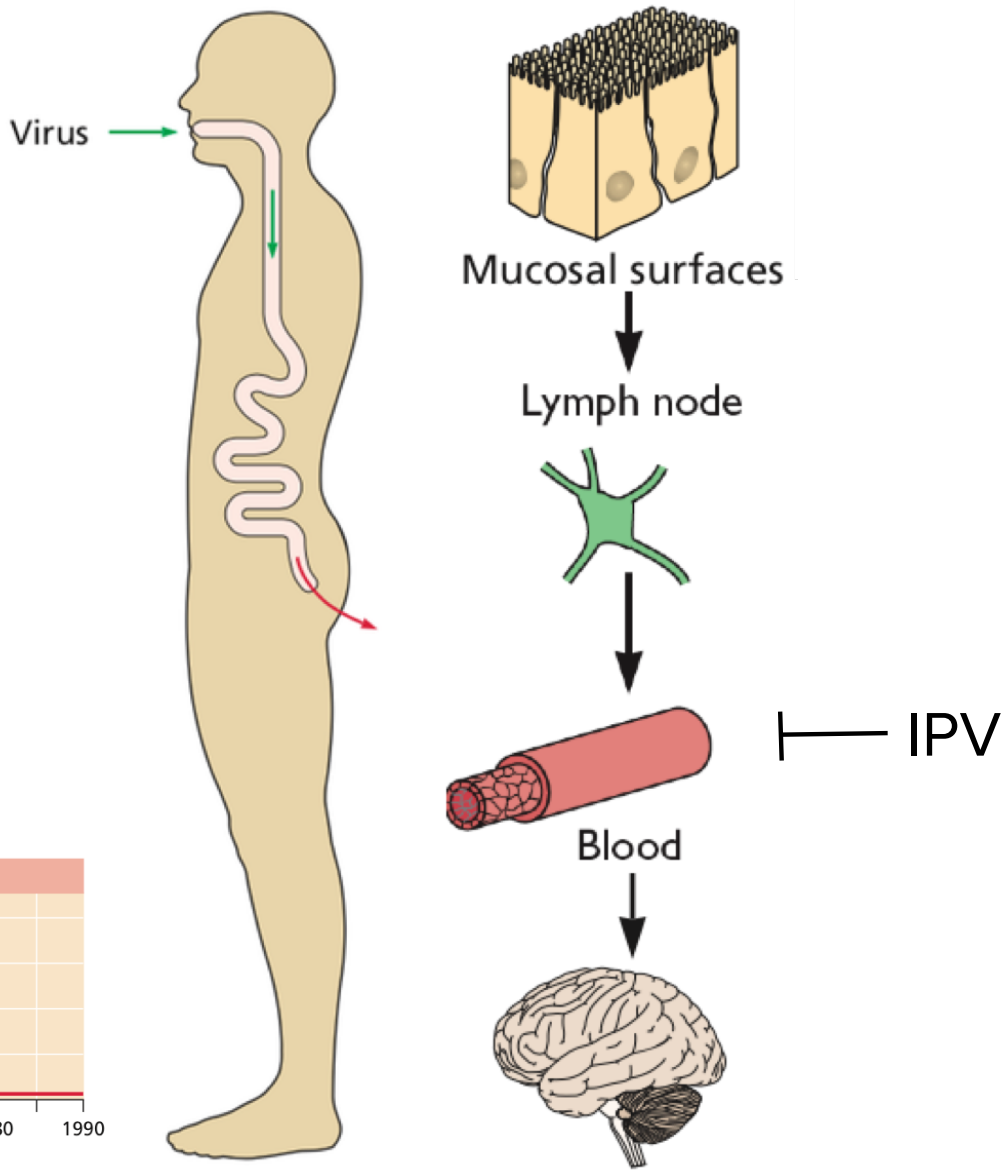
Inactivated poliovirus vaccine, IPV

- Poliovirus treated with formalin to destroy infectivity
- 1954: National Foundation for Infantile Paralysis-sponsored clinical trial of Jonas Salk's IPV, 1,800,000 children
- >50% protection, results announced 12 April 1955, licensed same day

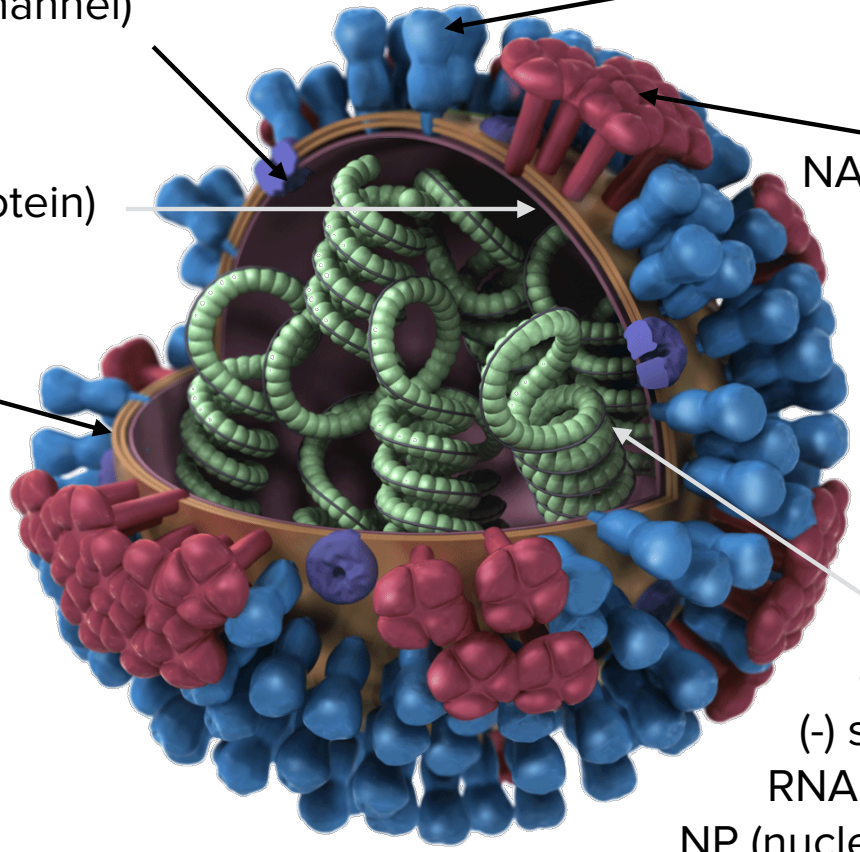

All Local Teams Are Rained Out
Journal American
7TH SPORTS WALL ST. SPECIAL
SALK'S VACCINE WORKS!
Official Count
Polio Vaccine Reported 80 to 90% Effective
All Local Teams Are Rained Out
Salk Sure Its 100% OK
Booster Shots Urged
Only 3 Escape
Salk's Formula:
The New York Times.
SALK POLIO VACCINE PROVES SUCCESS; MILLIONS WILL BE IMMUNIZED SOON; CITY SCHOOLS BEGIN SHOTS APRIL 25
Supply to Be Low for Time, But Output Will Be Rushed
6 VACCINE MAKERS GET U. S. LICENSES
Honor Gets Degree From Clark at The Citadel
NEW YORK Herald Tribune Late City Edition
on Way Out, Salk Report Shows; City Enabled to Treat 50% More
Two Shots Now, Third 7 Mo. Later
Shots Held 20 to 90% Effective
Blackmail Is Charged To South
New York World-Telegram The Sun
Dr. Francis' Official Report:
POLIO VACCINE IS 'SAFE, EFFECTIVE AND POTENT'
Rain Washes Out Three Local Clubs
Only 2 Shots Needed Now, Salk Reveals
Major Success Against Killing Paralytic Type
New York Post NIGHT EXTRA
fficial Salk Test Report:
POLIO ROUTED!
Daily Mirror
Triumph Over Polio!
They Vanished!
55 VACCINE WILL BE EVEN BETTER
Dr. Salk Visions 100% Effectiveness
Polio Victims Pray: May We Be Last


THE
Cutter Incident
HOW AMERICA'S FIRST POLIO VACCINE LED TO THE GROWING VACCINE CRISIS
Paul Offit, M.D.


Polio
Inactivated vaccine
Oral vaccine
Reported cases per 100,000 population
40
30
20
10
0
1940
1950
1960
1970
1980
1990


Virus
Mucosal surfaces
Lymph node
IPV
Blood

Influenza virus

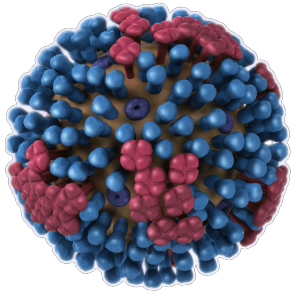


Three types: A, B, C

Inactivated influenza vaccine

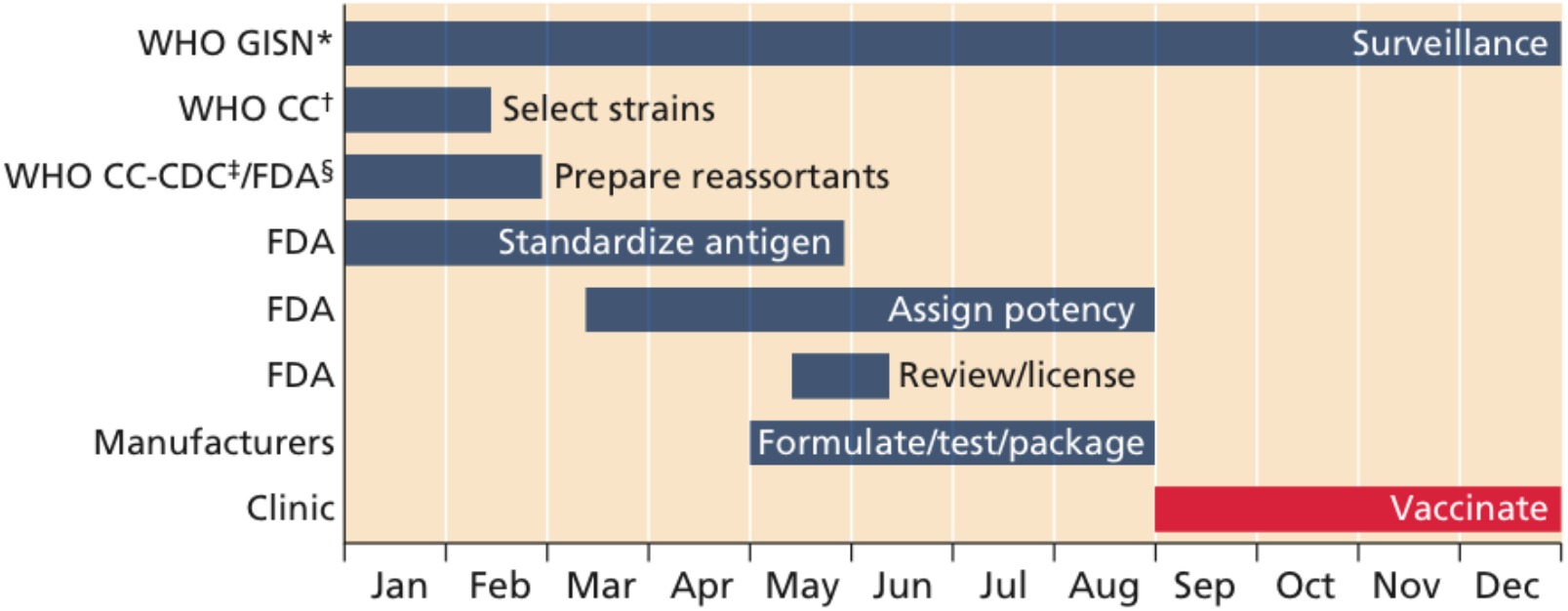
- 3000-49000 deaths/yr in US due to influenza virus
- Vaccine: virus grown in embryonated chicken eggs, formalin-inactivated or detergent or chemically disrupted virions
- 75-100 million doses manufactured each year US
- 60% effective in healthy children and adults <65 yr
- Protection correlates with serum antibodies to HA, NA
- Vaccine produced in cell culture avoids egg allergies (Flucelvax)

Inactivated influenza vaccine



- Envelope proteins change each year; new strains must be selected in the first few months for manufacture
- Use reassortants with most RNA segments from high-yielding strain, HA, NA from selected strain
- 2017-18 vaccine: A/Michigan/45/2015 (H1N1)pdm09-like virus; A/Hong Kong/4801/2014 (H3N2)-like virus; B/Brisbane/60/2008-like virus

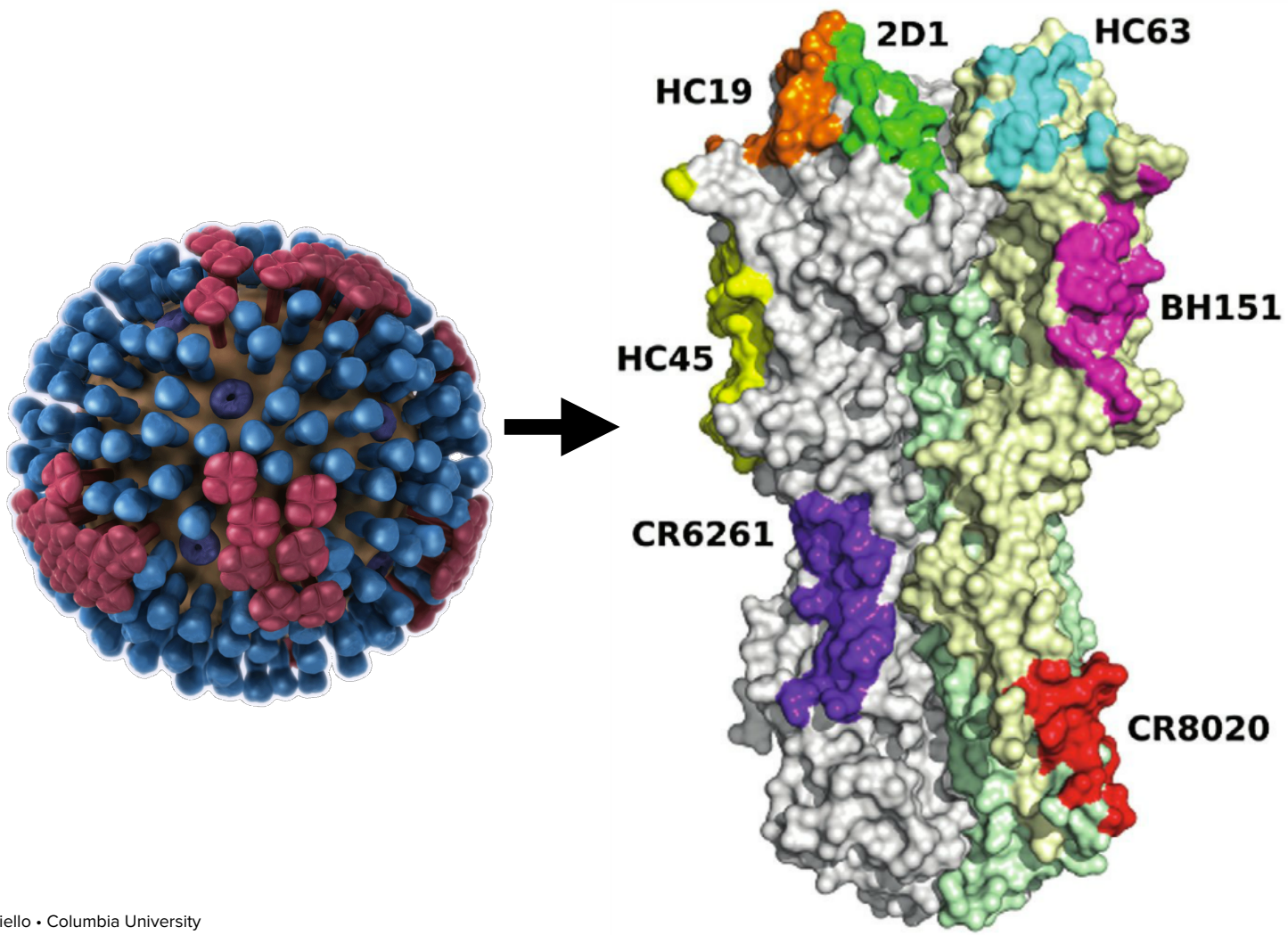
Selecting an influenza virus vaccine



*World Health Organization Global Influenza Surveillance Network
†WHO Collaborating Centres
‡US Centers for Disease Control and Prevention
§US Food and Drug Administration

http://www.microbe.tv/twiv/twiv-413/ on how strains are selected

Antigenic drift: Influenza virus



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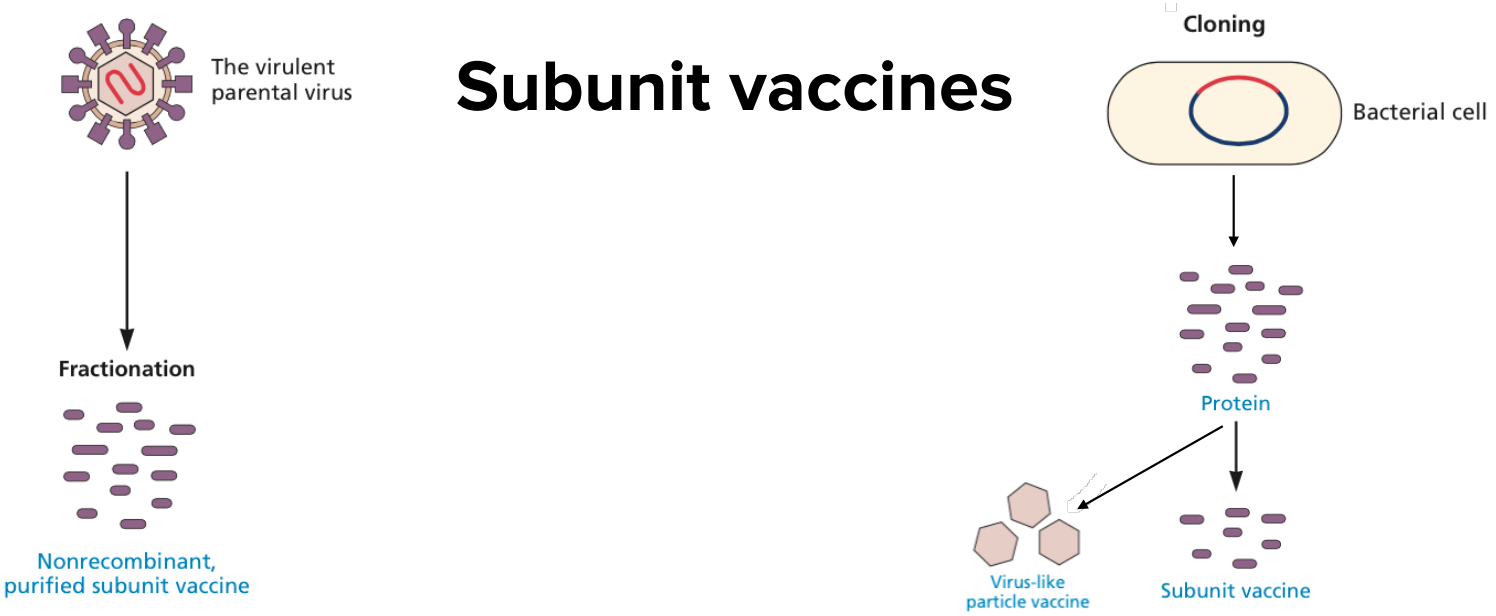
room number: virus

Which statement about inactivated viral vaccines is incorrect:

A. Chemicals can be used to inactivate infectivity
B. They do not replicate
C. They can be dangerous if inactivation is not complete
D. Antigenic variation can make them ineffective
E. None of the above are incorrect

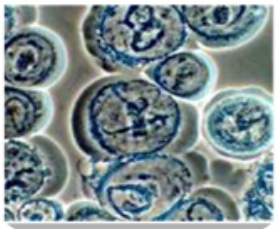
Subunit vaccines



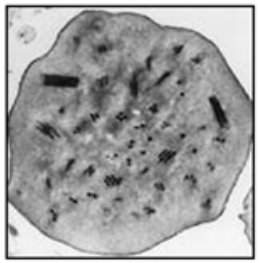
- Break virus into components, immunize with purified components
- Clone viral gene, express in bacteria, yeast, insect cells, cell culture, purify protein
- Antigen usually a capsid or membrane protein

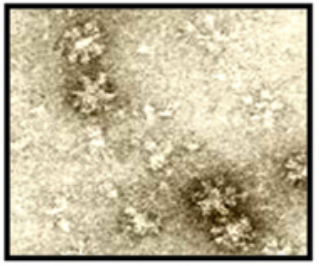
Flublok

Baculovirus Expression Vector System (BEVS) Technology



Vaccine prepared within 2 months





Baculovirus

- Engineer to express recombinant protein under powerful promoter

***expres*SF+® cells**

- Infect cells in fermenter
- Low-cost serum-free media

Pure protein

- Purify protein
- Formulate with PBS into vaccine

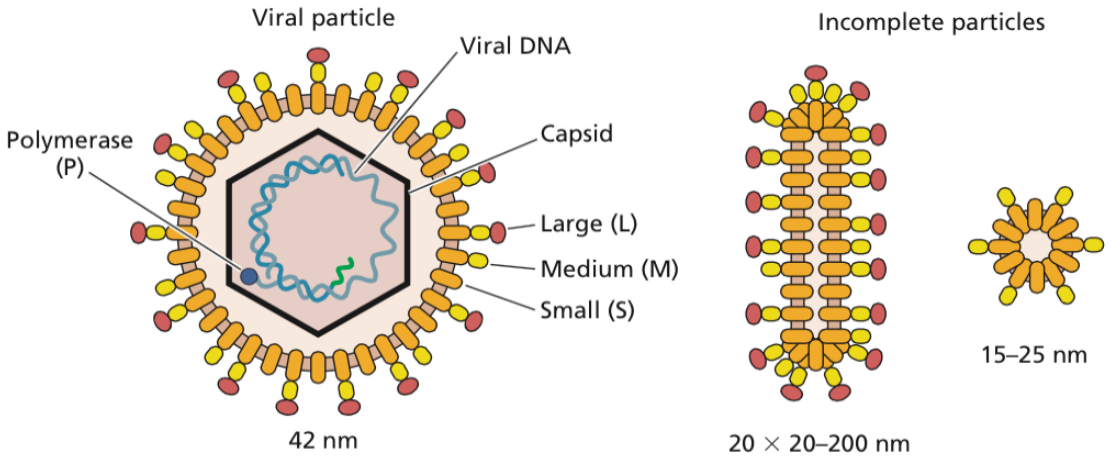
Approved for 18-49 years old

Protein Sciences Corp.

Some successful subunit vaccines

Cancer vaccine

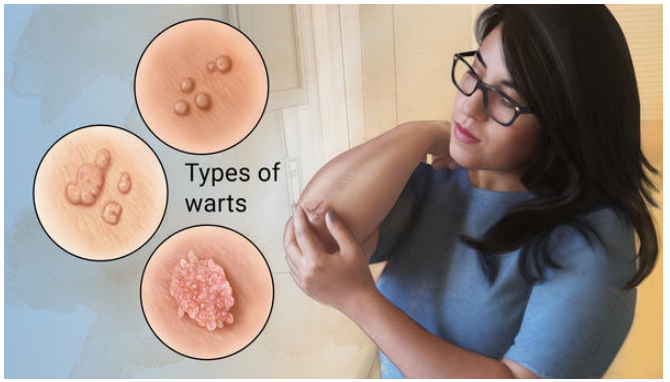
- Hepatitis B virus (HBV) - HBsAg protein produced in yeast
- Assembles into empty particles

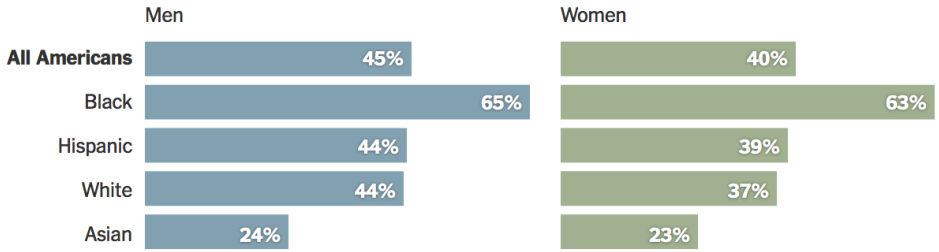


Principles of Virology, ASM Press

Human papillomaviruses

- Agents of warts (>170 types)
- Some are transmitted sexually, most common STD in USA
- Some cause low risk genital warts
- Others are high risk for cancers: cervix, vagina, penis, anus, oropharynx (31,000/yr; mostly 16, 18)
- Nearly half of Americans infected with genital HPV (18-59)



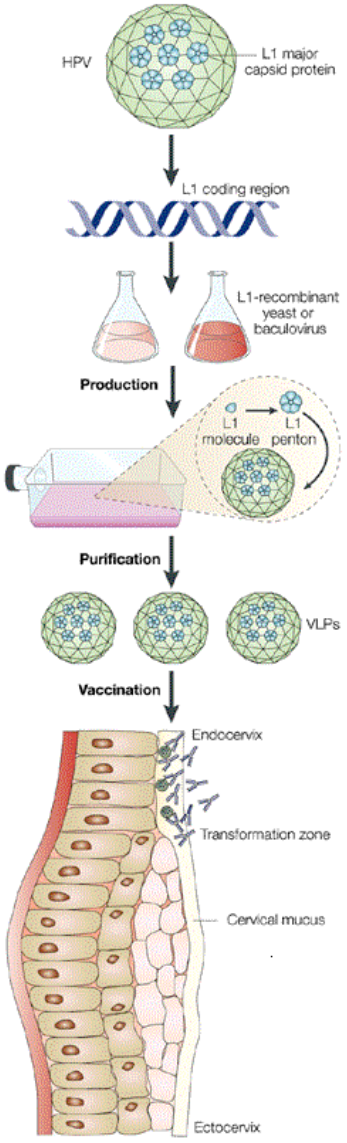


https://nyti.ms/2oFBTM2

Human papillomavirus vaccines

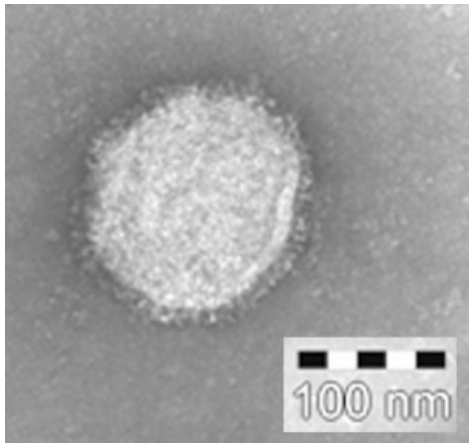
Cancer vaccines

- *Gardasil* (Merck): types 6, 11, 16, 18 produced in *S. cerevisiae*
- *Gardasil-9* (Merck): types 6, 11, 16, 18, 31, 33, 45, 52, 58
- *Cervarix* (GlaxoSmithKline): types 16, 18 produced in insect cells
- Should be given before becoming sexually active



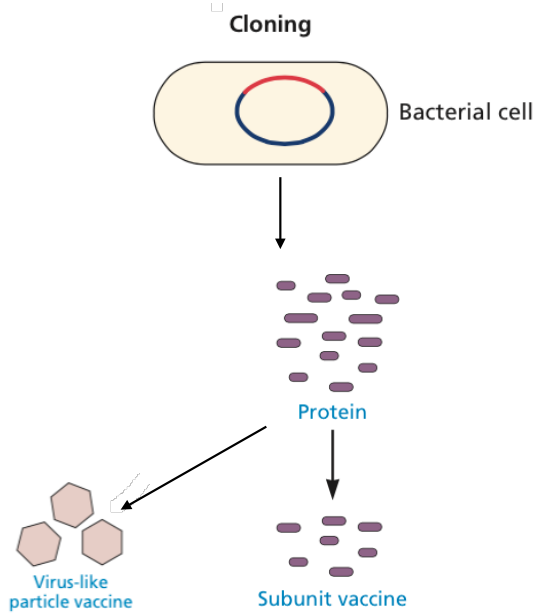
Future influenza vaccines?

- Virus-like particles: synthesis of HA alone in cells leads to production of immunogenic particles
- Has also been done in plants
- 1 square meter of plants produces 20,000 doses at under $0.20/dose



Subunit vaccine pro and con

- Advantages of a modern subunit vaccine
  - *Recombinant DNA technology*
  - *No viral genomes or infectious virus*
- Disadvantages
  - *Expensive*
  - *Injected*
  - *Poor antigenicity*



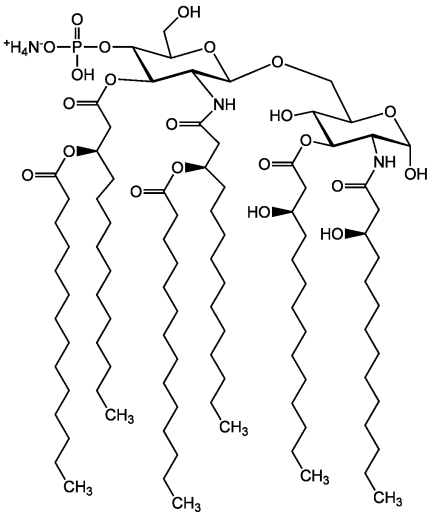
Principles of Virology, ASM Press

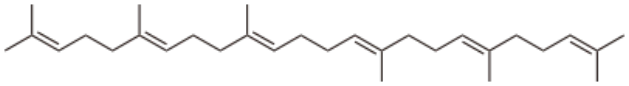
Inactivated and subunit vaccines have a common problem

- Viral proteins don’t replicate or infect
- Don’t cause inflammation
- Pure proteins often require *adjuvant* to mimic inflammatory effects of infection

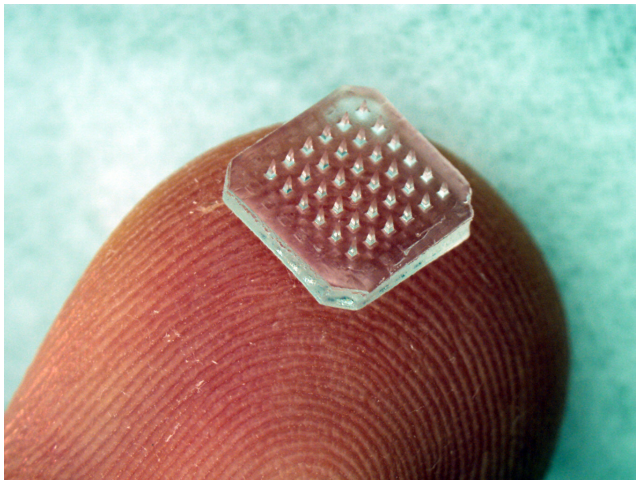
Adjuvants

- Stimulate early processes in immune recognition
- Produce a more robust acquired immune response with *less antigen*
  - *Slow release of antigen as site of inoculation*
  - *Inflammation*
- Licensed
  - *Alum (aluminum hydroxide or phosphate; in HBV vaccine) - US*
  - *AS04 in Cervarix (alum, monophosphoryl lipid A, TLR4 ligand) - US*
  - *MF59 - squalene oil-in-water emulsion (depot, innate stimulatory) - Europe*

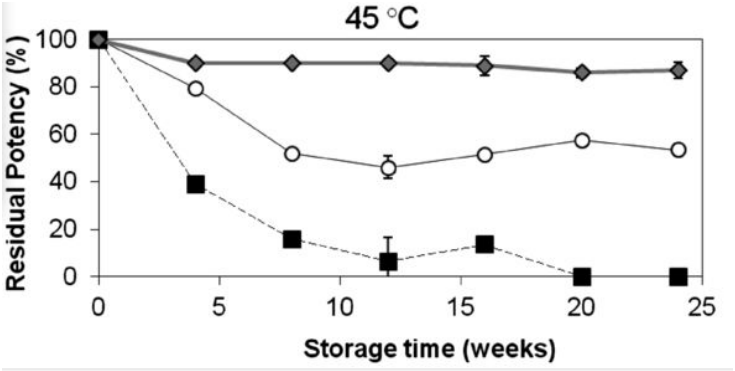




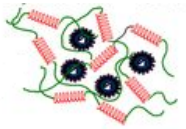
New vaccine technologies



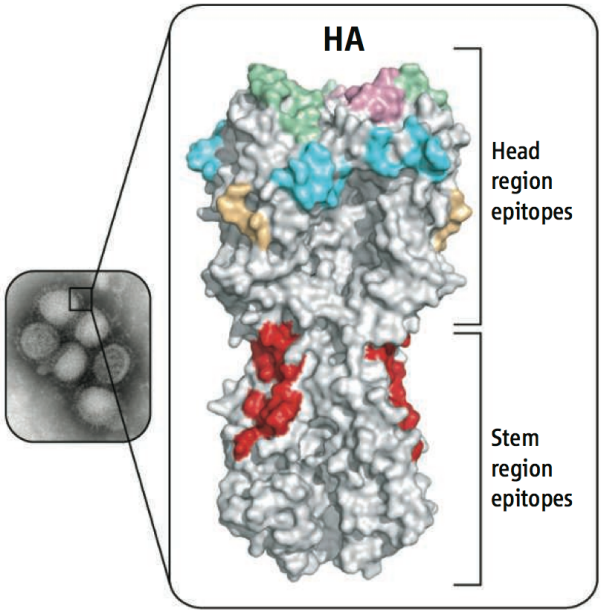
Microneedle patch

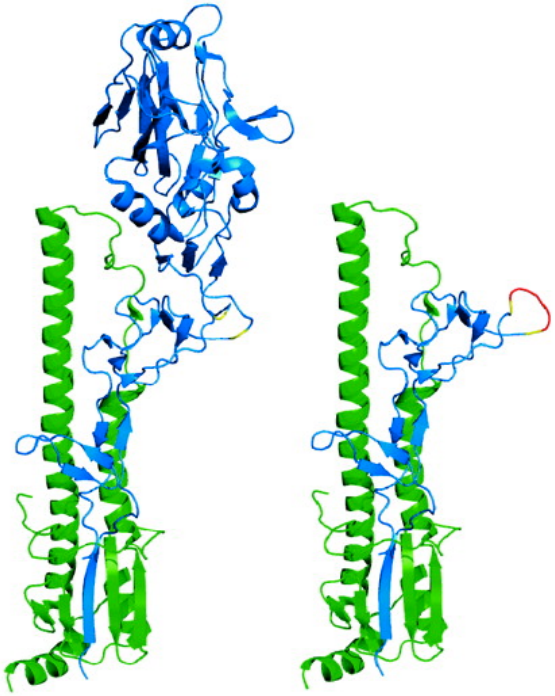


Thermostabilization in silk (or sugars)



Universal influenza vaccine





- Broadly neutralizing human mAbs
- Prime-boost
- HA stem antigen

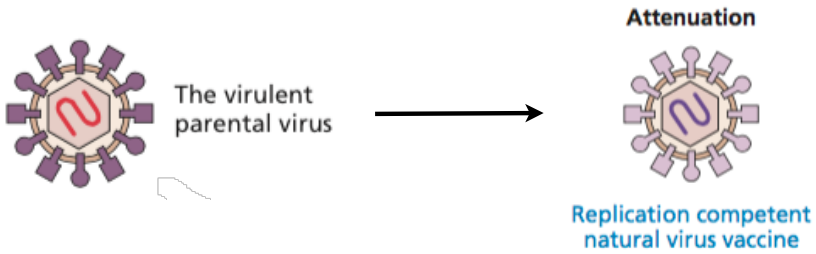
Go to:

b.socrative.com/login/student
room number: virus

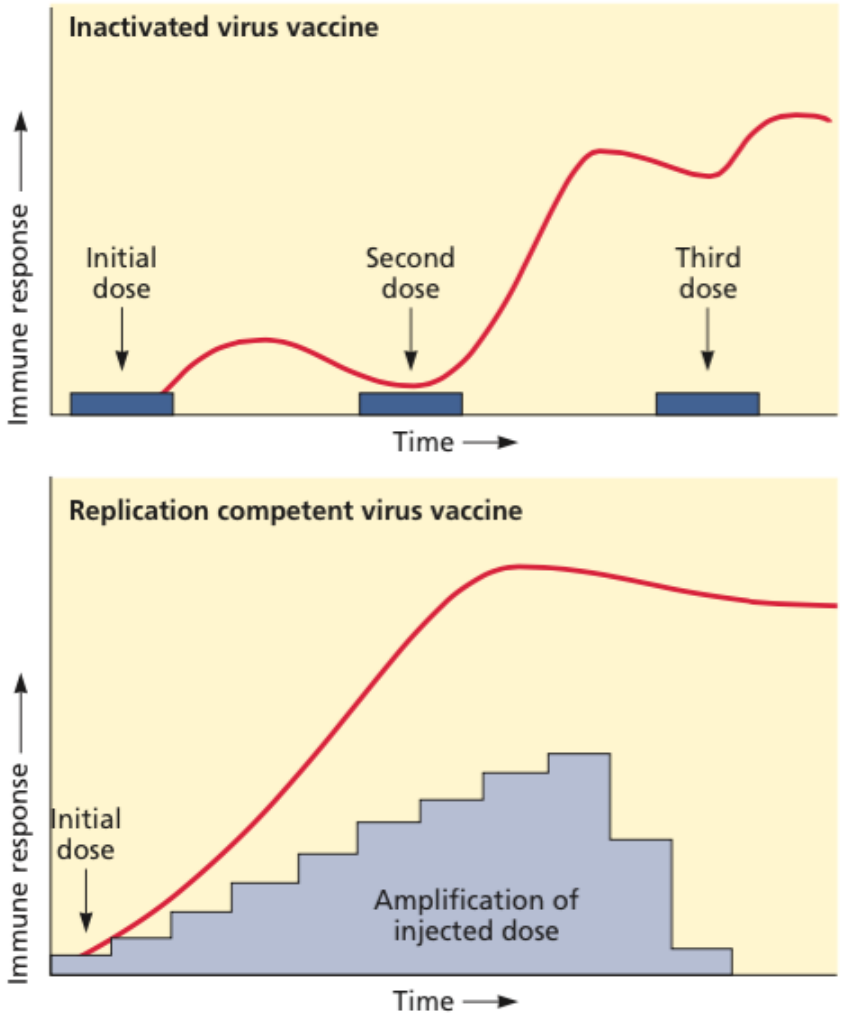
What are some requirements for an effective vaccine?

A. Low cost
B. Ease of administration
C. Provides long lasting immunity
D. Minimal side effects
E. All of the above

Replication competent, attenuated vaccines

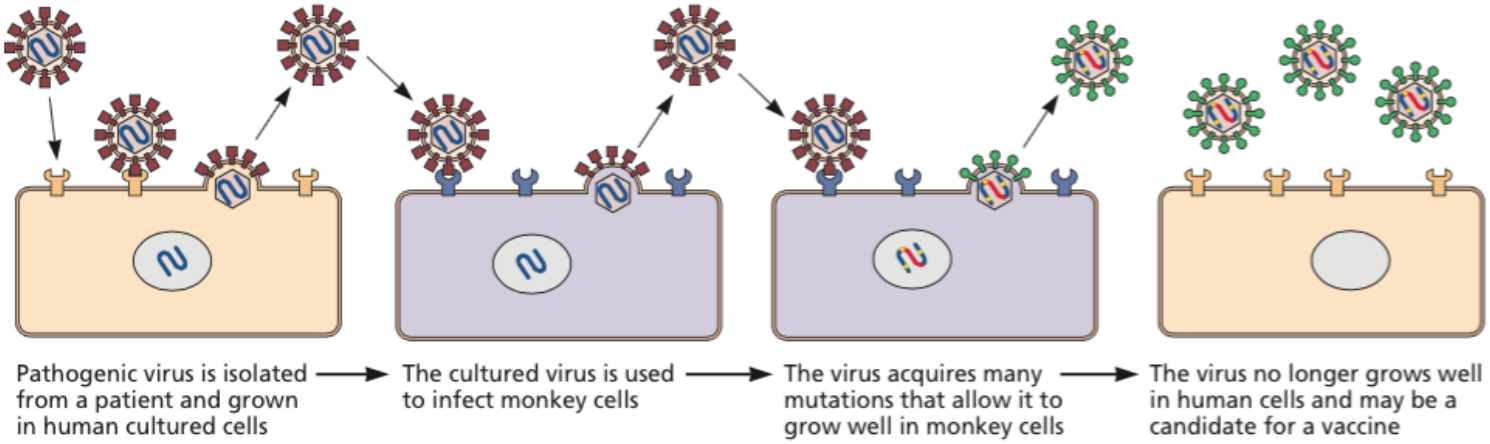


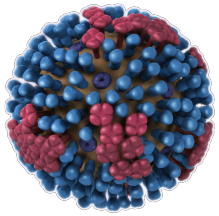
- Viral replication occurs, stimulates immune response
- Infection induces mild or inapparent disease



Principles of Virology, ASM Press

Empirically derived attenuated vaccines

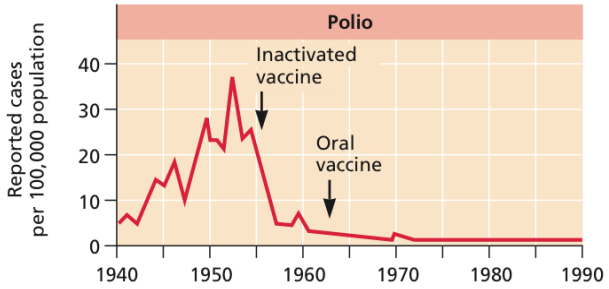


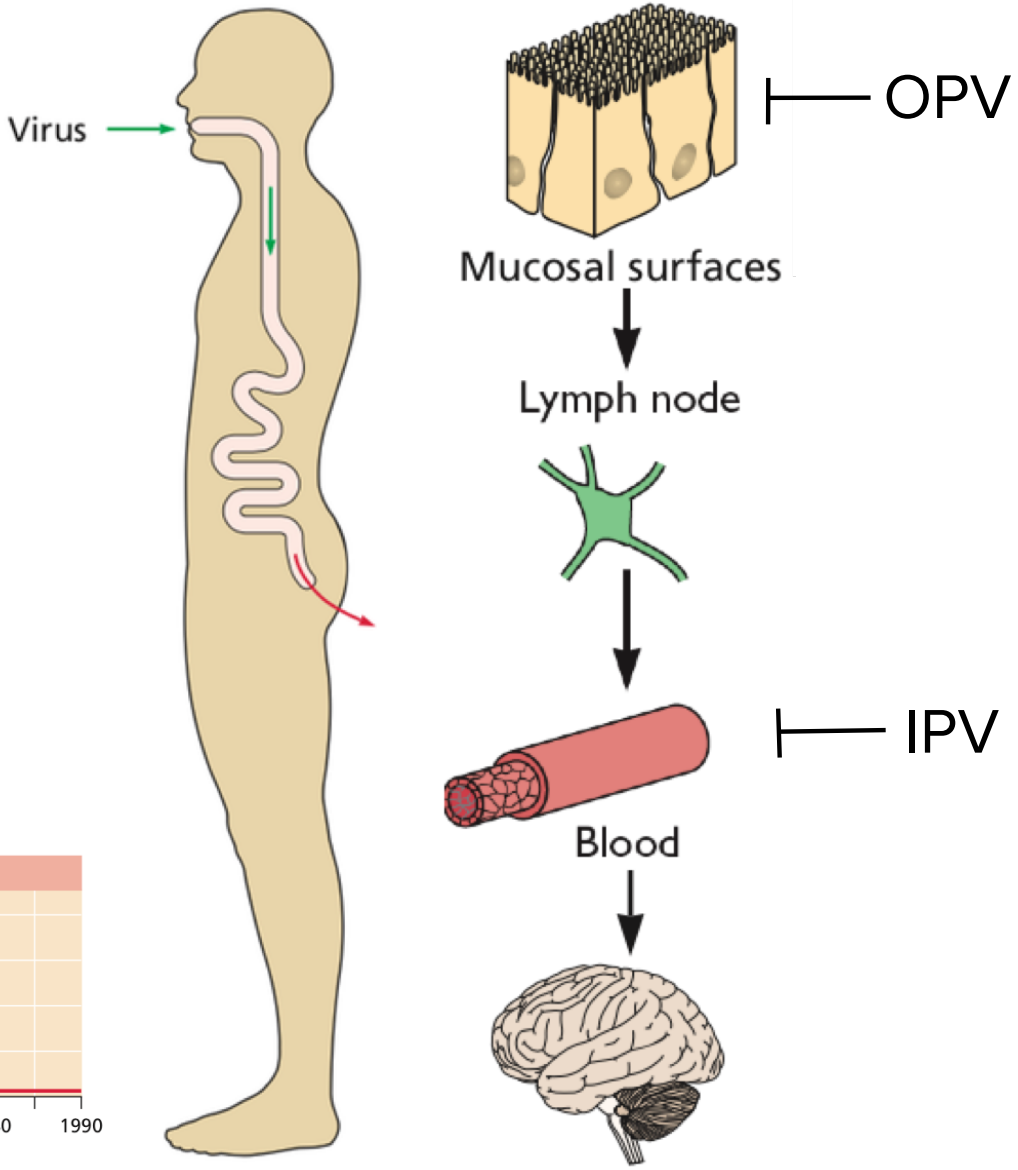


FluMist

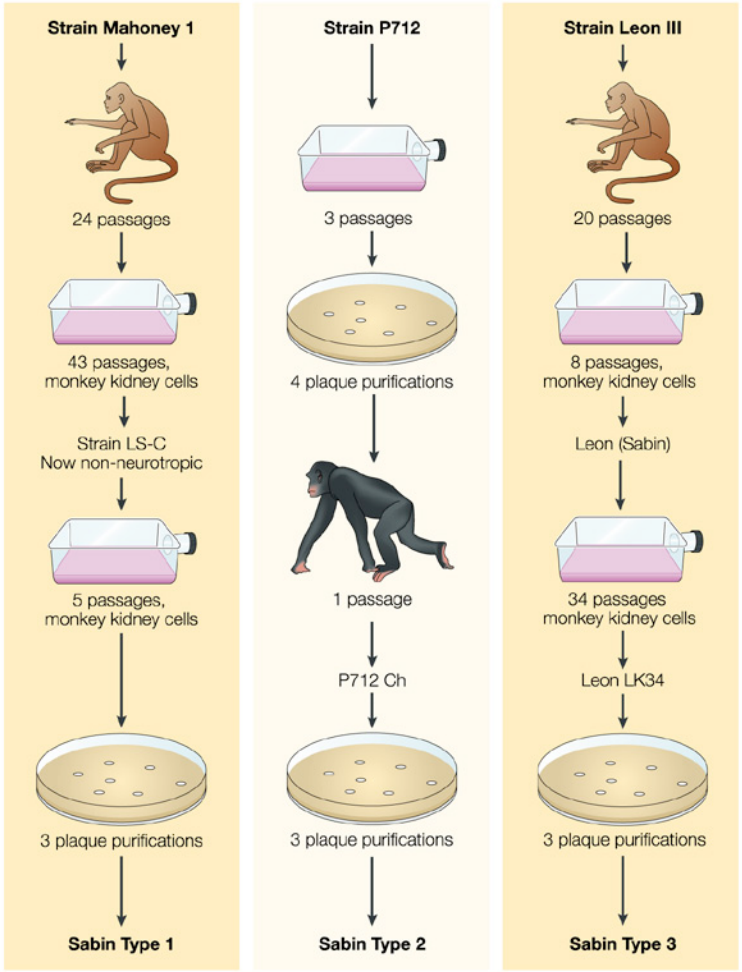
- Replication competent, intranasally administered influenza vaccine
- Multivalent
- Reassortants of master donor strain - HA, NA genes from current strains
- Viruses are cold-adapted, temperature-sensitive, and attenuated in a ferret model
- Replicate only in nasopharynx, produce protective immunity

Sabin oral poliovirus vaccine (OPV)





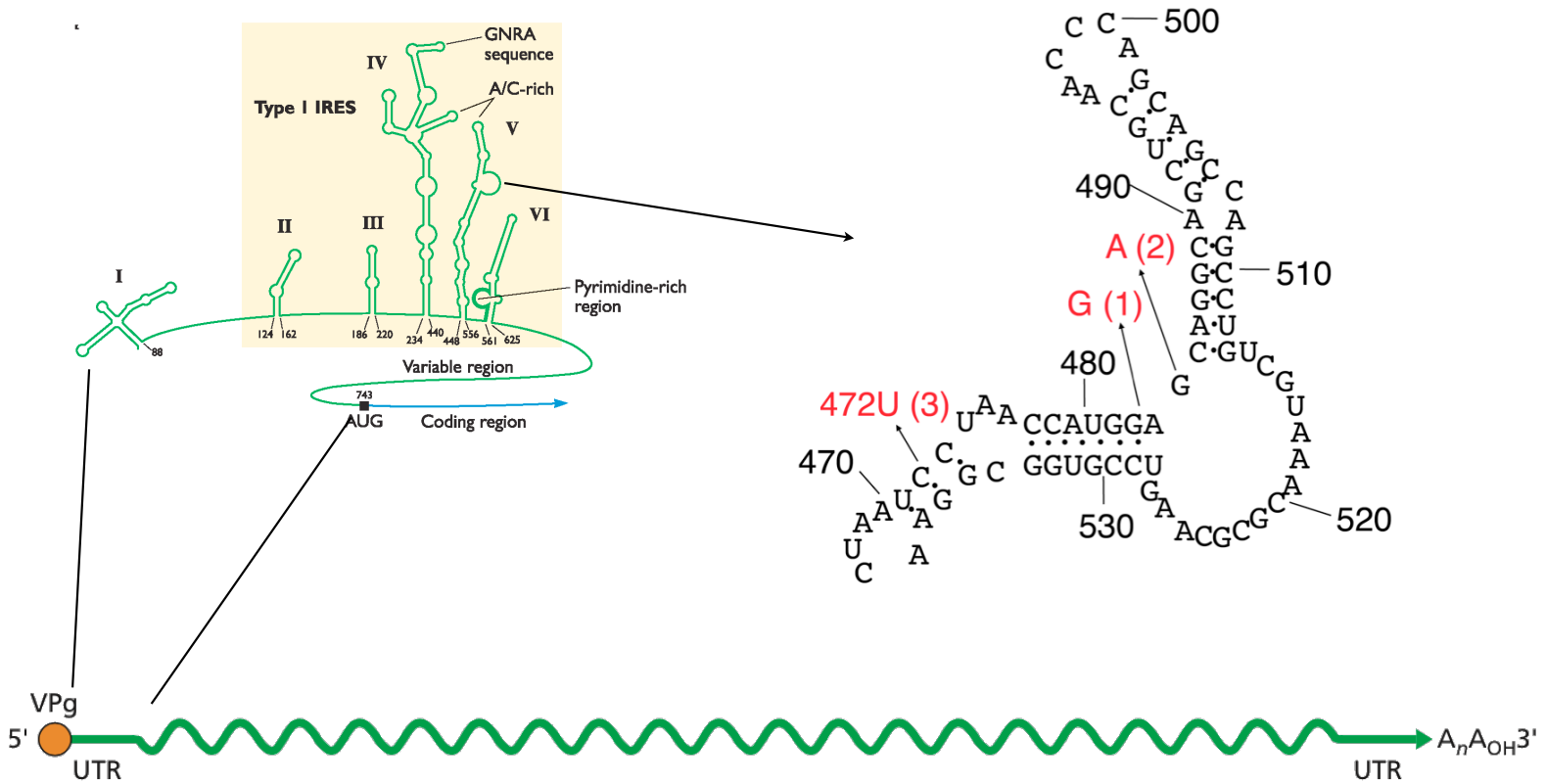
Attenuation of poliovirus neurovirulence



Albert Sabin's three strains of OPV licensed in the US in 1961

Determinants of Sabin vaccine strain attenuation

Virus	Mutation
P1/Sabin	5'-UTR nt 480 VP1 aa 1106 VP1 aa 1134 VP3 aa 3225 VP4 aa 4065
P2/Sabin	5'-UTR nt 481 VP1 aa 1143
P3/Sabin	5'-UTR nt 472 VP3 aa 3091

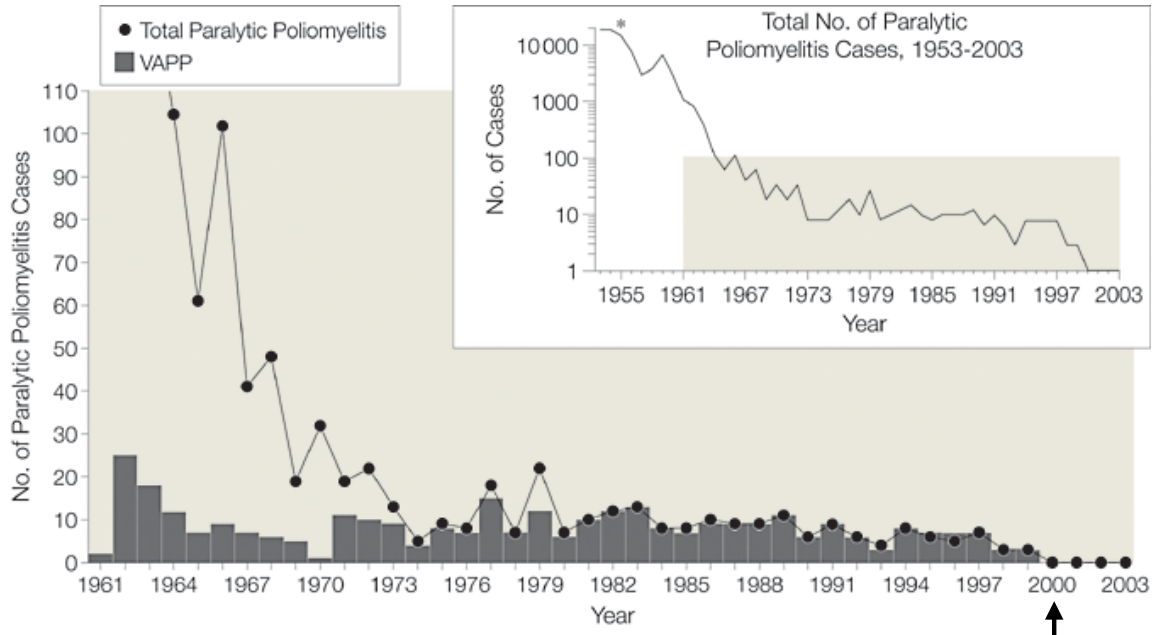

Type I IRES
GNRA sequence
A/C-rich
Pyrimidine-rich region
Variable region
AUG
Coding region
VPg
5'
UTR
UTR
$A_nA_{OH}3'$
500
490
A (2)
510
G (1)
480
472U (3)
470
530
520

Reversion of P3/Sabin

Virus	Base at 472	Time of isolation after vaccination	Histological lesion score
Sabin vaccine	U		0.36
DM1	U	24 h	ND
DM2	U	31 h	1.58
DM3	U/C	35 h	ND
DM4	C	47 h	2.48
DM38	C	18 da	ND
P3/119	C	3-4 weeks	3.34

Evans et al., Nature 314:548 (1985)

Reported Cases of Paralytic Poliomyelitis, United States, 1961-2003

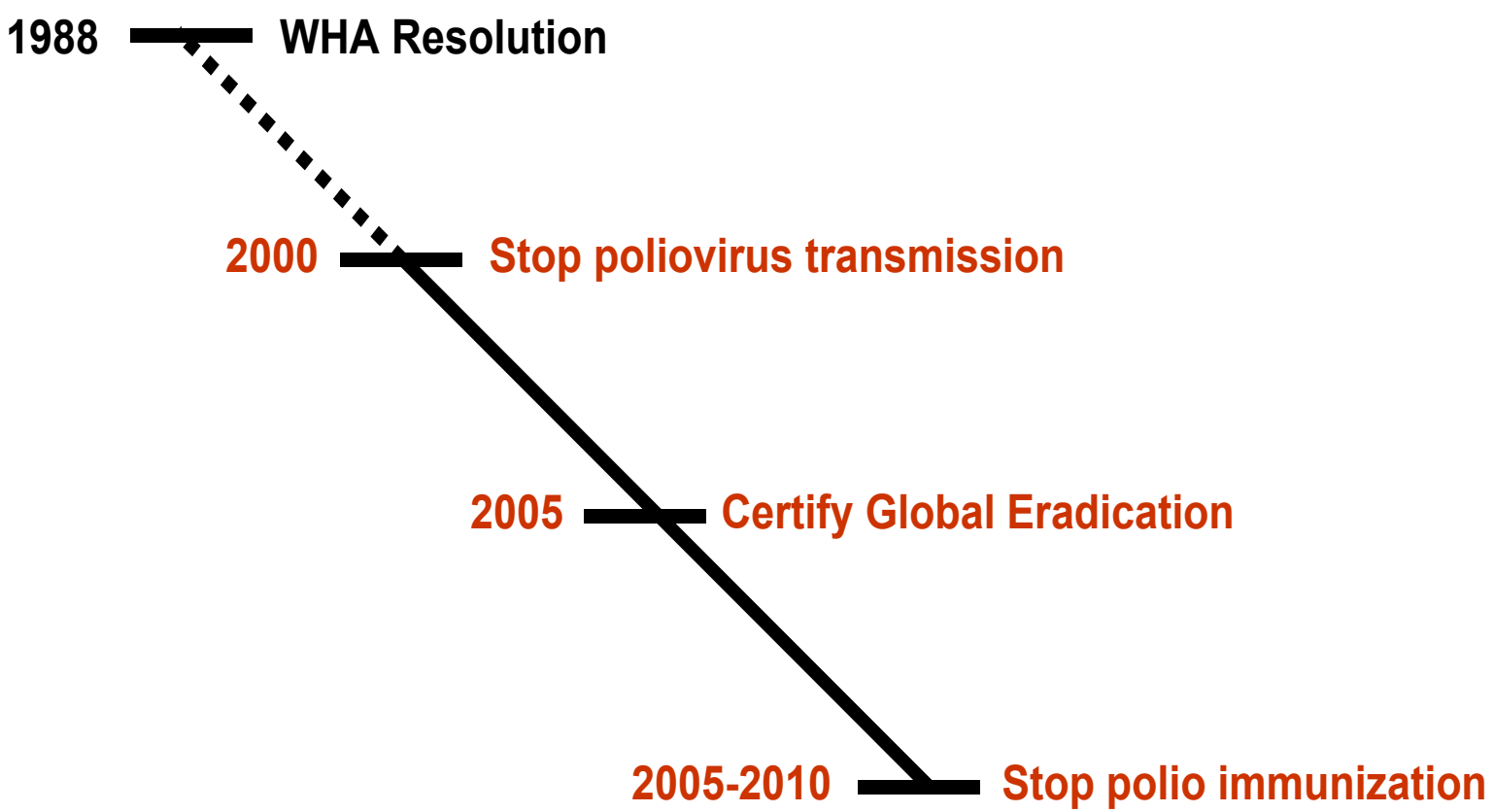


Alexander, L. N. et al. JAMA 2004;292:1696-1701.

switch to IPV

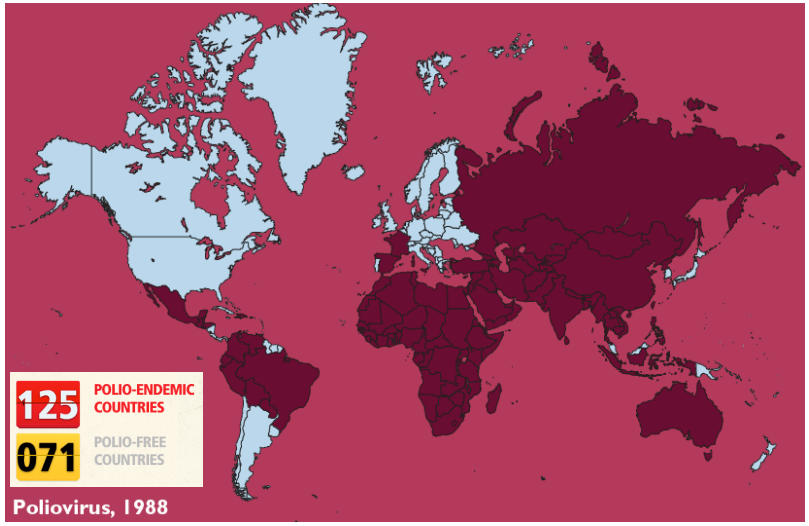
1 paralytic case/1.4 million doses

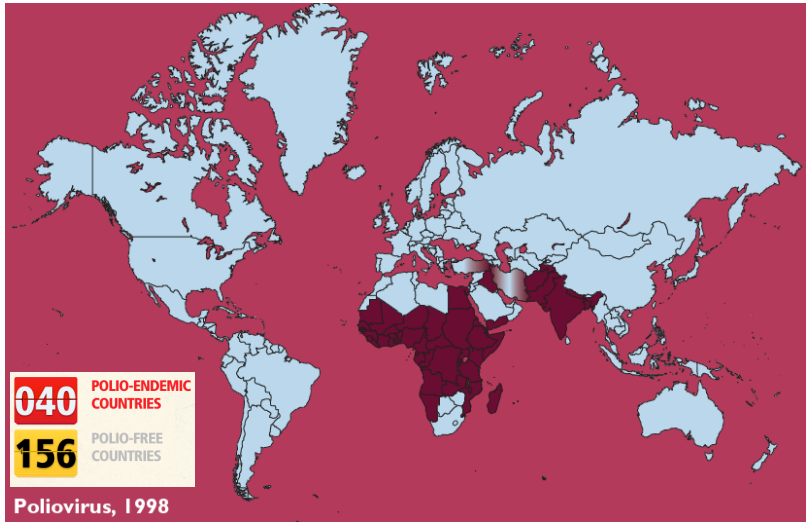
Eradication of poliomyelitis

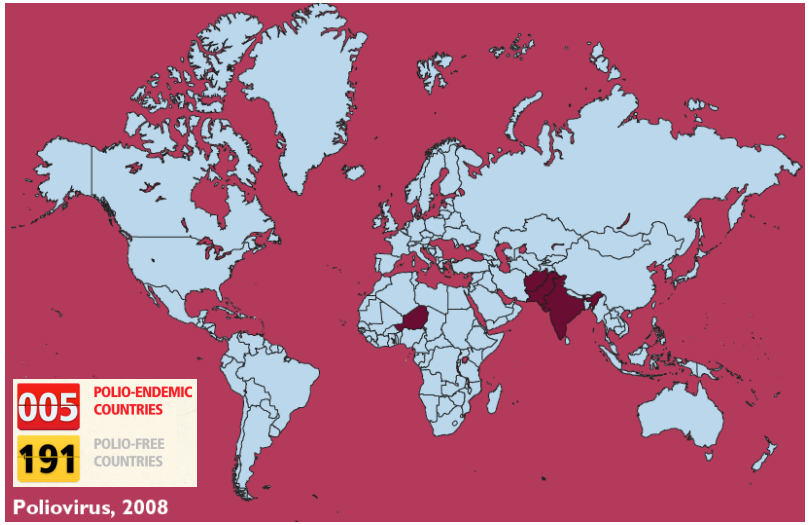


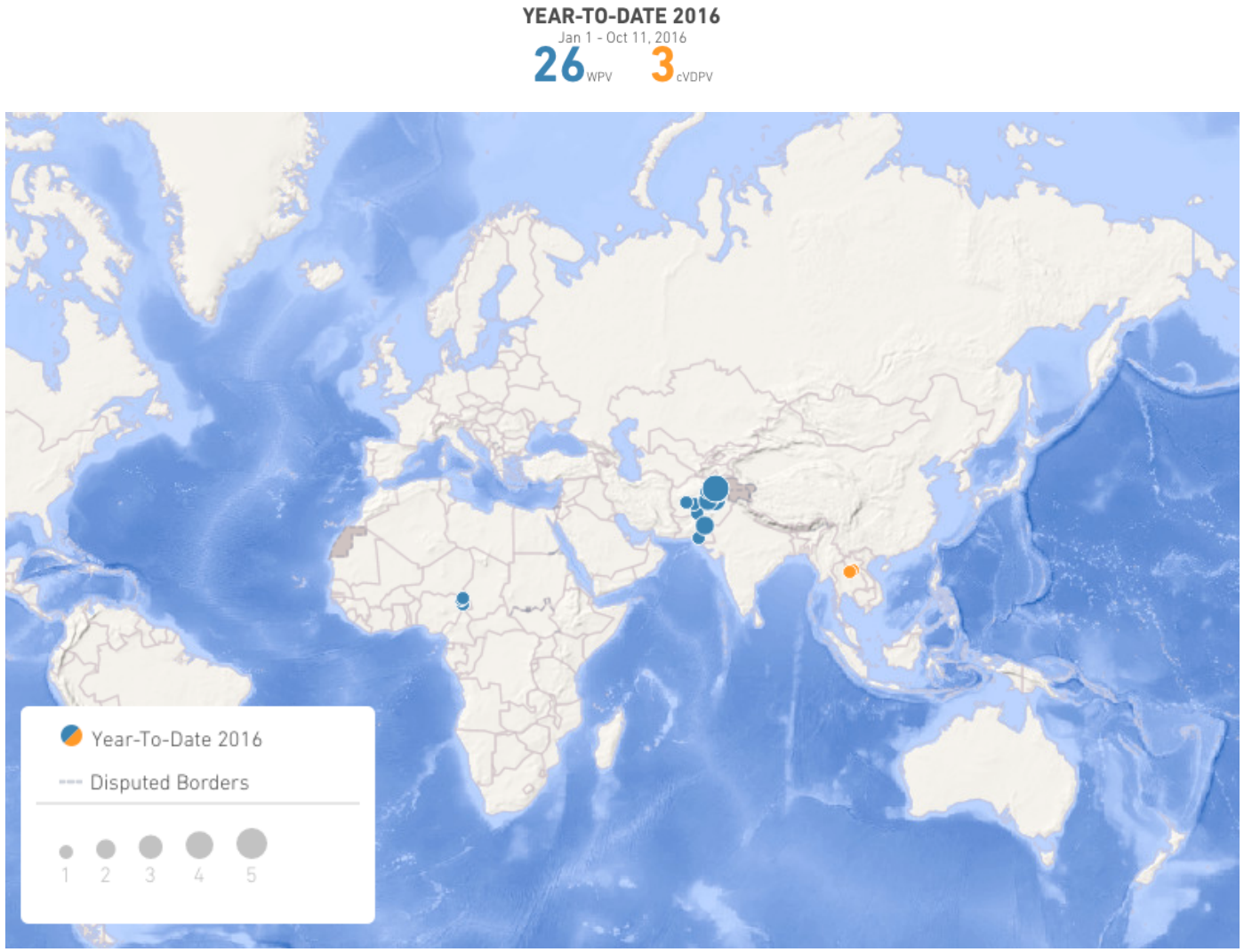
Can viral diseases be eradicated?

- Smallpox eradication program launched 1967, eradicated 1978
- Two features essential for eradication:
  - Replication in only one host
  - Vaccination induces lifelong immunity


125
POLIO-ENDEMIC COUNTRIES
071
POLIO-FREE COUNTRIES
Poliovirus, 1988


040
POLIO-ENDEMIC COUNTRIES
156
POLIO-FREE COUNTRIES
Poliovirus, 1998


005
POLIO-ENDEMIC COUNTRIES
191
POLIO-FREE COUNTRIES
Poliovirus, 2008



polioeradication.org

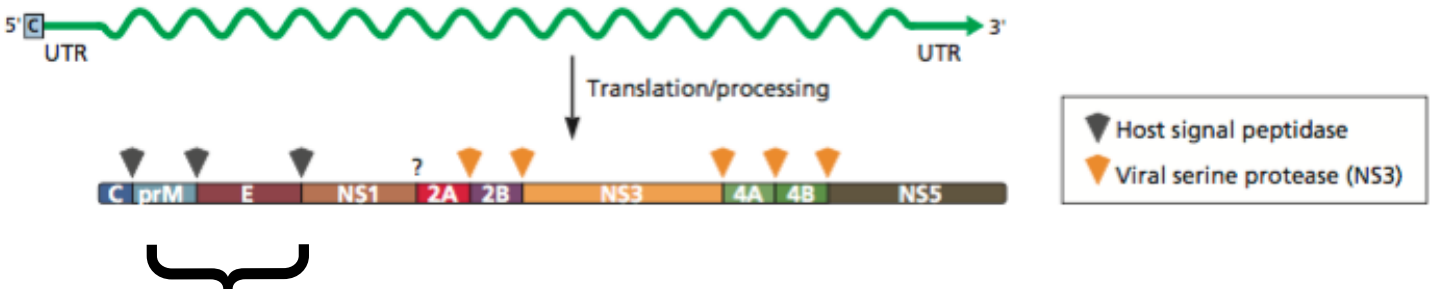
(wild type 2 declared eradicated, no type 3 since 2012)

Even if we eradicate a virus from the earth, as long as the nucleotide sequence is known...

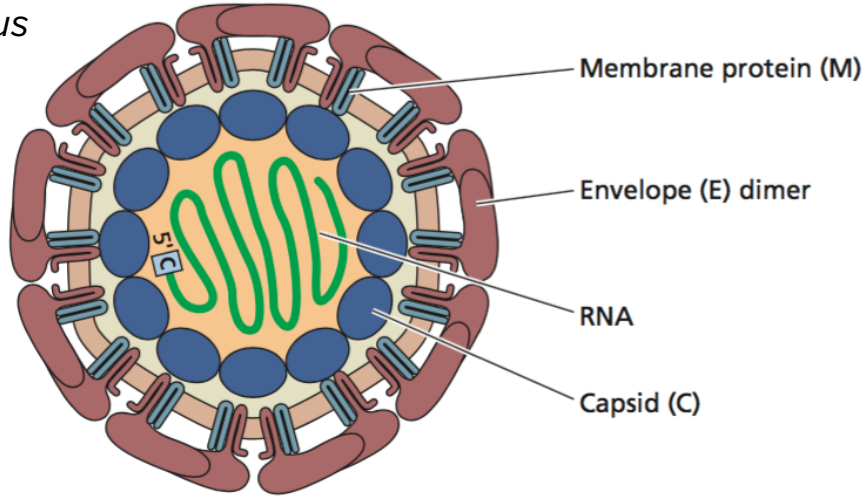
Engineering attenuated vaccines

- Yellow fever: first human virus identified, 1901
- Mosquito transmitted flavivirus
- Disease: fever and nausea to failure of major organ systems; high fatality
- Yellow fever vaccine 17D produced 1938 by 176 passages of virulent wild type Asibi strain in chick embryo tissue
- 500 million doses distributed; safe, effective

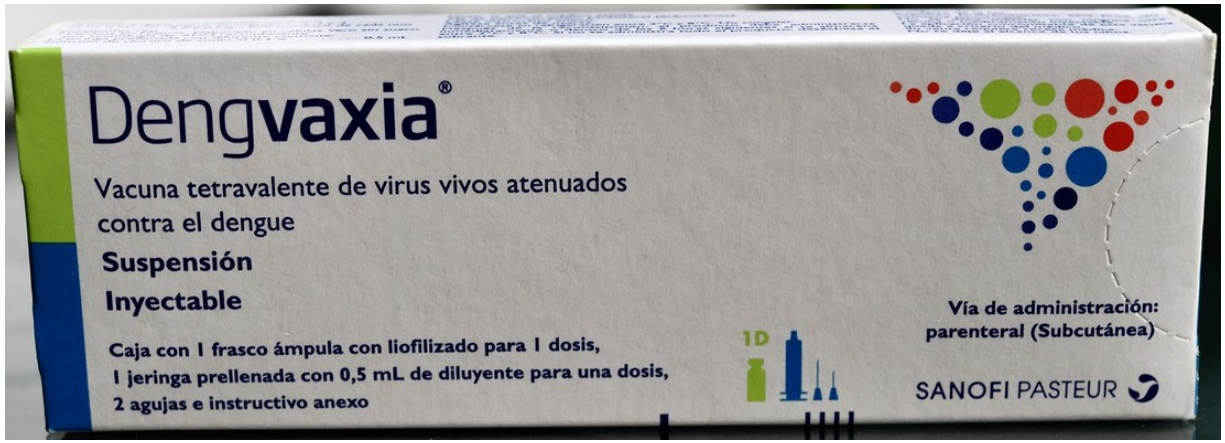
Building on success of YF 17D vaccine



Replace with dengue virus



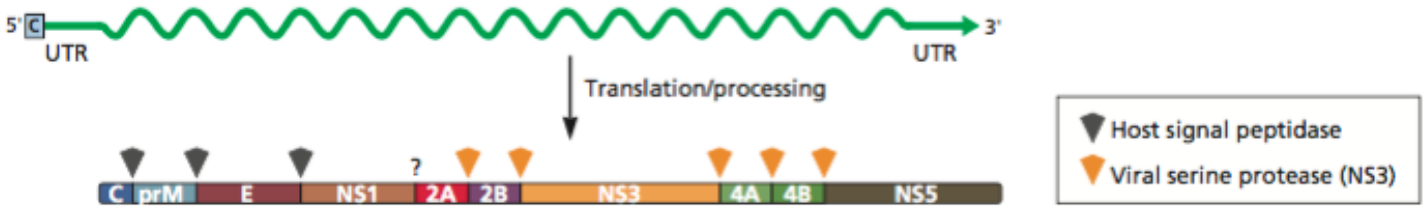
Dengvaxia



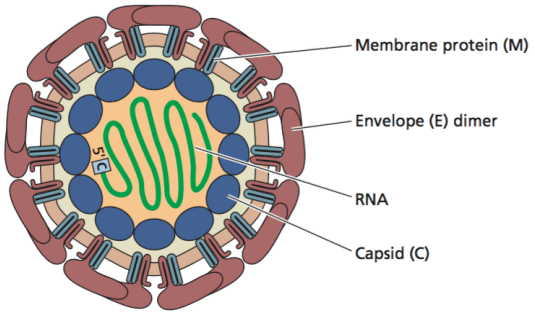
- E, prM of dengue virus 1, 2, 3, 4 in YF 17D backbone
- Licensed in Mexico, Brazil, Philippines
- No protection against DENV-2
- Lead to worse disease in 2-9 yo

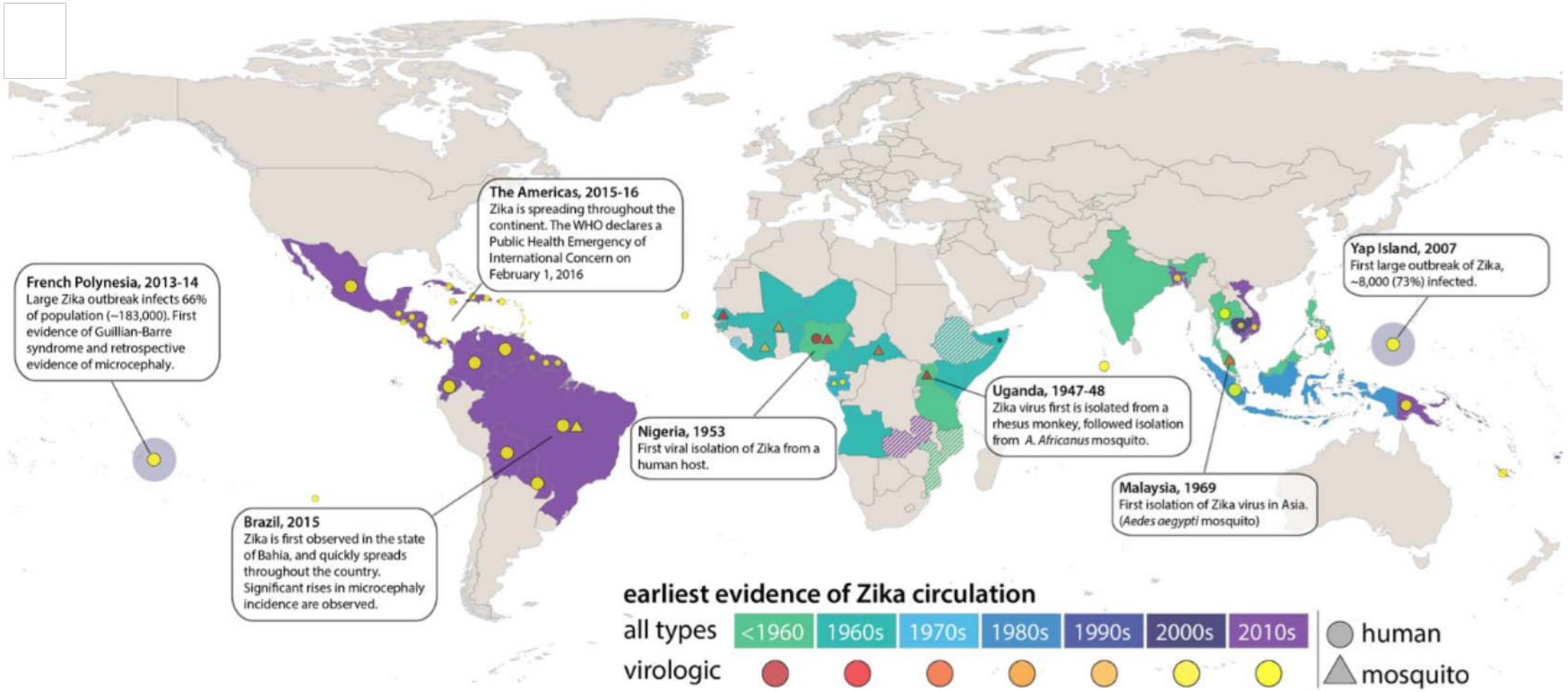
TV003

- Tetravalent, attenuated dengue virus vaccine produced by mutagenesis of infectious clone
- One dose, 100% protection vs challenge



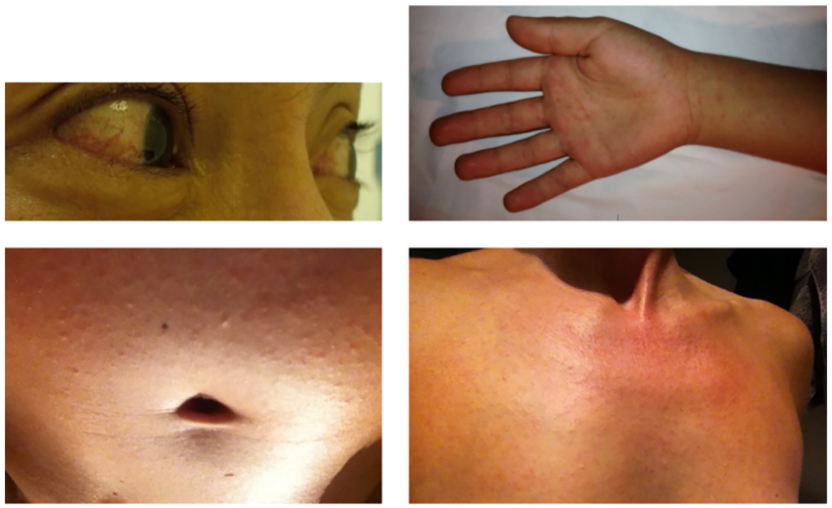
Zika virus


Membrane protein (M)
Envelope (E) dimer
RNA
Capsid (C)
5'
C


The Americas, 2015-16
Zika is spreading throughout the continent. The WHO declares a Public Health Emergency of International Concern on February 1, 2016
French Polynesia, 2013-14
Large Zika outbreak infects 66% of population (~183,000). First evidence of Guillian-Barre syndrome and retrospective evidence of microcephaly.
Brazil, 2015
Zika is first observed in the state of Bahia, and quickly spreads throughout the country. Significant rises in microcephaly incidence are observed.
Nigeria, 1953
First viral isolation of Zika from a human host.
Uganda, 1947-48
Zika virus first is isolated from a rhesus monkey, followed isolation from A. Africanus mosquito.
Malaysia, 1969
First isolation of Zika virus in Asia. (Aedes aegypti mosquito)
Yap Island, 2007
First large outbreak of Zika, ~8,000 (73%) infected.
earliest evidence of Zika circulation
all types
<1960
1960s
1970s
1980s
1990s
2000s
2010s
virologic
human
mosquito

Zika virus

- Disease: rash, fever, joint pain, conjunctivitis, headache (similar to dengue, chikungunya)
- Incubation period 2-10 days
- 1 in 5 develop symptoms; 5 day course
- Fatalities rare



Central nervous system complications associated with Zika virus infection

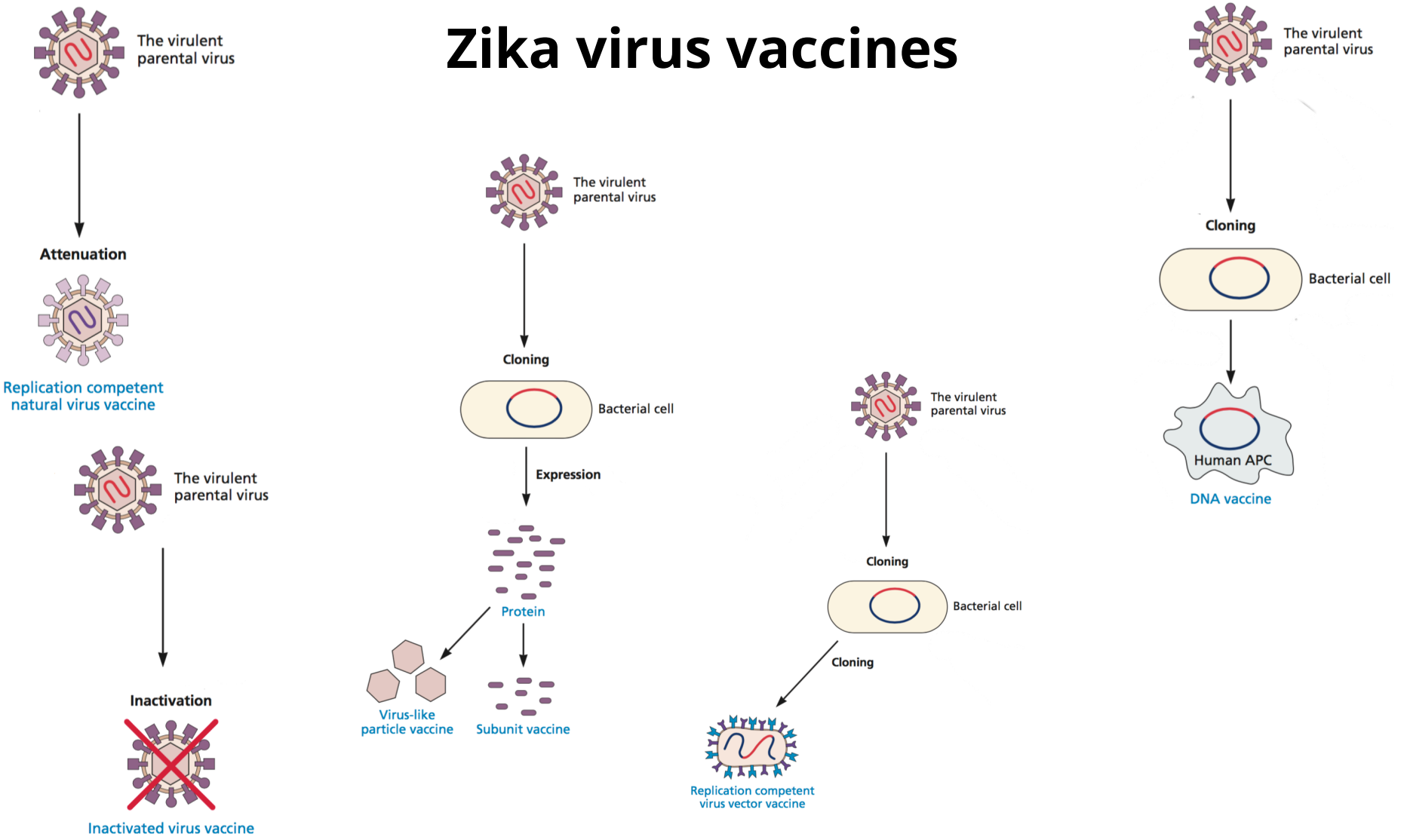
Adults

- Guillain-Barré Syndrome (post-infection autoimmune neuropathy; weakness, paralysis, death)
- Acute myelitis
- Encephalopathy
- Meningoencephalitis

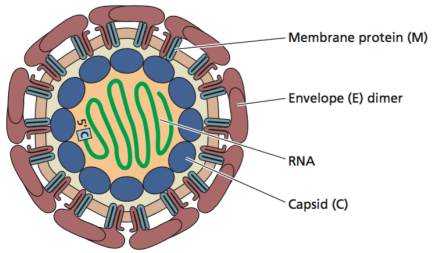
Infants

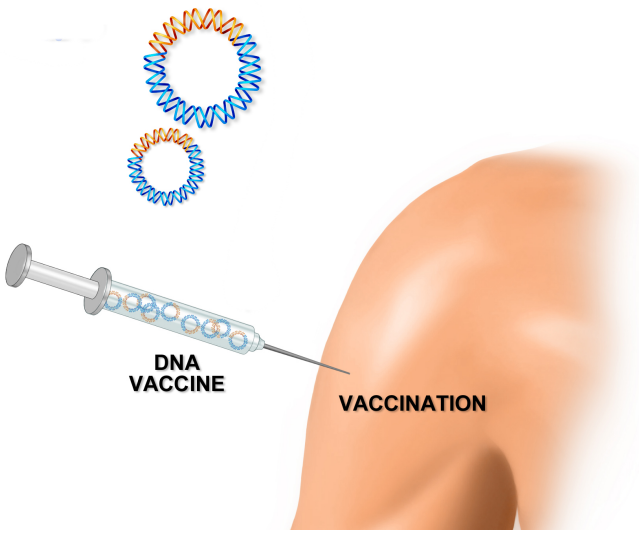
- Microcephaly
- Lissencephaly
- Macular atrophy

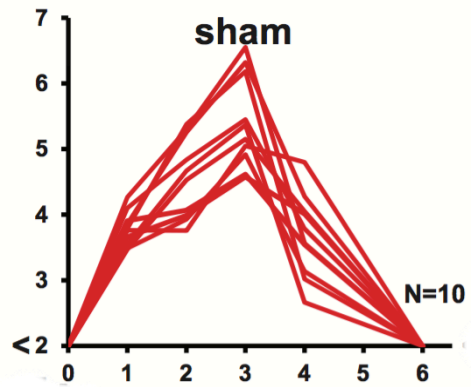
Zika virus vaccines

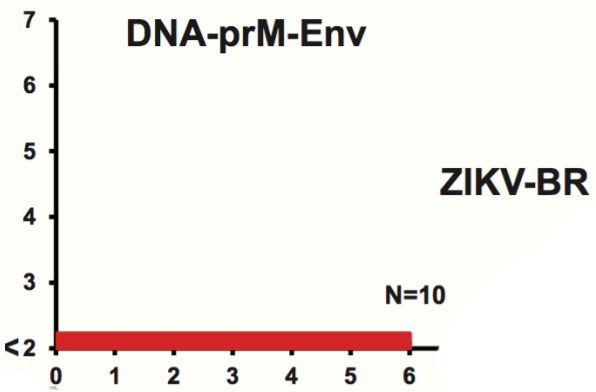


Zika virus DNA vaccine

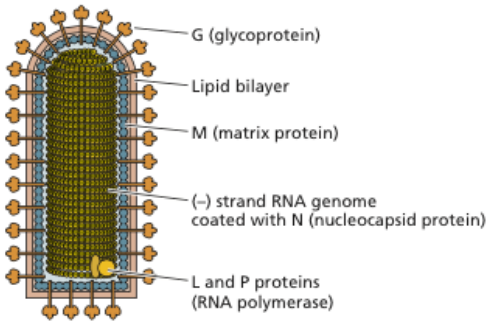


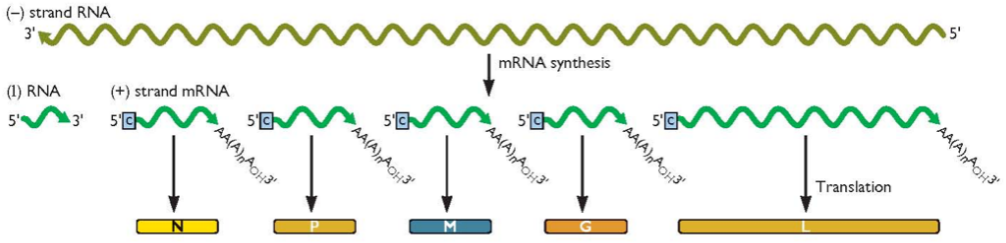


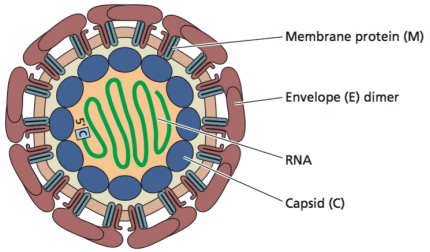


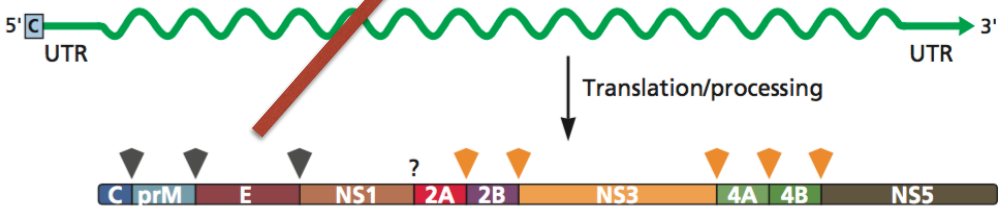


Vesicular stomatitis virus vector









Principles of Virology, ASM Press

For the US, a Zika virus vaccine
will be a travel vaccine (like yellow fever vaccine and others)