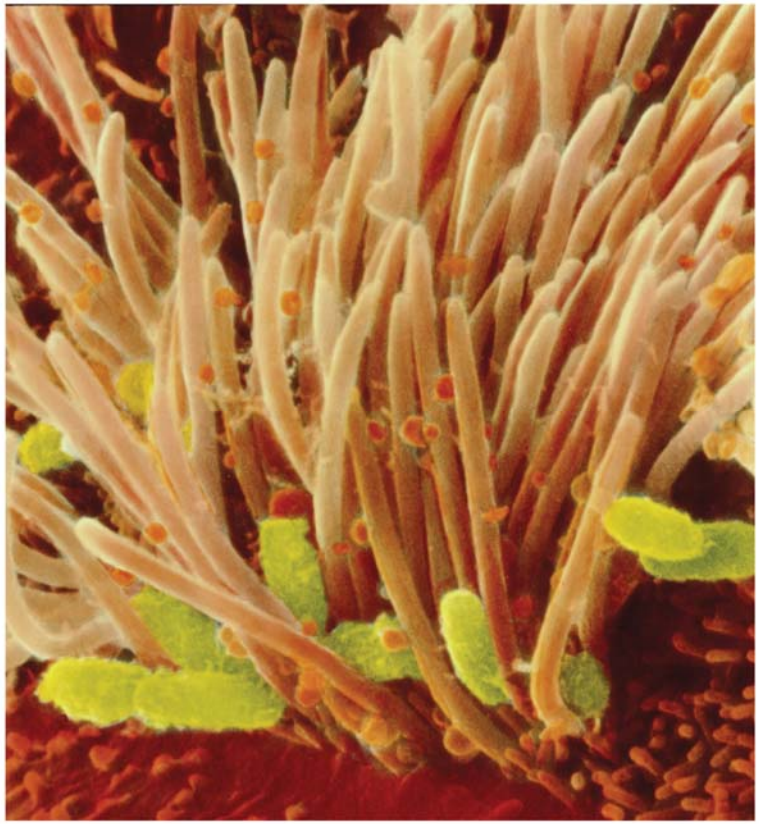
Chapter 18

Practical Applications of Immunology



History of Vaccines

- **Variolation**: practice which preceded vaccines
- As early as 1700s in Turkey / transfer “best sort of smallpox” with pin into venous blood of humans
- Infected person contract a mild illness that would last about a week
- Subsequently, protected from smallpox
- Technology was transferred to England where variolation was practiced
- Mortality rate associated with variolation was 1% // smallpox mortality rate was 50%

History of Vaccines

- At age 8 Edward Jenner, an English boy, received variolation.
- Years later Jenner became a physician and provided variolation to protect his patients from smallpox
- Noticed that one group (dairy maids) never became mildly sick following variolation
- Dairy maids did not fear smallpox // they already had cowpox
- Cowpox mild disease / causes lesions on cows' udders / dairy maids would become infected via hands during milking
- 1798 Jenner did experiments to show that infecting people with cowpox would protect them from smallpox
- Vaccine = suspension of organisms or fractions of organisms that is used to induce immunity
- Two centuries later / smallpox has been eliminated

History of Vaccines

- 1881 Louis Pasteur working on chicken cholera, pathogenic bacteria
- Went on vacation and left plated bacteria on workbench
- On return from vacation he used these bacteria to inoculate test chickens // they did not become sick // got second sample of "vegetative" bacteria and re-inoculated same chickens // they did not become sick
- Conclusion: culture he left on bench during vacation had become weakened and now unable to cause disease -- but rendered the chickens immune
- Experimented to deliberate weaken pathogens (attenuation) to make live attenuated vaccines
- Live vaccines more closely mimic actual infections // induce both cellular and humoral immunity

History of Vaccines

- Many communicable diseases can be controlled by behavioral or environmental methods
- E.g. good sanitation can prevent spread of cholera and use of condoms can slow spread of sexual transmitted diseases
- Viral diseases can not be effectively treated once contracted
- Vaccinations often only feasible method of controlling viral diseases
- If most of the population is vaccinated, virus can not spread // herd immunity
- Outbreaks become limited to sporadic // not enough host within community to allow spread of virus (or bacterial infection)

Reported numbers of measles cases in the United States, 1960–2010. (CDC, 2010)

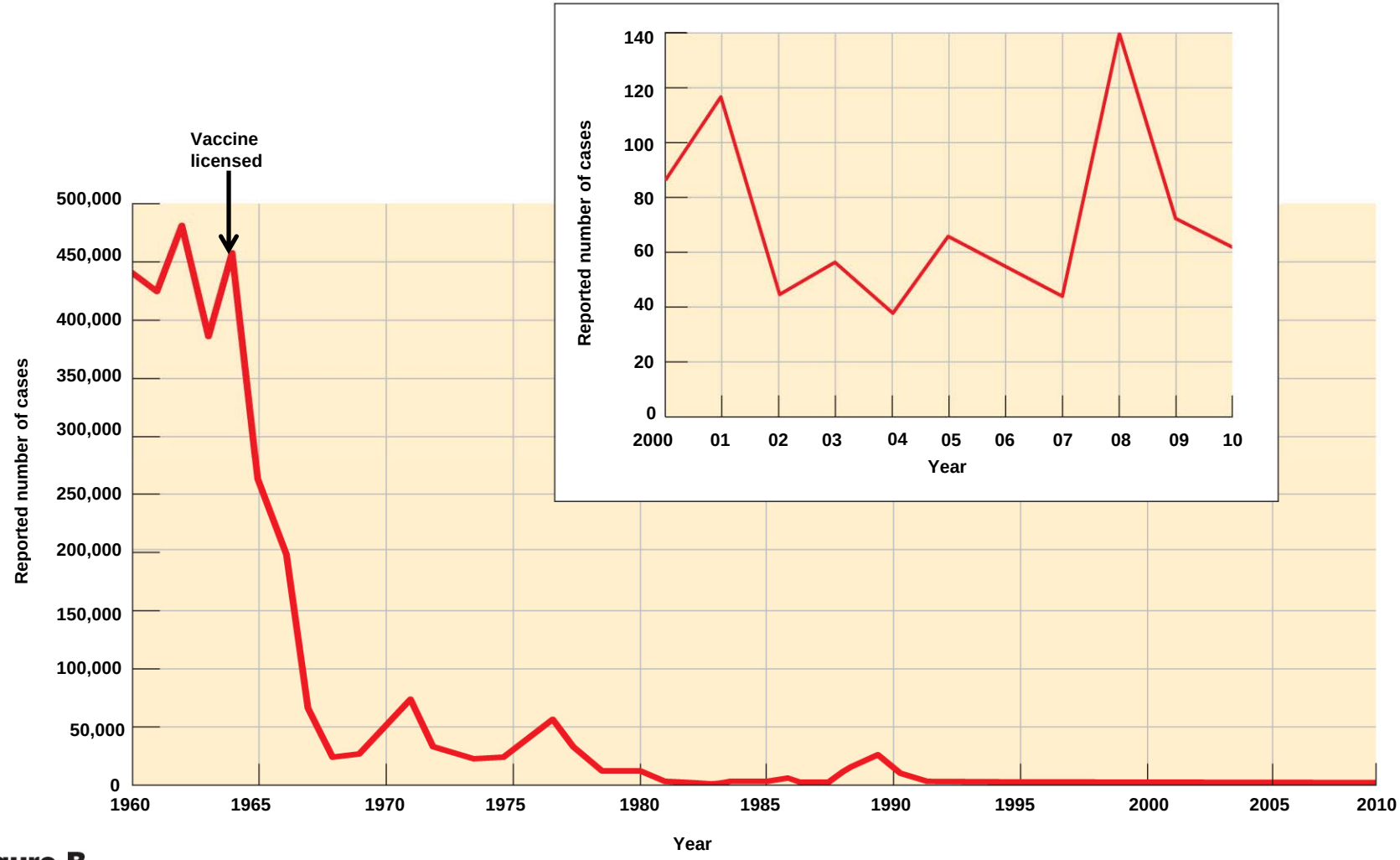


Figure B

Countries with the highest measles mortality.

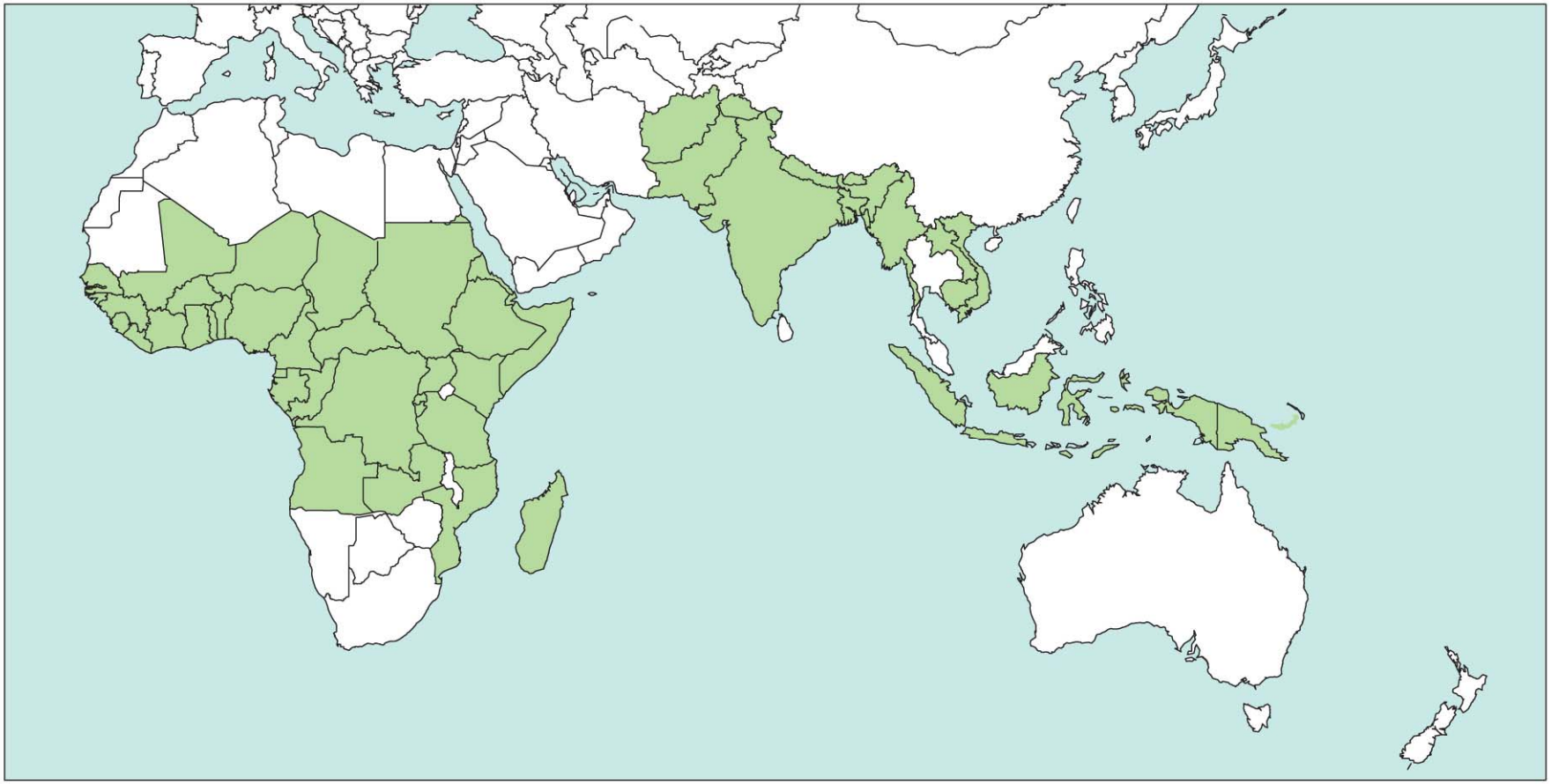


Figure C

Vaccines Used to Prevent Bacterial Diseases

Disease	**Vaccine**
Diphtheria	Purified diphtheria toxoid
Meningococcal meningitis	Purified polysaccharide from *Neisseria meningitidis*
Pertussis (whooping cough)	Inactivated toxin plus acellular fragments of *Bordetella pertussis*
Pneumococcal pneumonia	Purified polysaccharide from seven strains of *Streptococcus pneumoniae*
Tetanus	Purified tetanus toxoid
Haemophilus influenzae type b meningitis	Polysaccharide from *Haemophilus influenzae* type b conjugated with protein to enhance effectiveness

Vaccines Used to Prevent Viral Diseases

Disease	Vaccine
Influenza	Injected vaccine, inactivated virus (nasally administered: attenuated virus)
Measles	Attenuated virus
Mumps	Attenuated virus
Rubella	Attenuated virus
Chickenpox	Attenuated virus
Poliomyelitis	Killed virus

Vaccines Used to Prevent Viral Diseases

Disease	Vaccine
Rabies	Killed virus
Hepatitis B	Antigenic fragments of virus
Hepatitis A	Inactivated virus
Smallpox	Live vaccinia virus
Herpes zoster	Attenuated virus
Human papillomavirus	Antigenic fragments of virus

Types of Vaccines

- Live attenuated vaccines // often provided lifelong immunity // effectiveness 95%
- Inactivated killed vaccines // microbes that have been killed or virus that have been inactivated // often require booster shots // induce mostly humoral antibody immunity
- Subunit vaccines // use only antigenic fragments // avoids danger associated with live or killed pathogens // subunits produced by genetic modification of bacteria to produce subunit vaccines = recombinant vaccines
- Toxoids // inactivated toxins (only exotoxins) // often require booster shots // e.g. tetanus and diphtheria toxoids
- Currently // 20 separate injections are recommended for infants and children

Types of Vaccines

- **Attenuated whole-agent vaccines //**
  e.g. MMR vaccine

- **Inactivated whole-agent vaccines //**
  e.g. Salk polio vaccine

- **Toxoids //** **e.g.** Tetanus vaccine

Types of Vaccines

- **Subunit vaccines**
  - Acellular - pertussis
  - Recombinant - hepatitis B
- **Nucleic acid (DNA) vaccines**
  - West Nile (for horses)

Vaccines for Persons Aged 0–6 Years

- Hepatitis B
- Rotavirus
- DTaP
- *Haemophilus influenzae* type b
- Pneumococcal
- Inactivated poliovirus
- Influenza

Vaccines for Persons Aged 0–6 Years

- MMR
- Varicella
- Hepatitis A
- Meningococcal

TABLE 18.3 Recommended Immunization Schedule for Persons Aged 0–6 Years—United States, 2011 (CDC)

Vaccine \ Age	Birth	1 month	2 months	4 months	6 months	12 months	15 months	18 months	19–23 months	2–3 years	4–6 years
Hepatitis B	HepB	HepB			HepB						
Rotavirus			Rv	Rv	Rv						
Diphtheria, Tetanus, Pertussis			DTaP	DTaP	DTaP		DTaP				DTaP
***Haemophilus influenzae* type b**			Hib	Hib	Hib	Hib					
Pneumococcal*			PCV	PCV	PCV	PCV				PPSV	
Inactivated Poliovirus			IPV	IPV	IPV						IPV
Influenza					Influenza (Yearly)						
Measles, Mumps, Rubella						MMR					MMR
Varicella						Varicella					Varicella
Hepatitis A†						HepA (2 doses)					
Meningococcal‡										MCV	

Note: Vaccines are listed under routinely recommended ages. Bars indicate range of recommended ages for immunization. For those who fall behind or start late, see the catch-up schedule. Additional information at www.cdc.gov/vaccines/recs/schedules/

* PCV = Pneumococcal conjugate vaccine, PPSV = Pneumococcal polysaccharide vaccine.

† The two doses at least 6 mo. apart.

‡ Meningococcal conjugate vaccine (MCV) for children aged 2–10 years with defective immune systems and certain other high risk situations.

TABLE 18.1 Principal Vaccines Used in the United States to Prevent Bacterial Diseases in Humans

Disease(s)	Vaccine	Recommendation	Booster
Tetanus, diphtheria, and pertussis	DTaP (children younger than 3), Tdap (older children and adults), Td (booster for tetanus and pertussis)	DTaP (months 2, 4, 6, 15–18; years 4–6);* Td (adults every 10 years); Tdap (similar to Td; single dose for children aged 11–12 years, or adults aged 19–64); booster every 10 years	Tdap (booster) every 10 years
Meningococcal meningitis	Purified polysaccharide from *Neisseria meningitidis*	For people with substantial risk of infection Recommended for college freshmen, especially if living in dormitories	Need not established
Pneumococcal pneumonia	Purified polysaccharide from seven strains of *Streptococcus pneumoniae*	For adults with certain chronic diseases; people over 65; children 2–23 months	None if first dose administered ≥ 24 months
Haemophilus influenzae type b meningitis	Polysaccharide from *Haemophilus influenzae* type b conjugated with protein to enhance effectiveness	Children prior to school age; see Table 18.3	None recommended

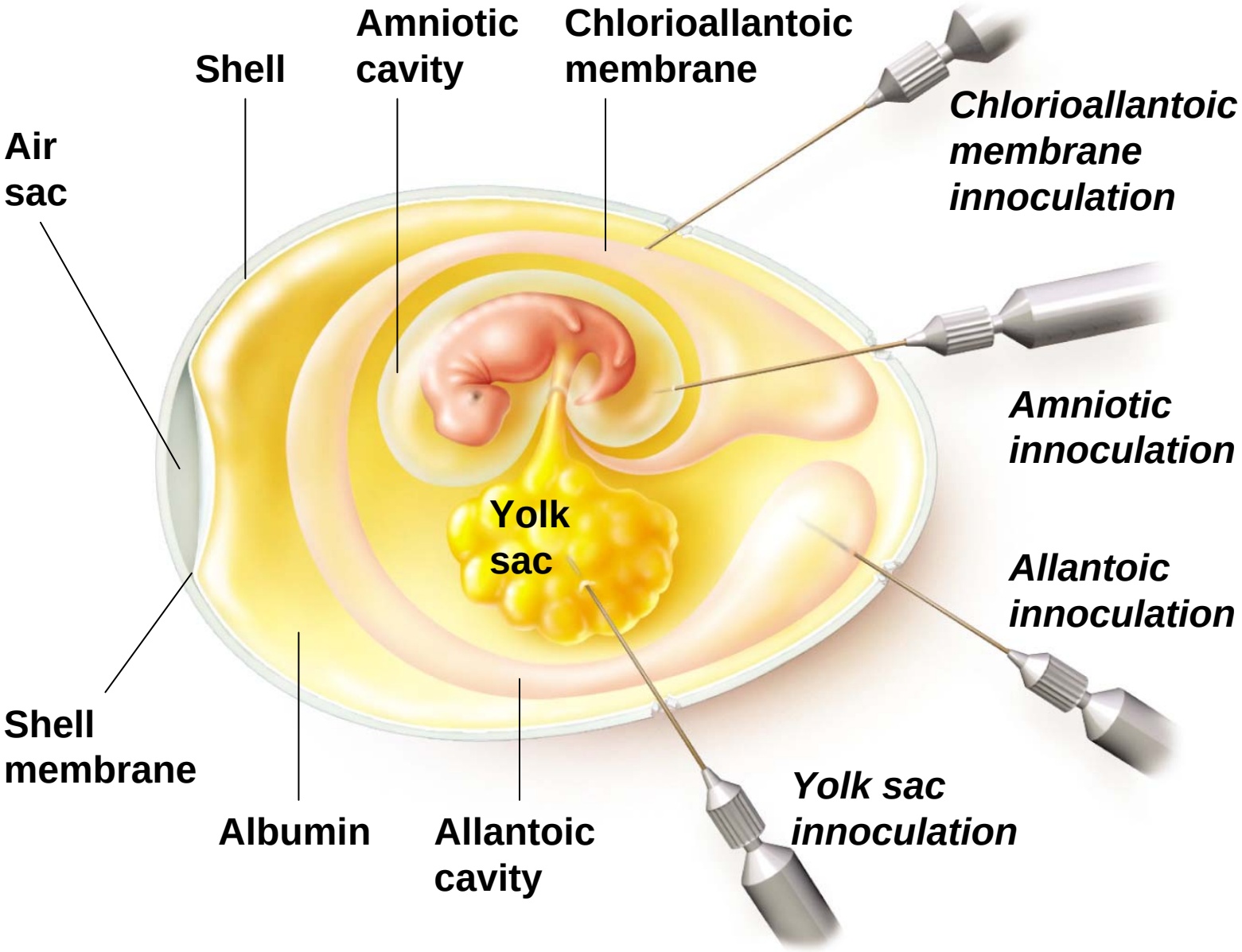
* For details, see www.cdc.gov/vaccines/vdp-vac/pertussis/

TABLE 18.2 Principal Vaccines Used in the United States to Prevent Viral Diseases in Humans

Disease	Vaccine	Recommendation	Booster
Influenza	Injected vaccine, inactivated virus (nasally administered vaccine with attenuated virus is now available for some)	For chronically ill, including children over 6 months. Adults over age 65. Healthy children aged 6–23 months (because higher risk of related hospitalizations). Health care workers and others in contact with high risk groups. Healthy persons aged 5–49 years can receive intranasal vaccine.	Annual
Measles	Attenuated virus	For infants aged 15 months	Adults if exposed during outbreak
Mumps	Attenuated virus	For infants aged 15 months	Adults if exposed during outbreak
Rubella	Attenuated virus	For infants aged 15 months; for women of childbearing age who are not pregnant	Adults if exposed during outbreak
Chickenpox	Attenuated virus	For infants aged 12 months	(Duration of immunity not known)
Poliomyelitis	Killed virus	For children, see Table 18.3; for adults, as risk to exposure warrants.	(Duration of immunity not known)
Rabies	Killed virus	For field biologists in contact with wildlife in endemic areas; for veterinarians; for people exposed to rabies virus by bites.	Every 2 years
Hepatitis B	Antigenic fragments of virus	For infants and children, see Table 18.3; for adults, especially health care workers, homosexual men, injecting drug users, heterosexual people with multiple partners, and household contacts of hepatitis B carriers.	Duration of protection at least
Hepatitis A	Inactivated virus	Mostly for travel to endemic areas and protecting contacts during outbreaks	Duration of protection estimated at about 10 years
Smallpox	Live vaccinia virus	Certain military and health care personnel	Duration of protection estimated
Herpes zoster	Attenuated virus	Adults over age 60	None recommended
Human papilloma virus	Antigenic fragments of virus	All females under age 26. Boys optional.	Duration at least 5 years

Influenza viruses are grown in embryonated eggs.

Inoculation of an embryonated egg.



Adjuvants

- Early days of commercial vaccine // occasional contaminations
- After vaccine was further purified // vaccine because less effective
- Led to understanding that chemical additives could increase effectiveness of vaccines
- These chemical additives called adjuvants // e.g. aluminum salts

Safety of Vaccines

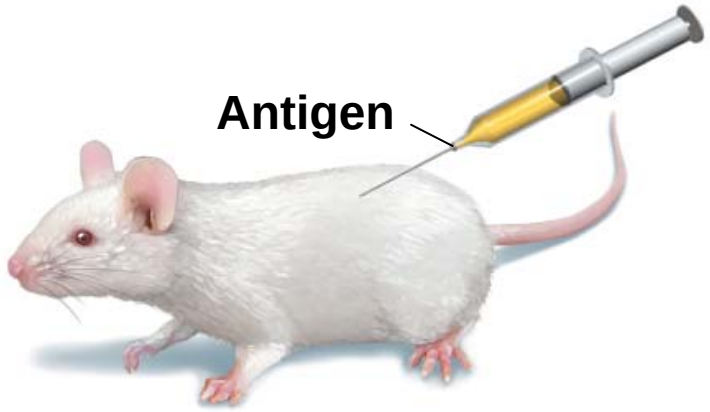
- Therapeutic index // Risk-versus-benefit calculation // note – even asprin has a risk factor (more dangerous than any vaccine)
- 1999 // vaccine to prevent infant diarrhea caused by rotaviruses was withdrawn because in some cases it caused life-threatening intestinal obstructions
- Rumors started in England about incorrect correlation between the MMR vaccine and autism led to many parents not immunizing their children
- Rumors have been discredited // autism is a genetic disease which starts during fetal life but exhibits symptoms at same age when MMR vaccine given
- Failure to immunize children has broken down the herd immunity and now diseases like measles are once again becoming a problem

Monoclonal Antibodies (Mabs)

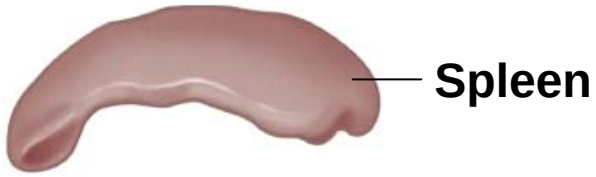
- **Hybridoma**: “immortal” cancerous B cell fused with an antibody-producing normal B cell
  - Produces **monoclonal antibodies**
  - **Used for treating and diagnosing disease**
  - highly specific and they can be produced in large quantities.

The Production of Monoclonal Antibodies.

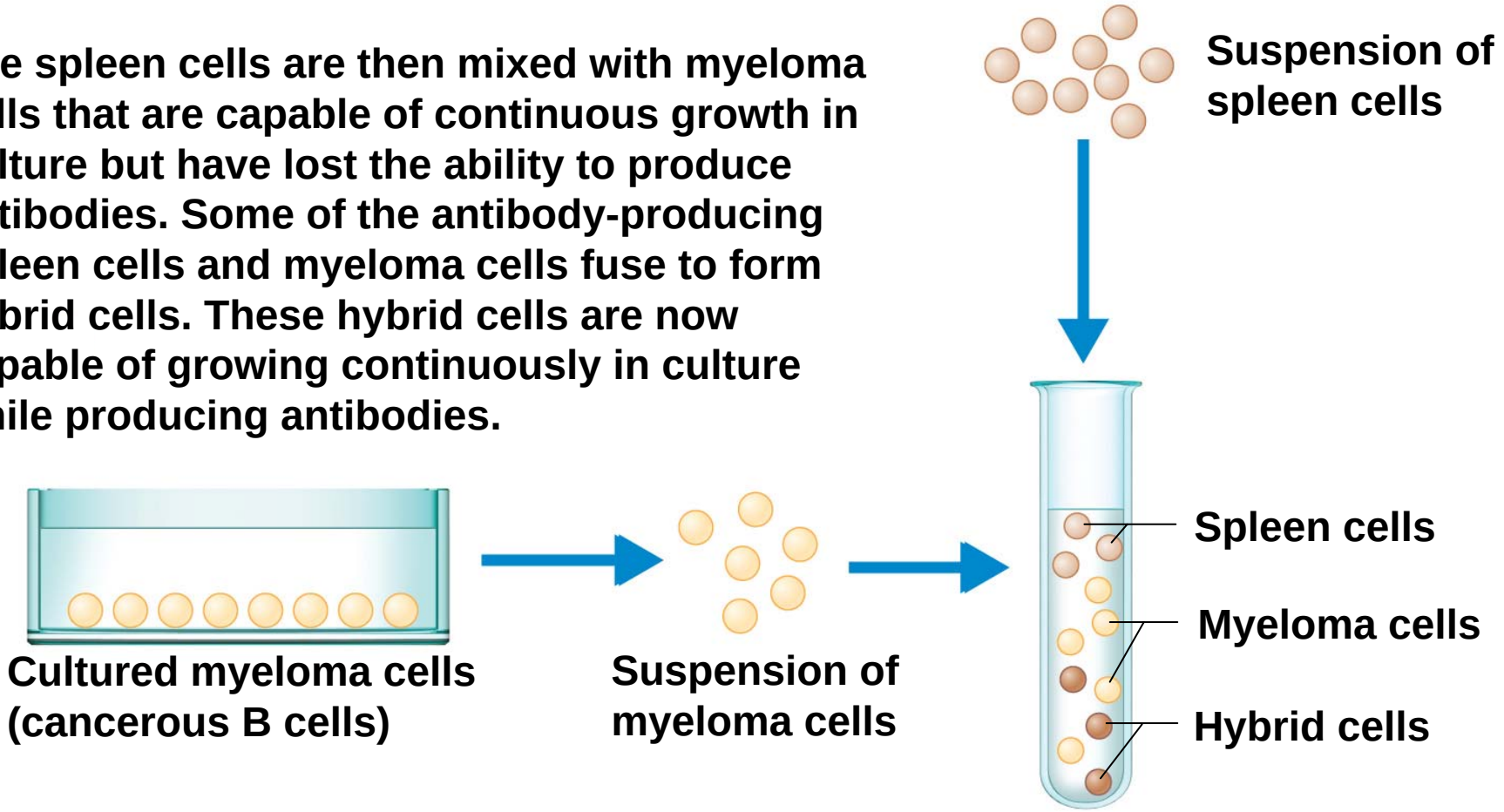
1. **A mouse is injected with a specific antigen that will induce production of antibodies against that antigen.**



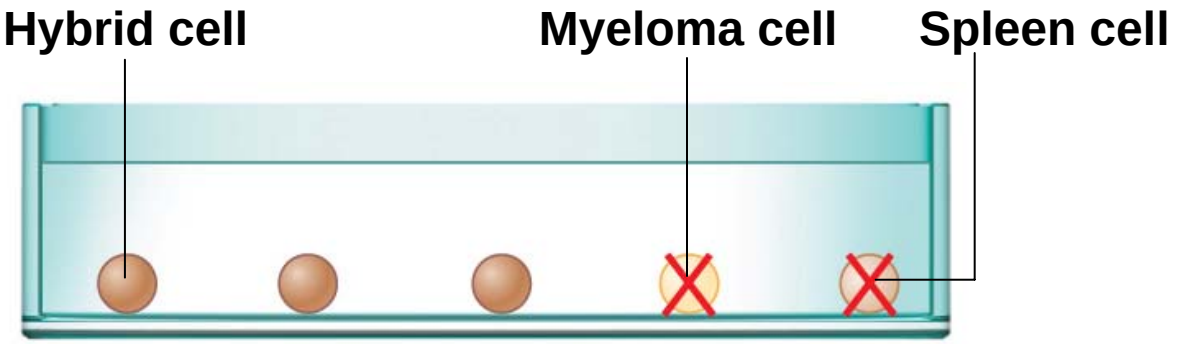
2. **The spleen of the mouse is removed and homogenized into a cell suspension. The suspension includes B cells that produce antibodies against the injected antigen.**



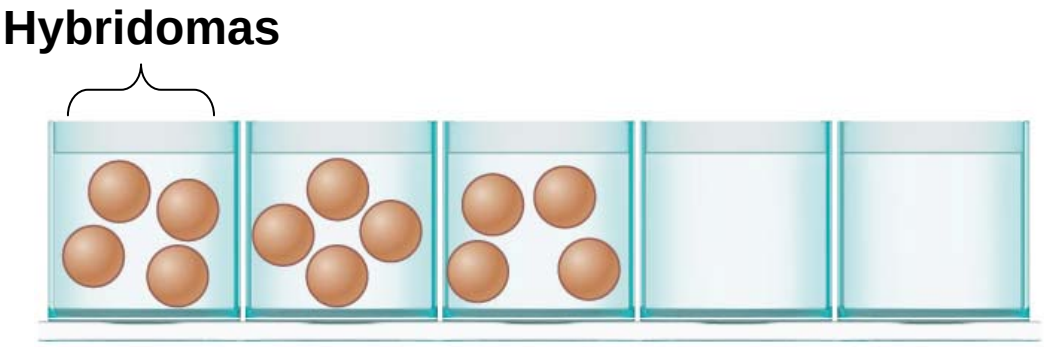
3. The spleen cells are then mixed with myeloma cells that are capable of continuous growth in culture but have lost the ability to produce antibodies. Some of the antibody-producing spleen cells and myeloma cells fuse to form hybrid cells. These hybrid cells are now capable of growing continuously in culture while producing antibodies.



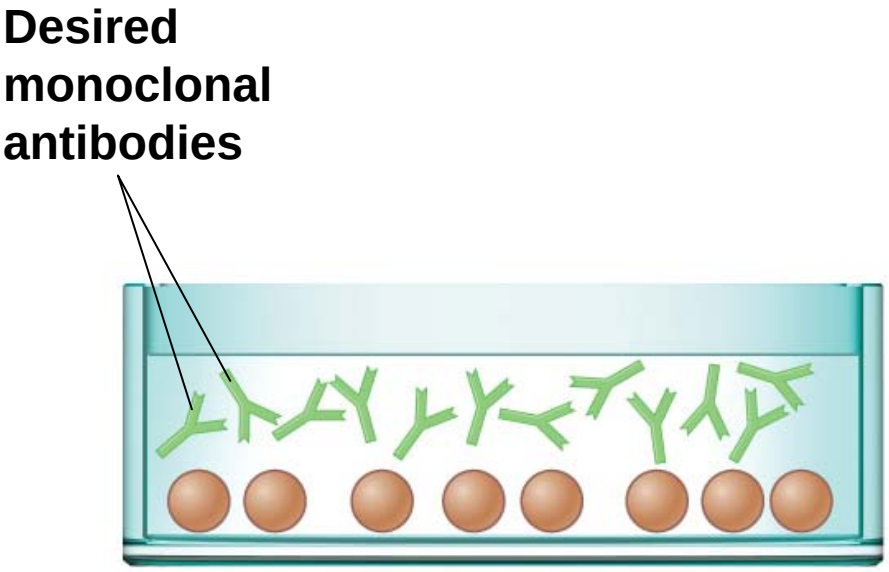
4. The mixture of cells is placed in a selective medium that allows only hybrid cells to grow.



5. **Hybrid cells proliferate into clones called hybridomas. The hybridomas are screened for production of the desired antibody.**



6. **The selected hybridomas are then cultured to produce large quantities of monoclonal antibodies. Isolated antibodies are used for treating and diagnosing disease.**



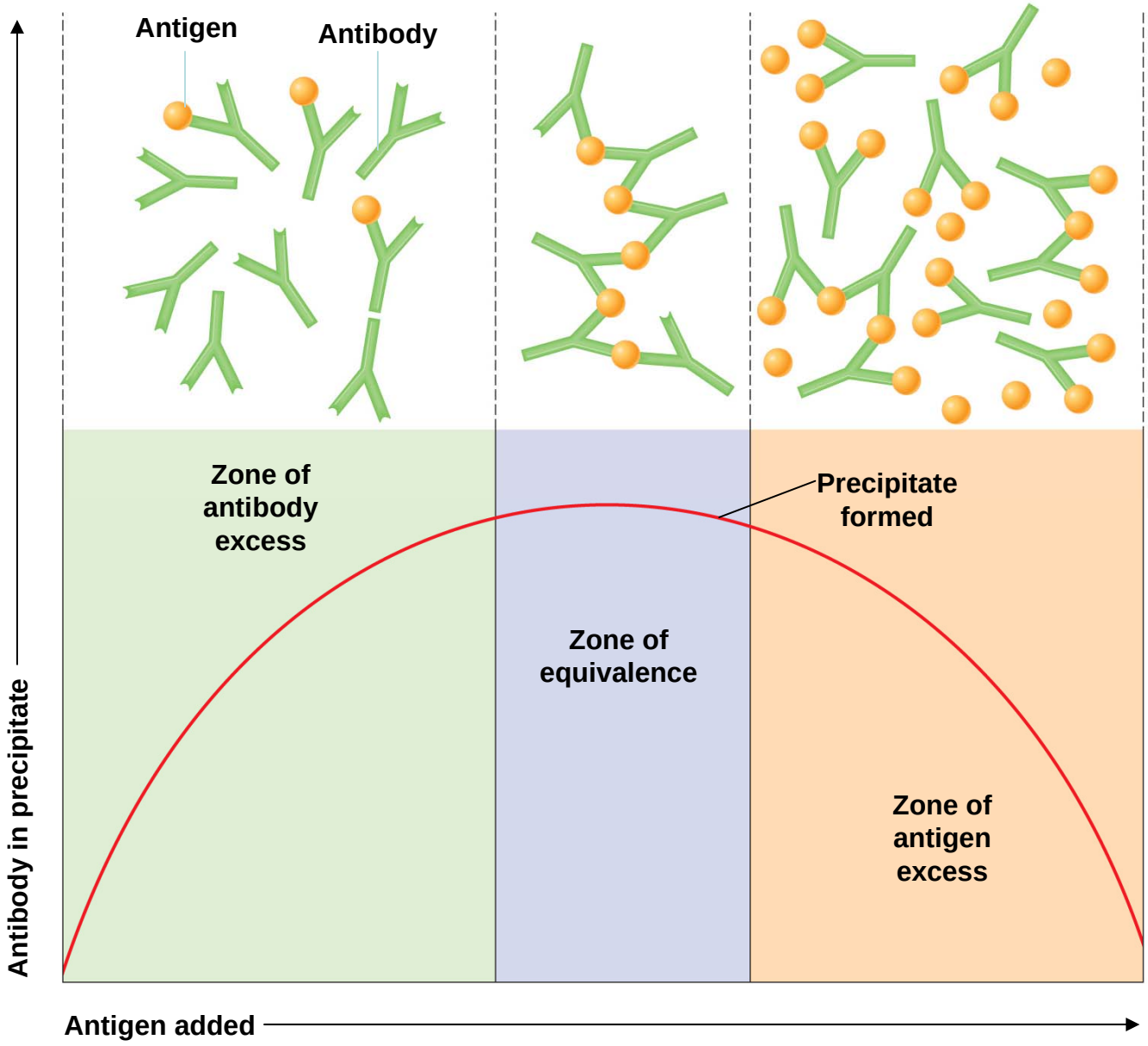
Monoclonal Antibodies (Mabs)

- Muromo**nab**-CD3: for kidney transplant
- Inflixi**mab**: for Crohn's disease
- Comalizu**mab**: for allergic asthma
- Rituxi**mab**: rheumatoid arthritis
- Trastuzu**mab**: Herceptin for breast cancer

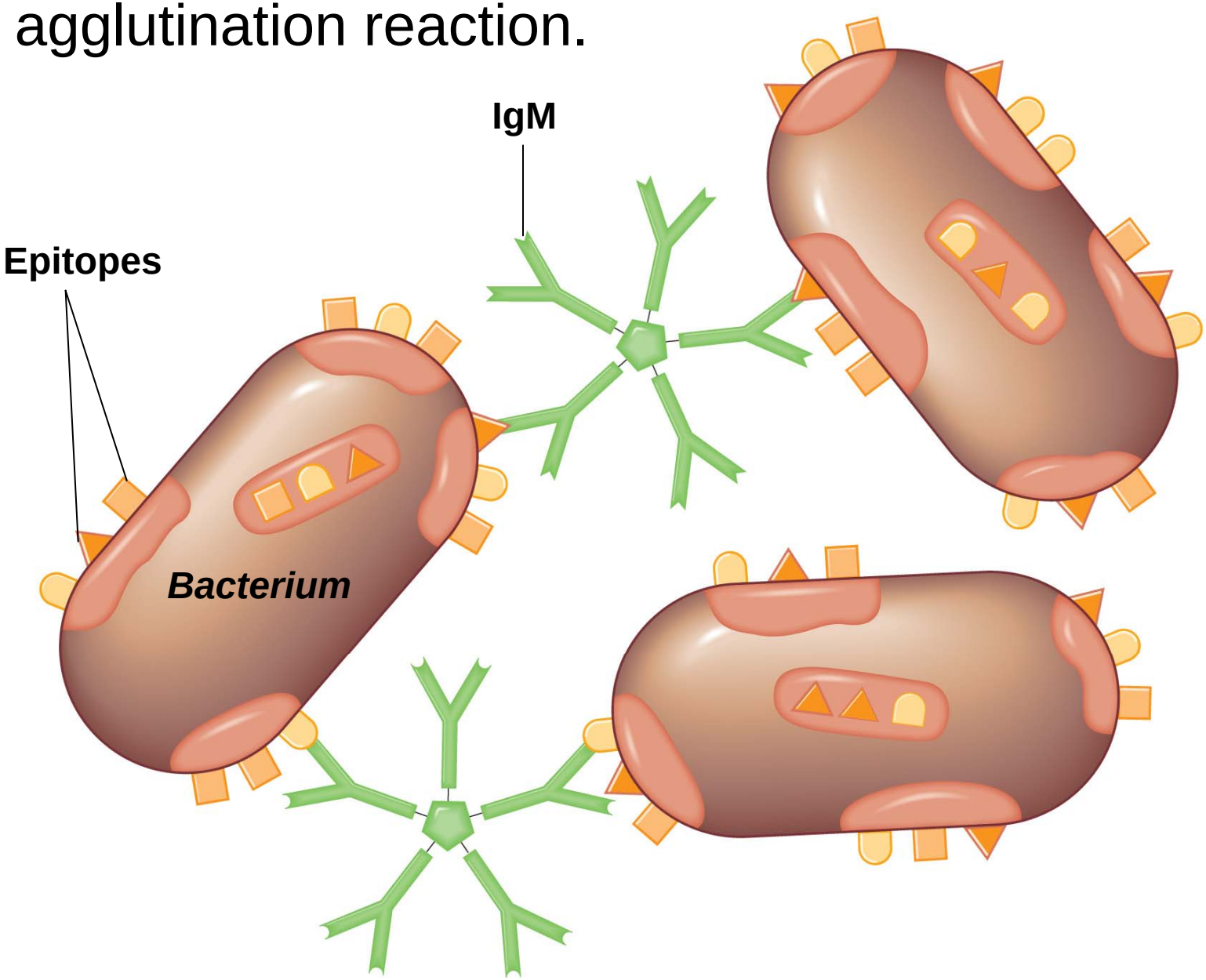
Serological Tests

- **Precipitation**: soluble antigens
- **Agglutination**: particulate antigens
- **Hemagglutination**: agglutination of RBCs
- **Neutralization**: inactivates toxin or virus
- **Fluorescent-antibody technique**: antibodies linked to fluorescent dye
- **Complement fixation**: RBCs are indicator
- **ELISA**: peroxidase enzyme is the indicator

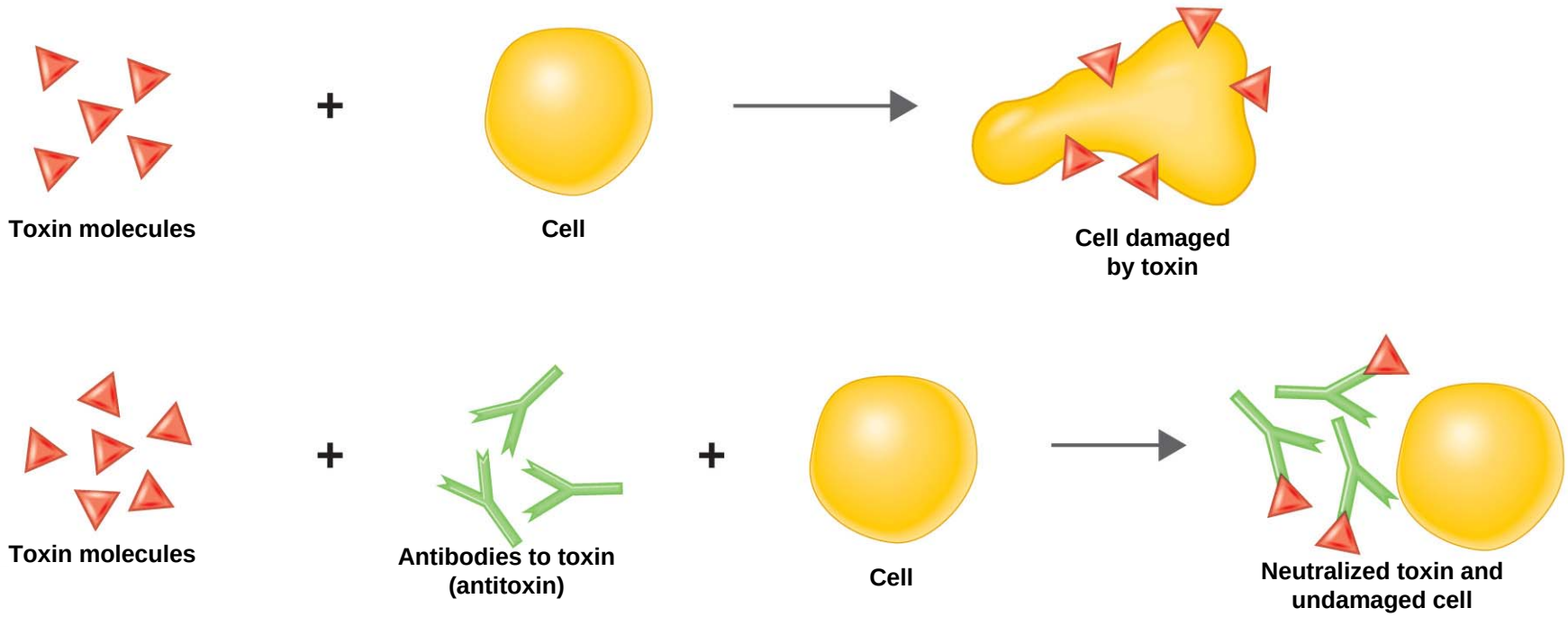
Precipitation Curve.



An agglutination reaction.

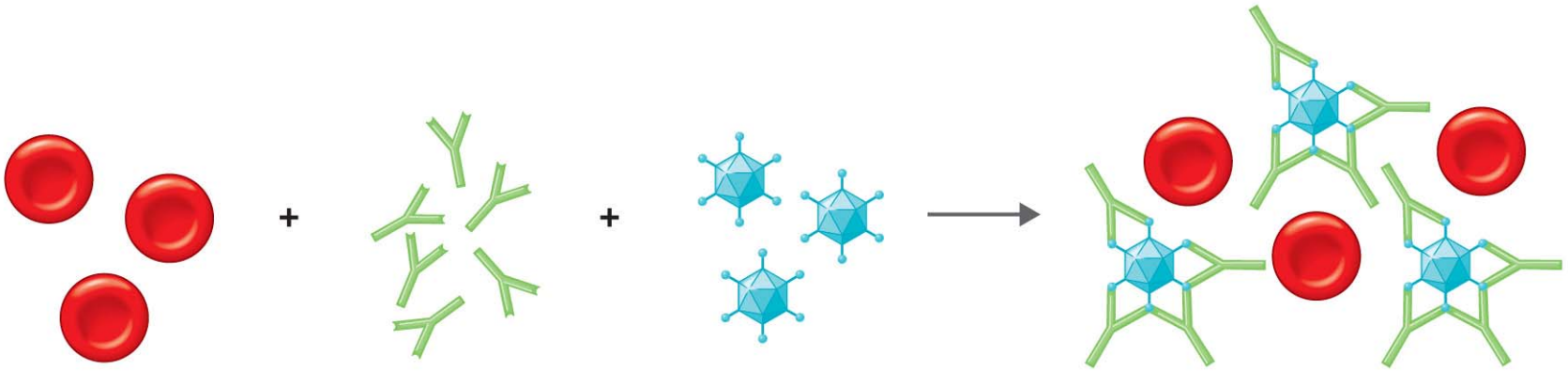


Reactions in neutralization tests.



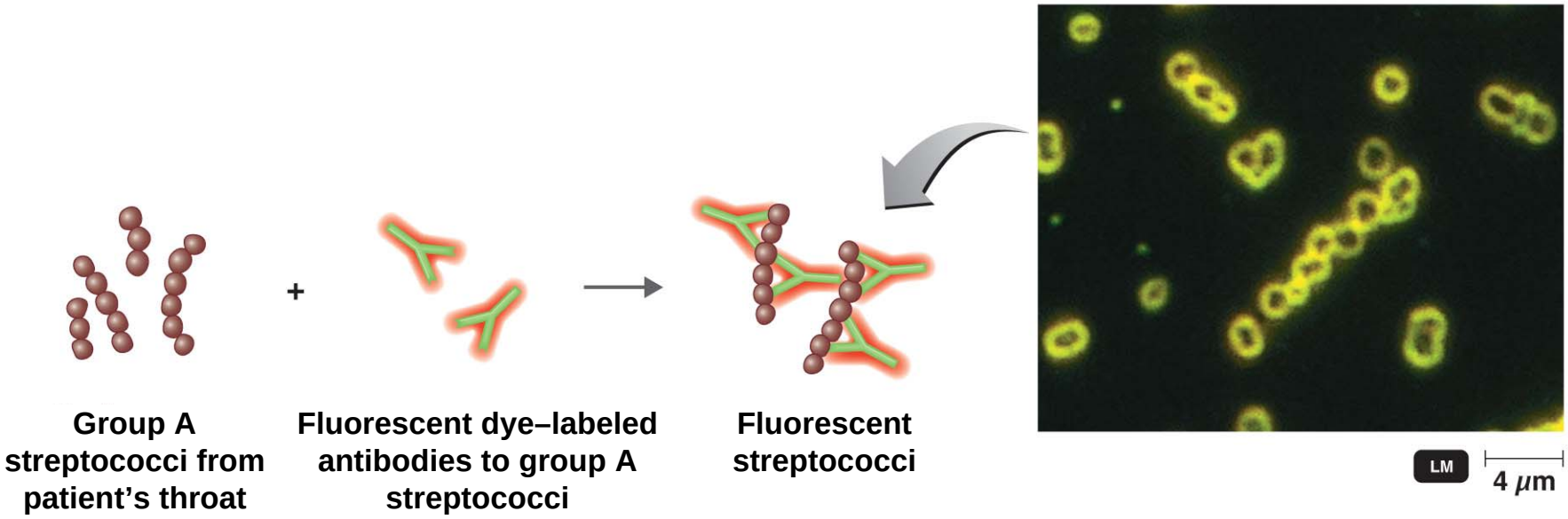
(a) The effects of a toxin on a susceptible cell and neutralization of the toxin by antitoxin

Reactions in neutralization tests.



(b) **Viral hemagglutination test to detect antibodies to a virus. These viruses will normally cause hemagglutination when mixed with red blood cells. If antibodies to the virus are present, as shown here, they neutralize and inhibit hemagglutination.**

Fluorescent-antibody (FA) techniques.



(a) **Reactions in a positive direct fluorescent-antibody test**