Antibiotics

What are antibiotics?
Who are the main producers?
Biological functions?
Resistance
New developments

First antimicrobial drugs

Louis Pasteur (1822-1895):
"pasteurization"
Fermentation: wine contamination
Germ theory: silkworn disease
Vaccine: anthrax, fowl cholera
Rabies

First antimicrobial drugs

Paul Ehrilch (1854-1915):

- Methylene blue: malaria

-Toxin and antitoxin

-Salvarsan: magic bullet against syphilis, *Treponema pallidum*

First antimicrobial drugs

- Gerhard Domagk (Nobel Prize 1939)
  - Sulfa drugs
  - Prontosil
  - Sulfanilamide, analog of p-aminobenzoic acid
  - (part of folic acid, precursor of nucleic acids)
  - Development of antituberculosis compounds thiosemicarbasone and isoniazid

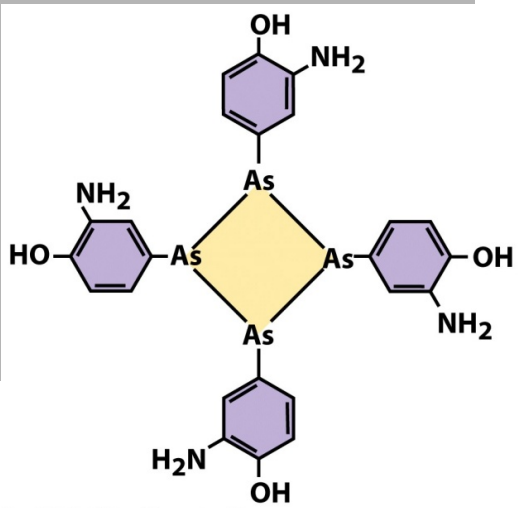
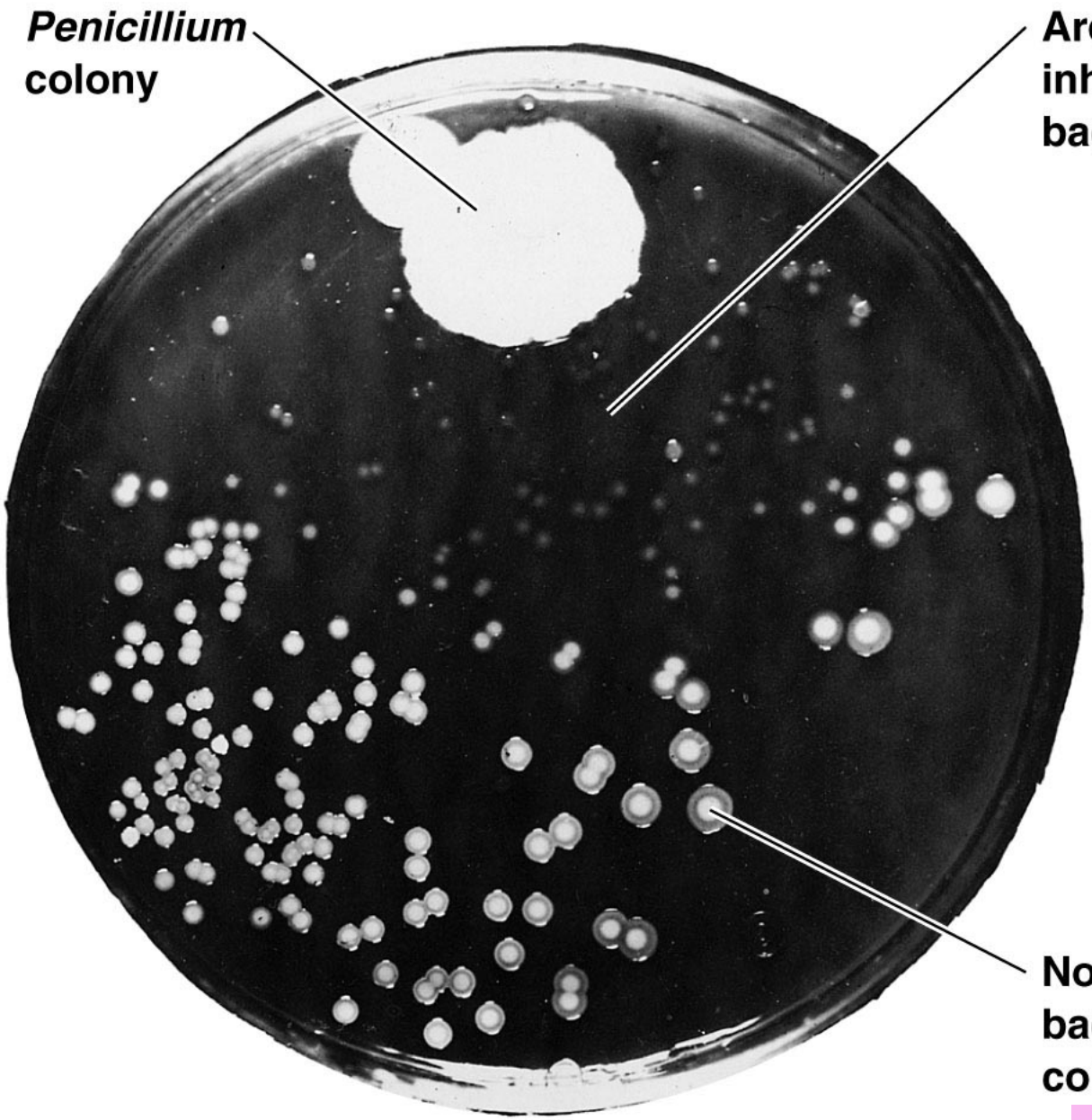


Figure 20-12 Brock Biology of Microorganisms 11/e



1928 Alexander Fleming

1940 Howard Florey , Ernst Chain

1944 Salman Waksman- streptomycin

Staphylococcus aureus

Figure 20.1

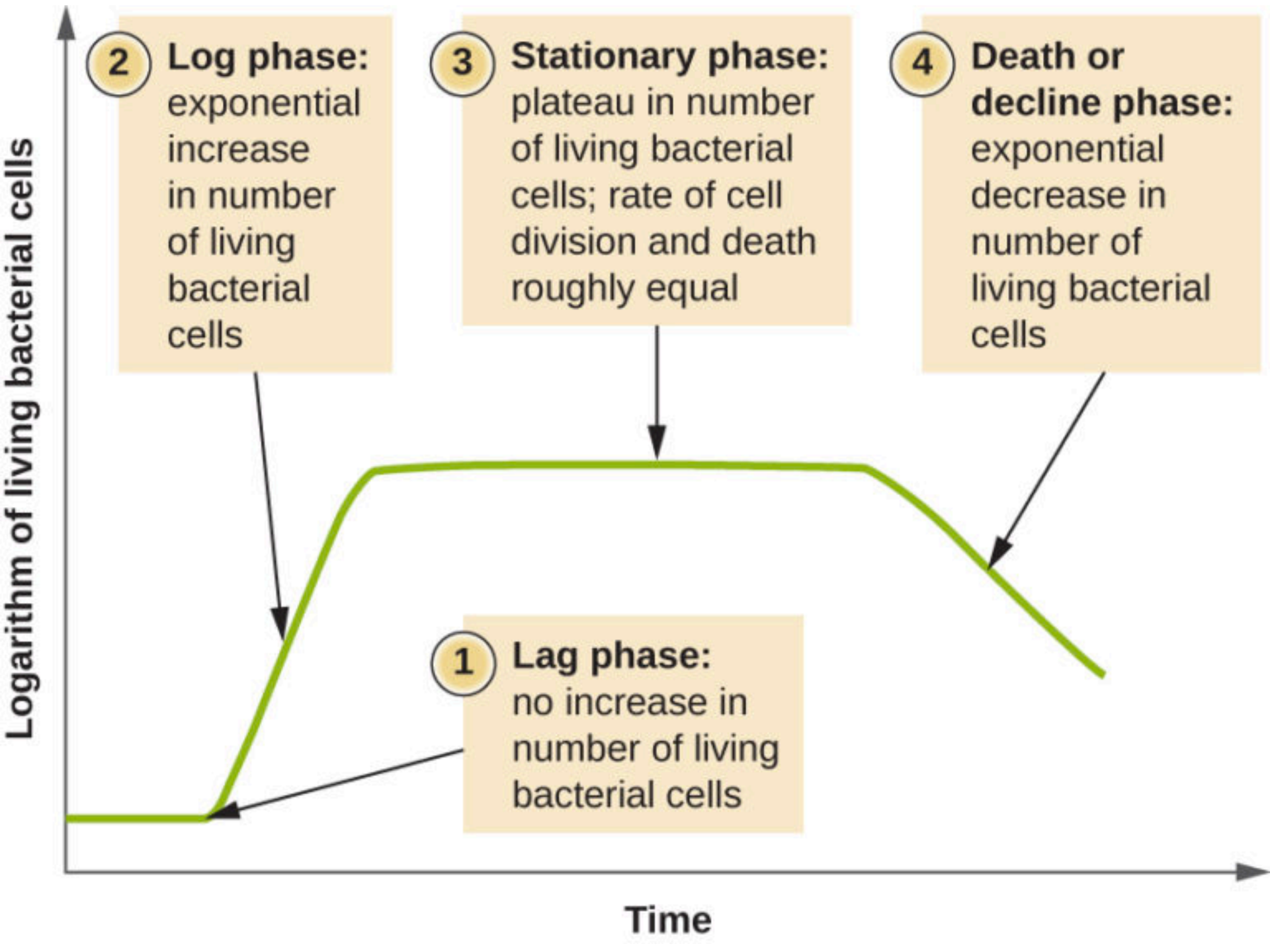
What are antibiotics?

- Any compund able to kill a target cell
- Secondary metabolites synthesized by some microorganisms
- Biological activity

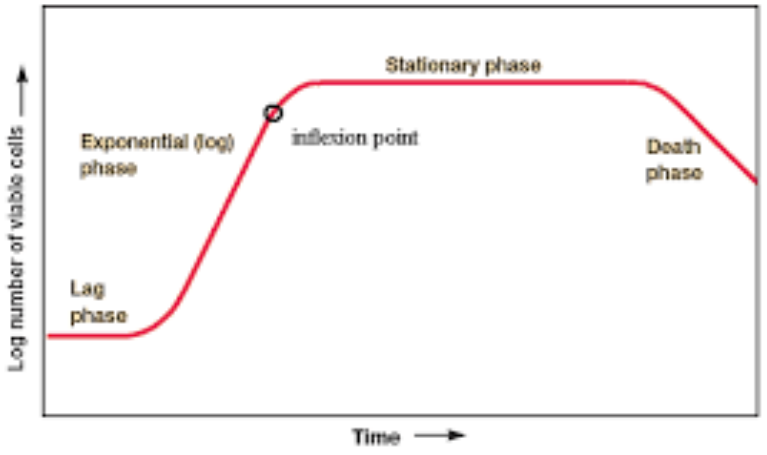
Primary and Secondary Metabolism

Difference between primary and secondary metabolites

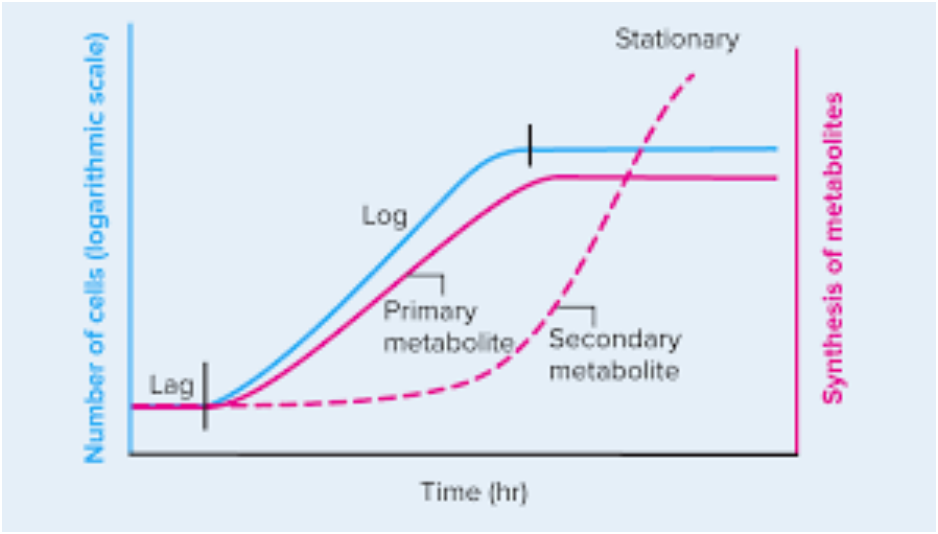
Primary metabolites	Secondary metabolites
They are involved in normal growth, development and reproduction.	They are not directly involved in the normal growth, development and reproduction.
Examples for primary metabolites are carbohydrates , fats and proteins.	Examples for secondary metabolites are alkaloids, tannis, resins, gums and latex etc.
They are not poisonous.	Some of these compounds are poisonous.


Logarithm of living bacterial cells
Time
1 Lag phase: no increase in number of living bacterial cells
2 Log phase: exponential increase in number of living bacterial cells
3 Stationary phase: plateau in number of living bacterial cells; rate of cell division and death roughly equal
4 Death or decline phase: exponential decrease in number of living bacterial cells

Primary and secondary metabolism



Microbial Growth Curve in a Closed System.



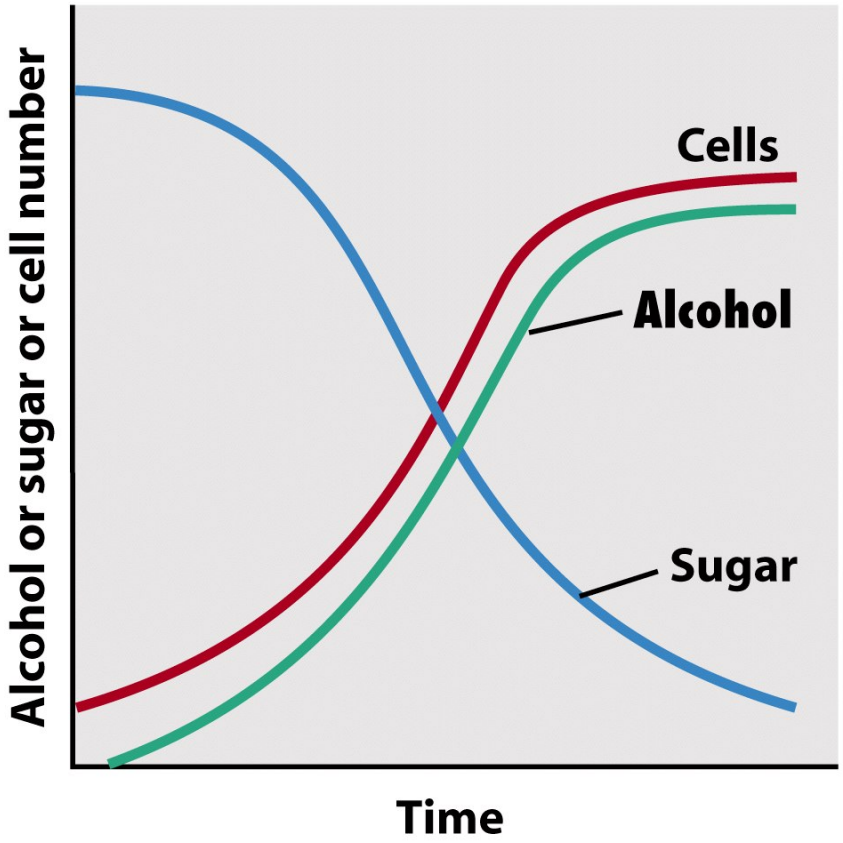


Figure 30-2a Brock Biology of Microorganisms 11/e

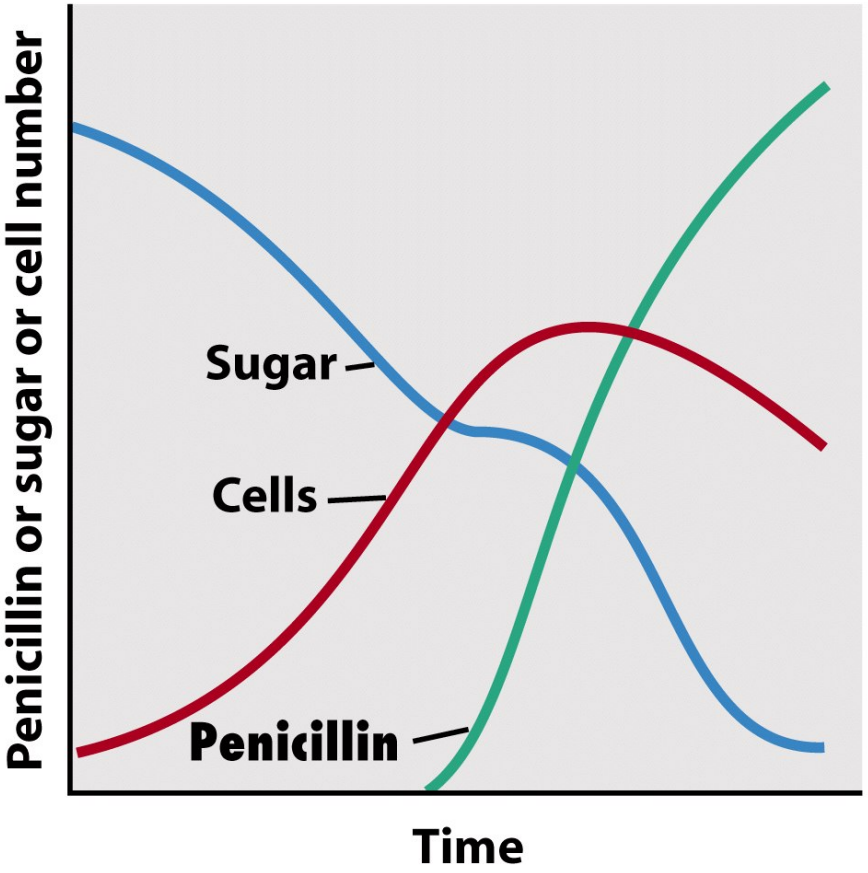


Figure 30-2b Brock Biology of Microorganisms 11/e

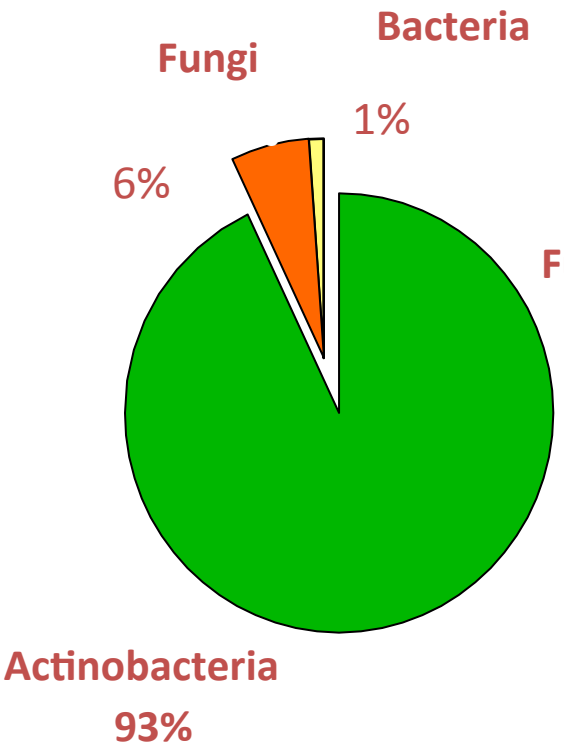
Primary and secondary metabolism

Who are the main producers

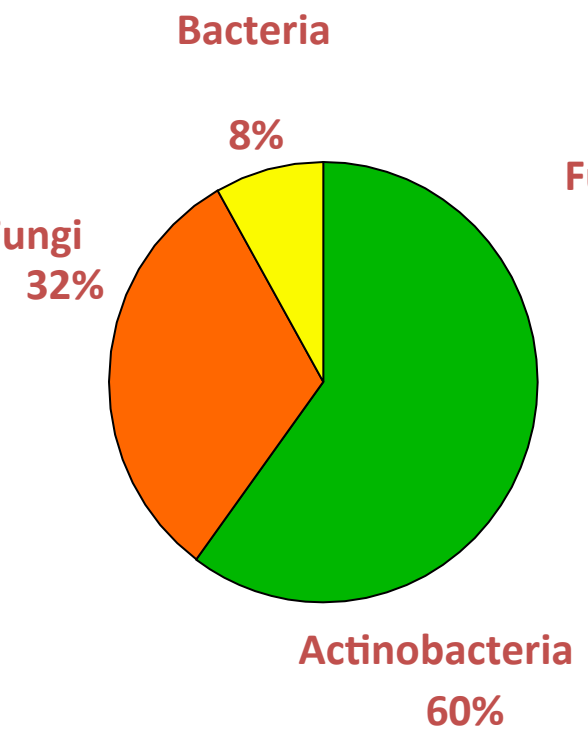
- Bacteria

  Gram positive *Streptomyces*
- Fungi
- Other bacteria

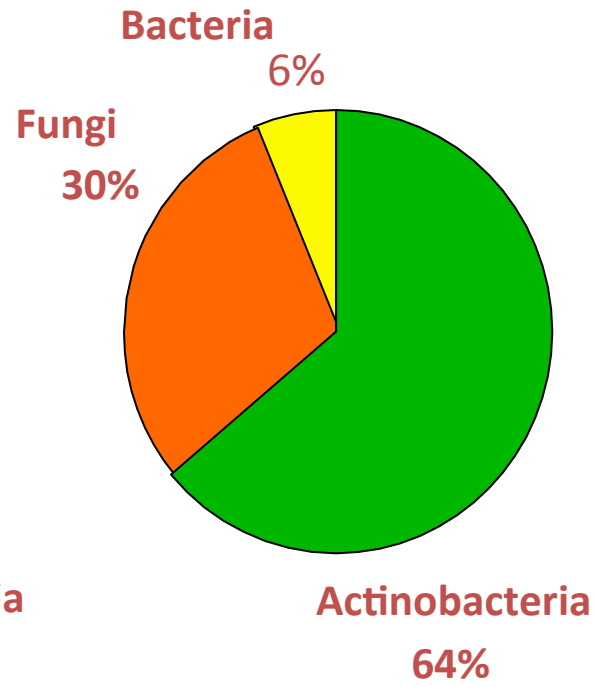
MICROORGANISMS and BIOACTIVE COMPOUNDS



Antitumorals



Antifungals



Bioactives

Biological functions of antibiotics?

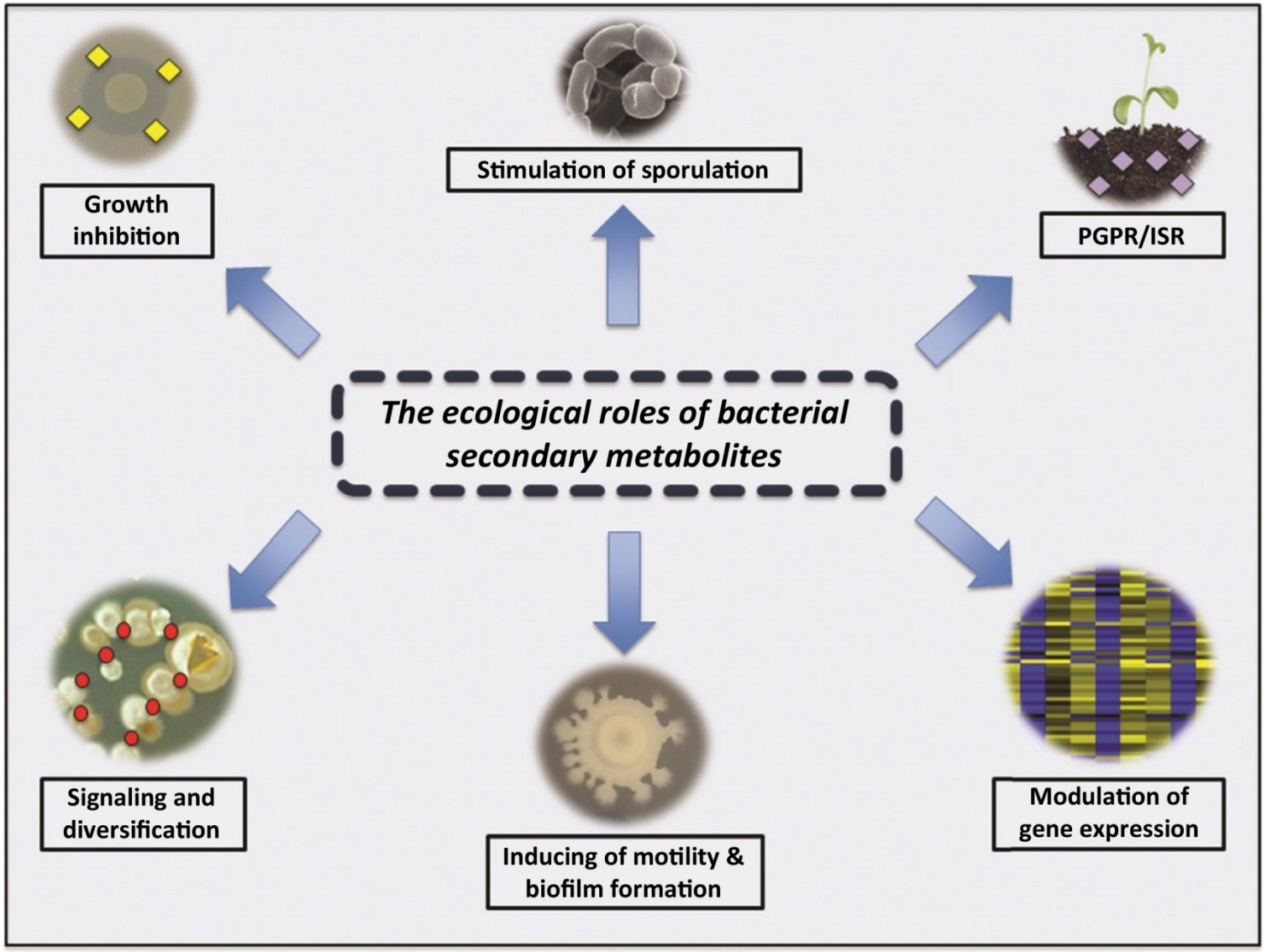
- In the producer:

  Activators of morphological differentiation, UV protector, communication

- In the target microorganism:

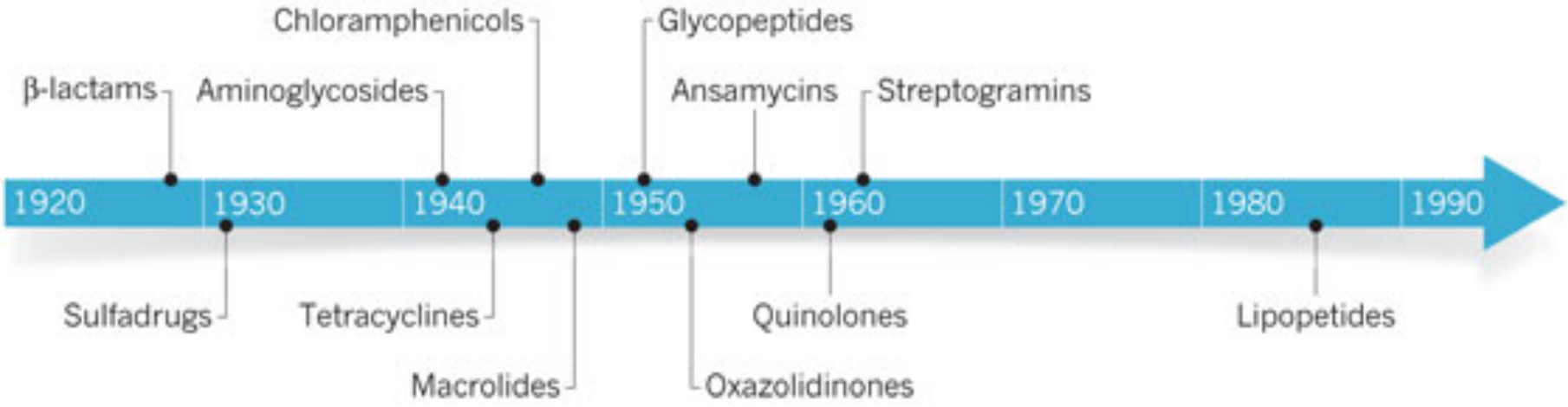
  Toxicity

Different Ecological Roles of Bacterial Secondary Metabolites in Nature



ANTIBIOTIC DISCOVERY TIMELINE

Decades without identifying antibiotics that go on to be used for the treatment of patients has put our defence against bacteria at risk. This timeline pinpoints the year that the antibiotics were first discovered.



Diminishing returns in finding natural products: Genetics to the rescue?

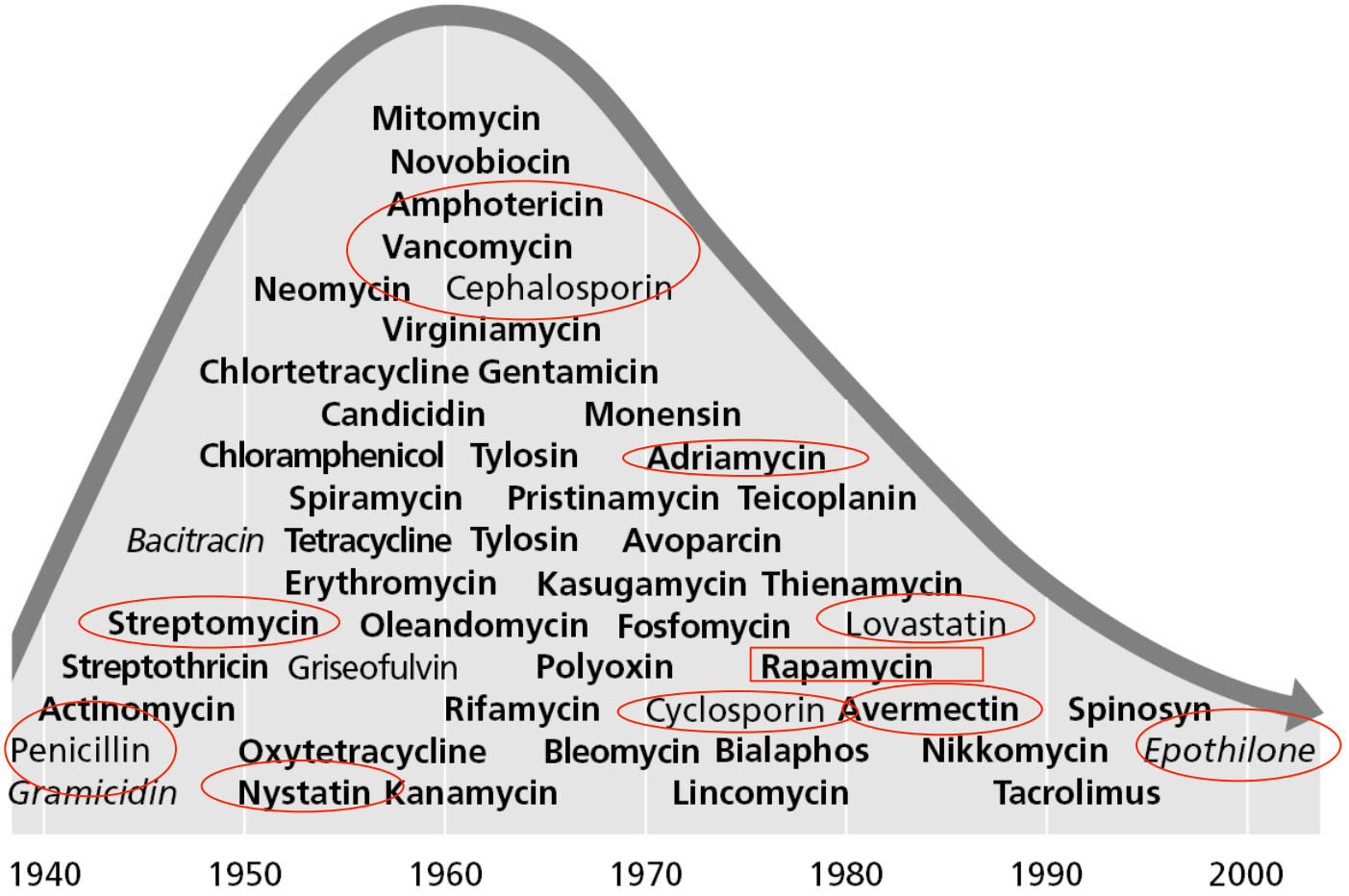


TABLE 20.1 Representative Sources of Antibiotics

Microorganism	Antibiotic
Gram-Positive Rods	
Bacillus subtilis	Bacitracin
Paenibacillus polymyxa	Polymyxin
Actinomycetes	
Streptomyces nodosus	Amphotericin B
Streptomyces venezuelae	Chloramphenicol
Streptomyces aureofaciens	Chlortetracycline and tetracycline
Saccharopolyspora erythraea	Erythromycin
Streptomyces fradiae	Neomycin
Streptomyces griseus	Streptomycin
Micromonospora purpurea	Gentamicin
Fungi	
Cephalosporium spp.	Cephalothin
Penicillium griseofulvum	Griseofulvin
Penicillium chrysogenum	Penicillin

BIOACTIVE COMPOUNDS SYNTHESIZED BY ACTINOBACTERIA

ANTIBACTERIALS

Erythromycin
Tetracycline
Gentamicin

ANTIFUNGALS

Amphotericin B
Nystatin

ANTIPARASITICS

Avermectins

ANTITUMORALS

Doxorubicin
Mitramycin
Bleomycin

IMUNOSUPRESSANTS

Rapamycin
FK506/Tacrolimus

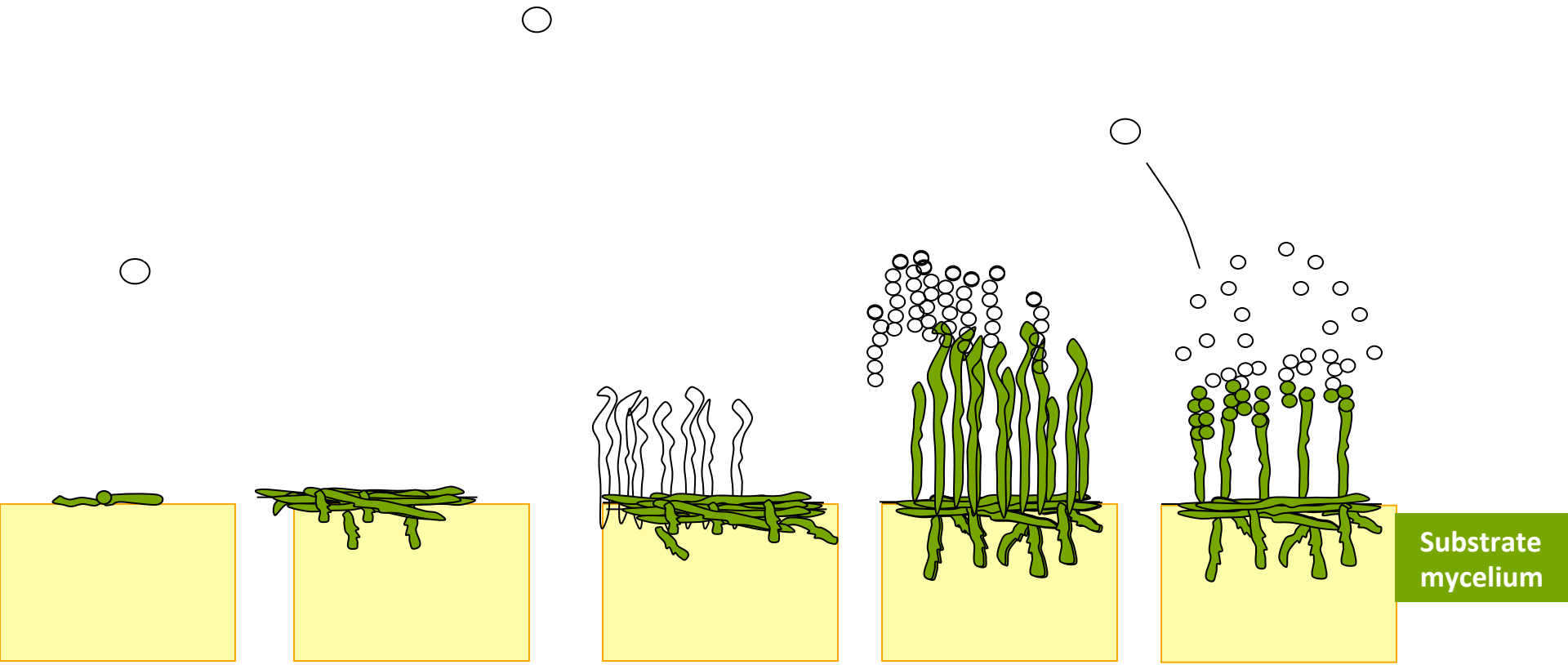
INSETICIDES

Espinosin

HERBICIDES

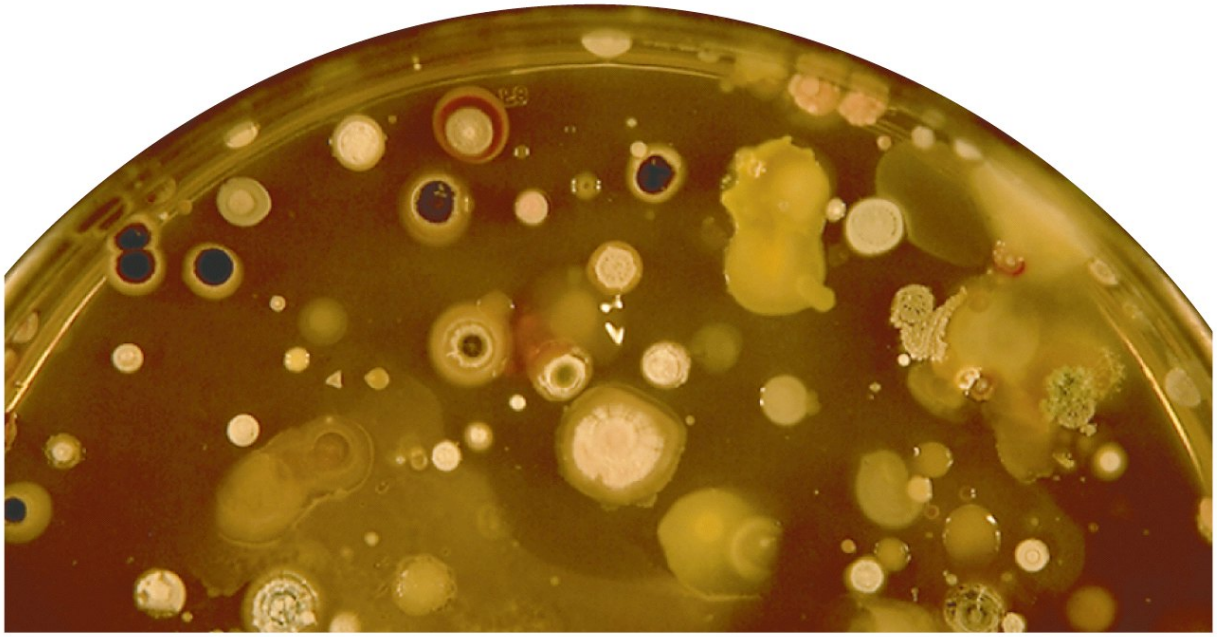
Bialaphos

LIFE CYCLE OF *Streptomyces*



Production of secondary metabolites

(antibiotics, fungicides, antitumorals,..)

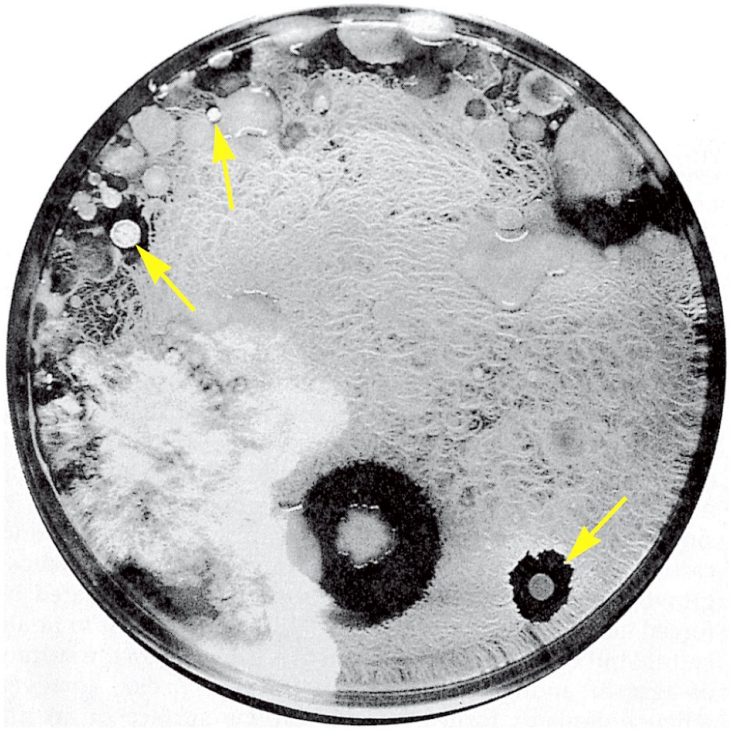


M. T. Madigan

Figure 12-76a Brock Biology of Microorganisms 11/e

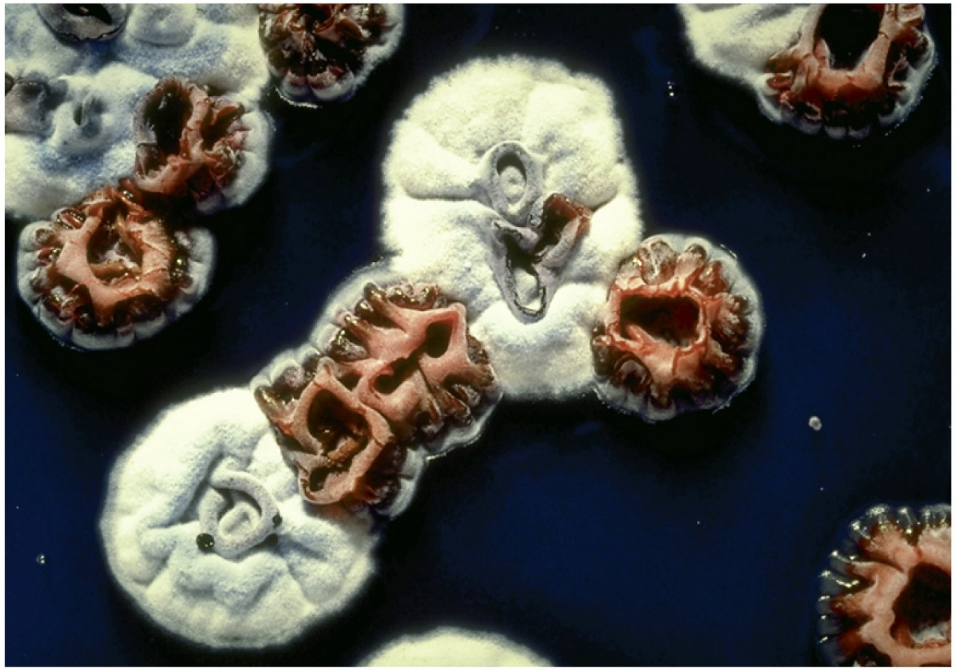
David A. Hopwood

Figure 12-76b Brock Biology of Microorganisms 11/e



© Eli Lilly & Co. Used with permission.

Figure 12-77a Brock Biology of Microorganisms 11/e
© 2006 Pearson Prentice Hall, Inc.



David A. Hopwood

Figure 12-77b Brock Biology of Microorganisms 11/e
© 2006 Pearson Prentice Hall, Inc.

Antibiotics classifications

Origin: natural, semisynthetic, syntethic

Chemical structures (11 groups)

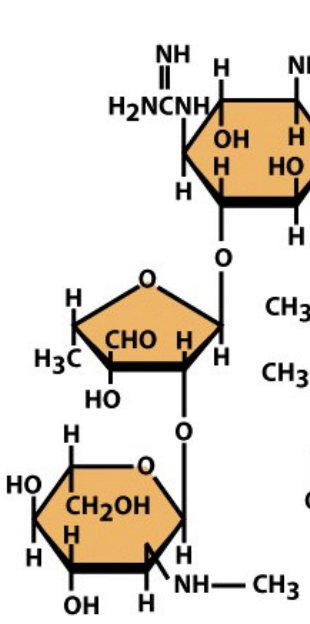
Biological Activity

Spectrum of Activity

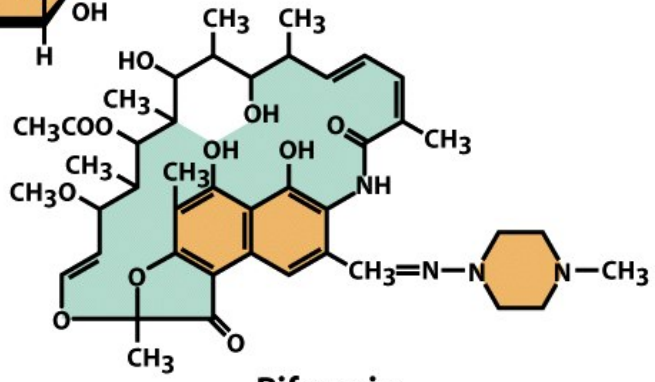
Biological Target

Antibiotic classification		Subclassification	Example
I.	Carbohydrate-containing compounds	Pure sugars	Nojirimycin
		Aminoglycosides	Streptomycin
		Orthosomycins	Everninomicin
		N-Glycosides	Streptothricin
		C-Glycosides	Vancomycin
		Glycolipids	Moenomycin
II.	Macrocyclic lactones	Macrolide antibiotics	Erythromycin
		Polyene antibiotics	Candicidin
		Ansamycins	Rifampin
		Macrotetrolides	Tetranactin
III.	Quinones and related compounds	Tetracyclines	Tetracycline
		Anthracyclines	Adriamycin
		Naphthoquinones	Actinorhodin
		Benzoquinones	Mitomycin

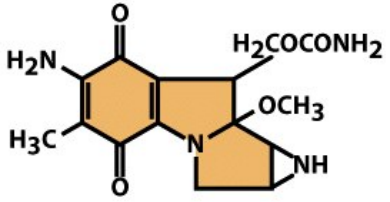
Representative structure



Streptomycin



Rifampin



Mitomycin C

Figure 20-13 part 1 Brock Biology of Microorganisms 11/e

Antibiotic classification		Subclassification	Example
IV.	Amino acid and peptide analogs	Amino acid derivatives	Cycloserine
		β-Lactam antibiotics	Penicillin, ceftriaxone
		Peptide antibiotics	Bacitracin
		Chromopeptides	Actinomycin
		Depsipeptides	Valinomycin
		Chelate-forming peptides	Bleomycin
V.	Heterocyclic compounds containing nitrogen	Nucleoside antibiotics	Polyoxins
VI.	Heterocyclic compounds containing oxygen	Polyether antibiotics	Monensin

Representative structure

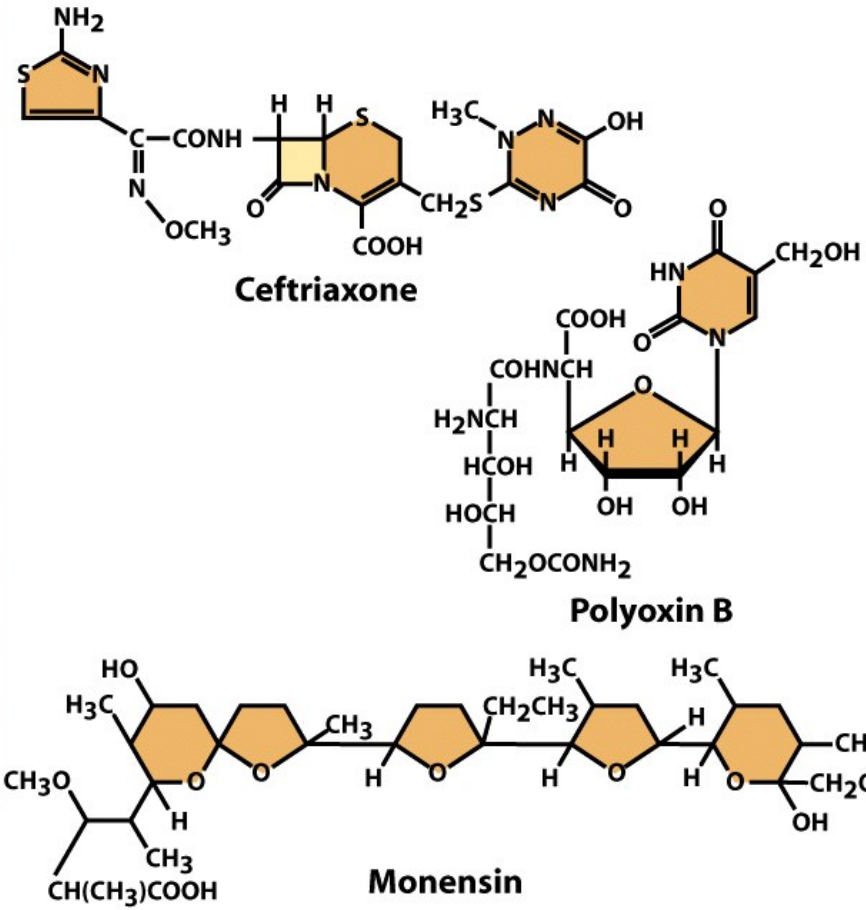


Figure 20-13 part 2 Brock Biology of Microorganisms 11/e

Antibiotic classification	Subclassification	Example
VII. Alicyclic derivatives	Cycloalkane derivatives	Cycloheximide
	Steroid antibiotics	Fusidic acid
VIII. Aromatic compounds	Benzene derivatives	Chloramphenicol
	Condensed aromatics	Griseofulvin
	Aromatic ether	Novobiocin
IX. Aliphatic compounds	Compounds containing phosphorus	Fosfomycin

Representative structure

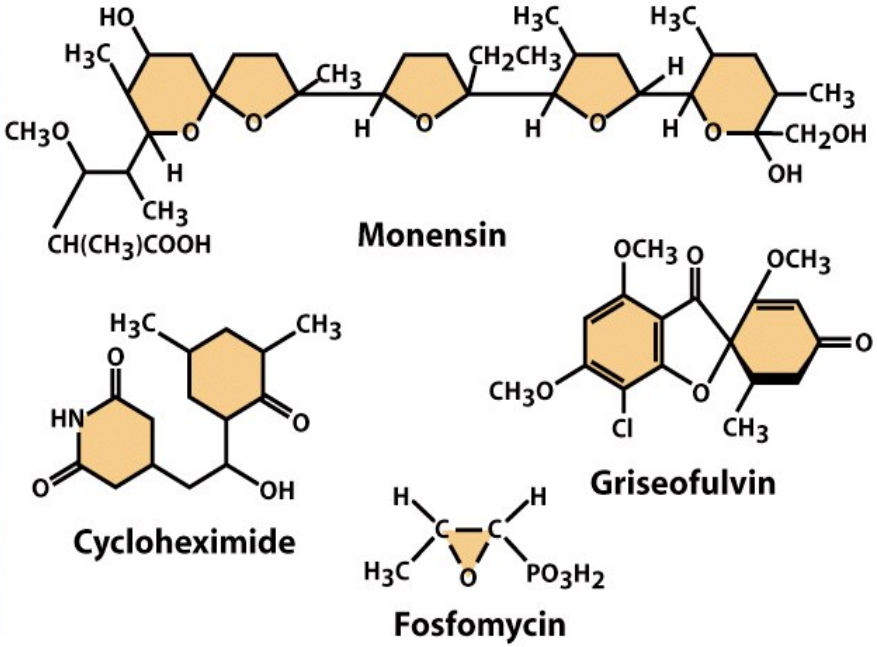
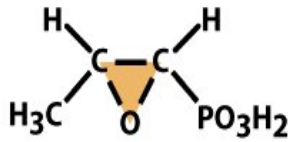
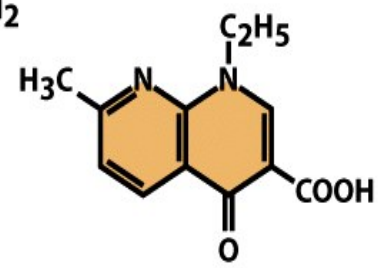


Figure 20-13 part 3 Brock Biology of Microorganisms 11/e

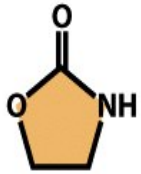
Antibiotic classification		Subclassification	Example	Representative structure
X.	Quinolone compounds	4-Quinolone Fluoro-4-quinolones	Nalidixic acid Ciprofloxacin	
XI.	Oxazolidinone	Cyclic lactone	2-Oxazolidinone	



Fosfomycin



Nalidixic acid



2-Oxazolidinone

Figure 20-13 part 4 Brock Biology of Microorganisms 11/e

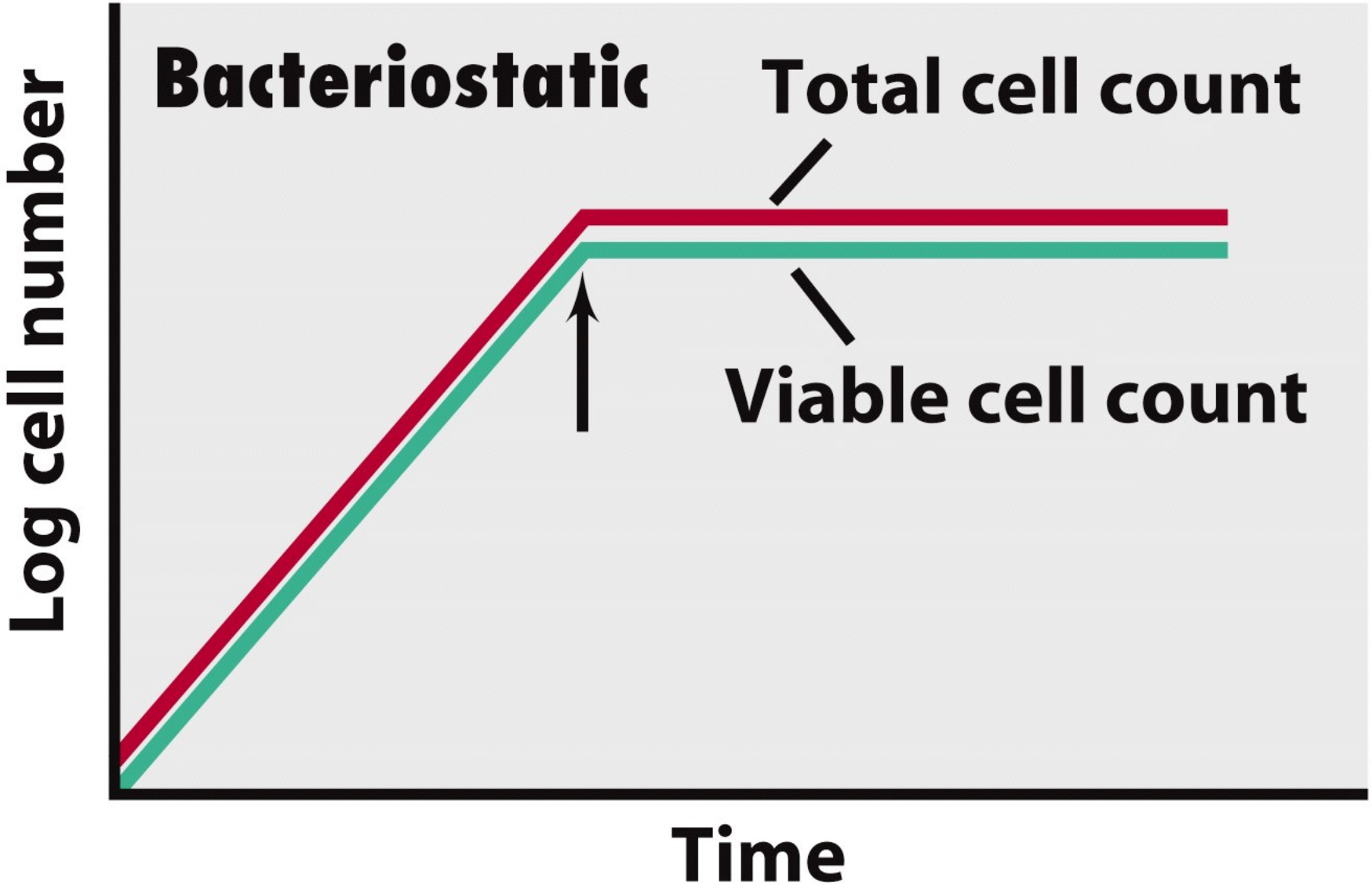


Figure 20-9a Brock Biology of Microorganisms 11/e

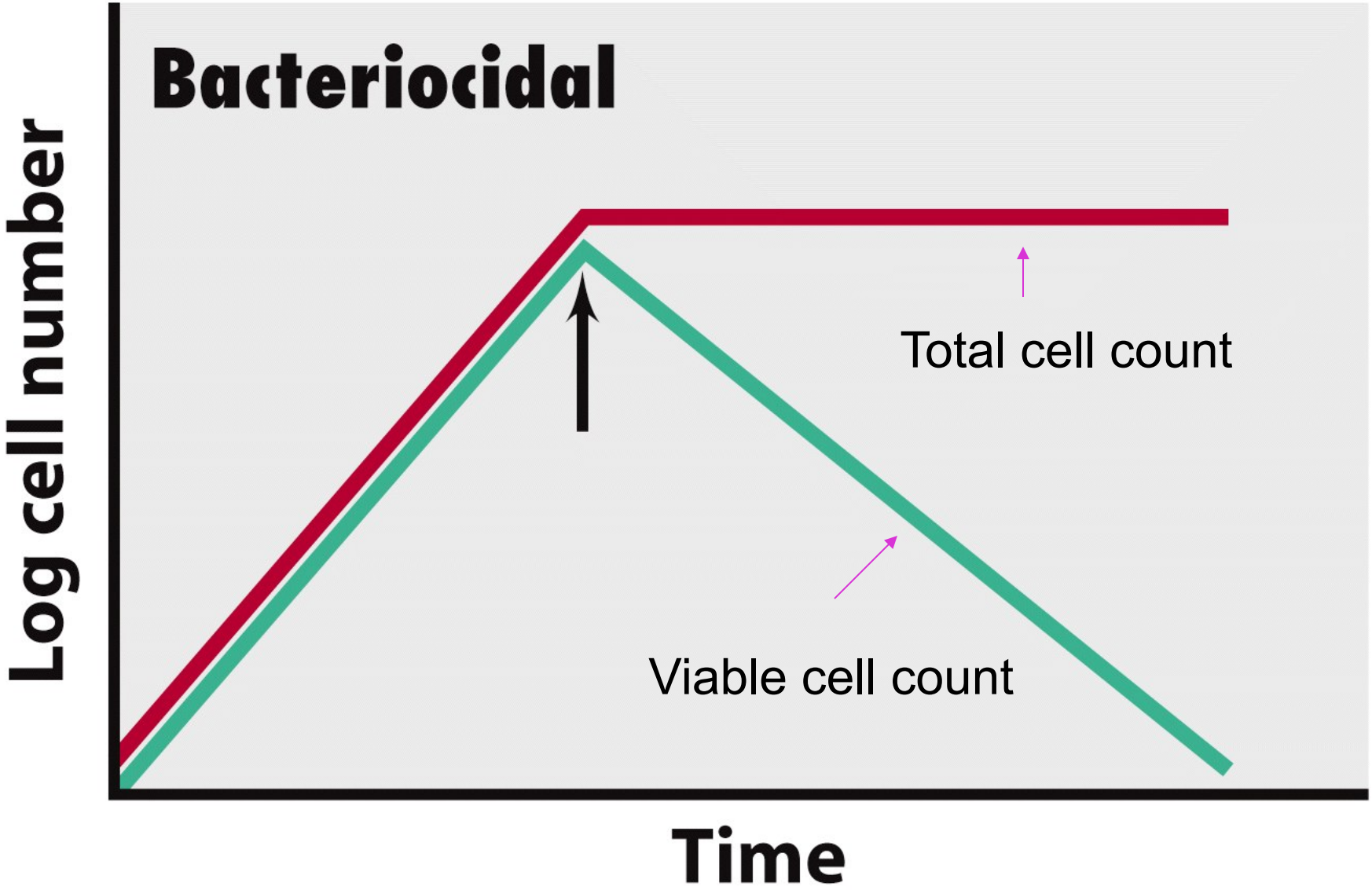


Figure 20-9b Brock Biology of Microorganisms 11/e

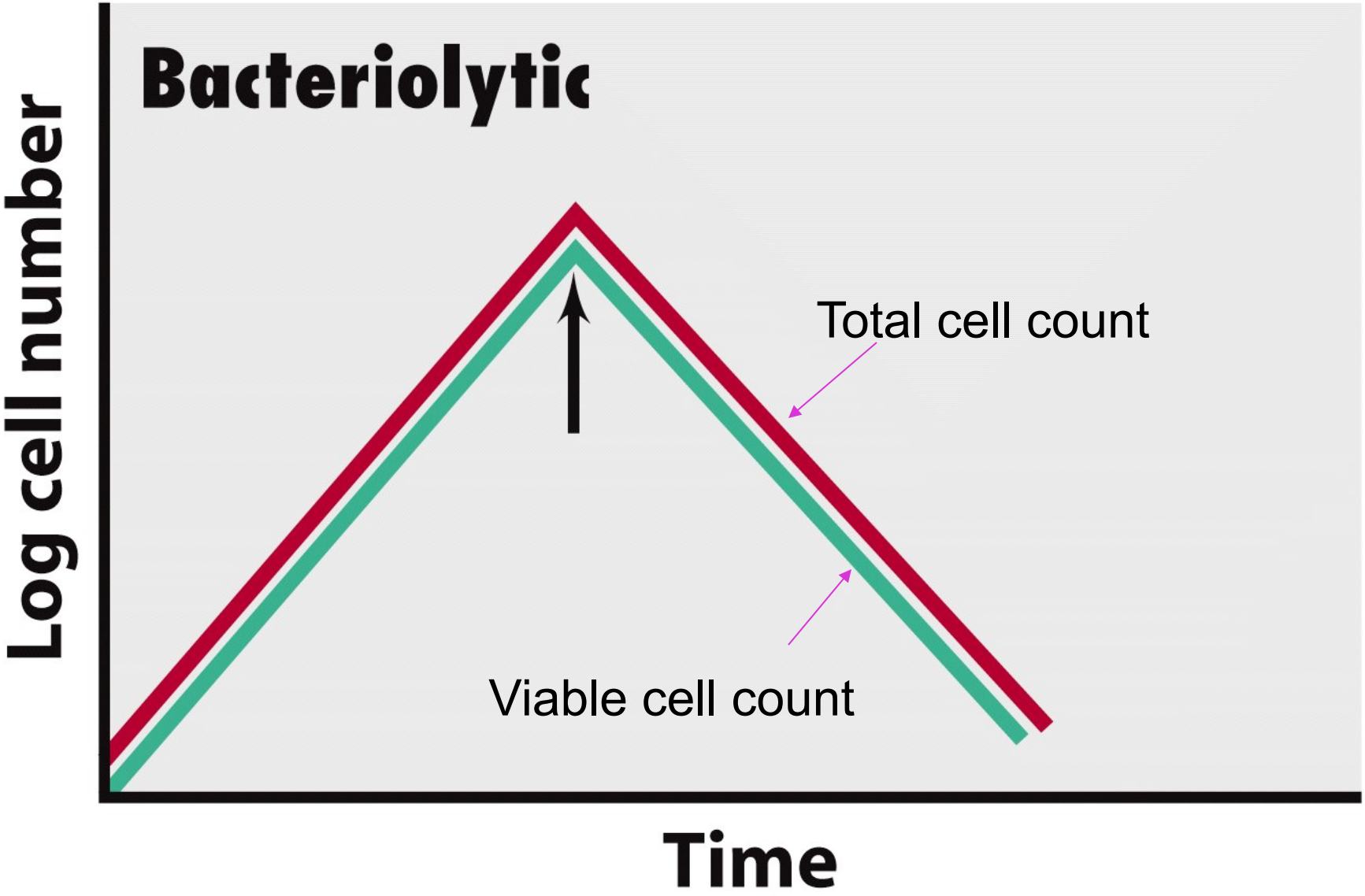


Figure 20-9c Brock Biology of Microorganisms 11/e

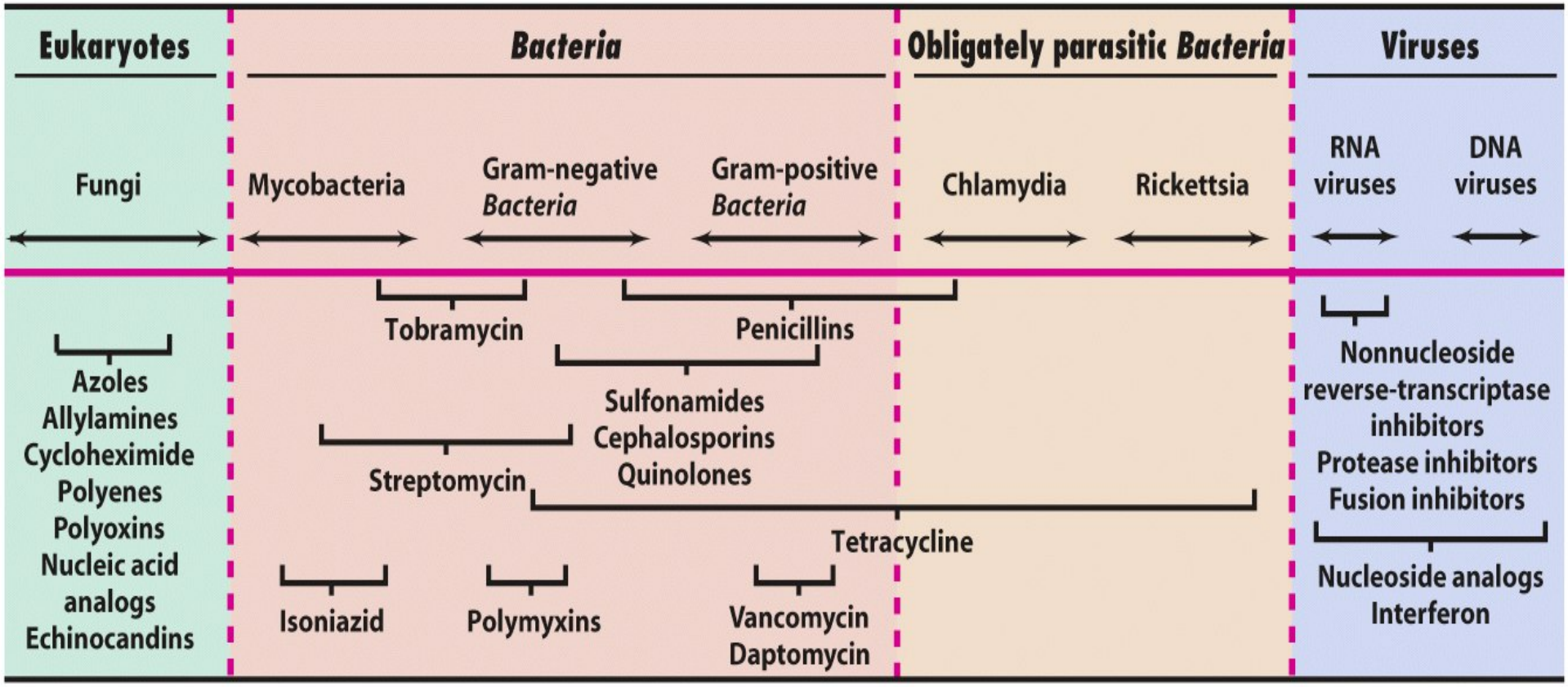


Figure 20-15 Brock Biology of Microorganisms 11/e

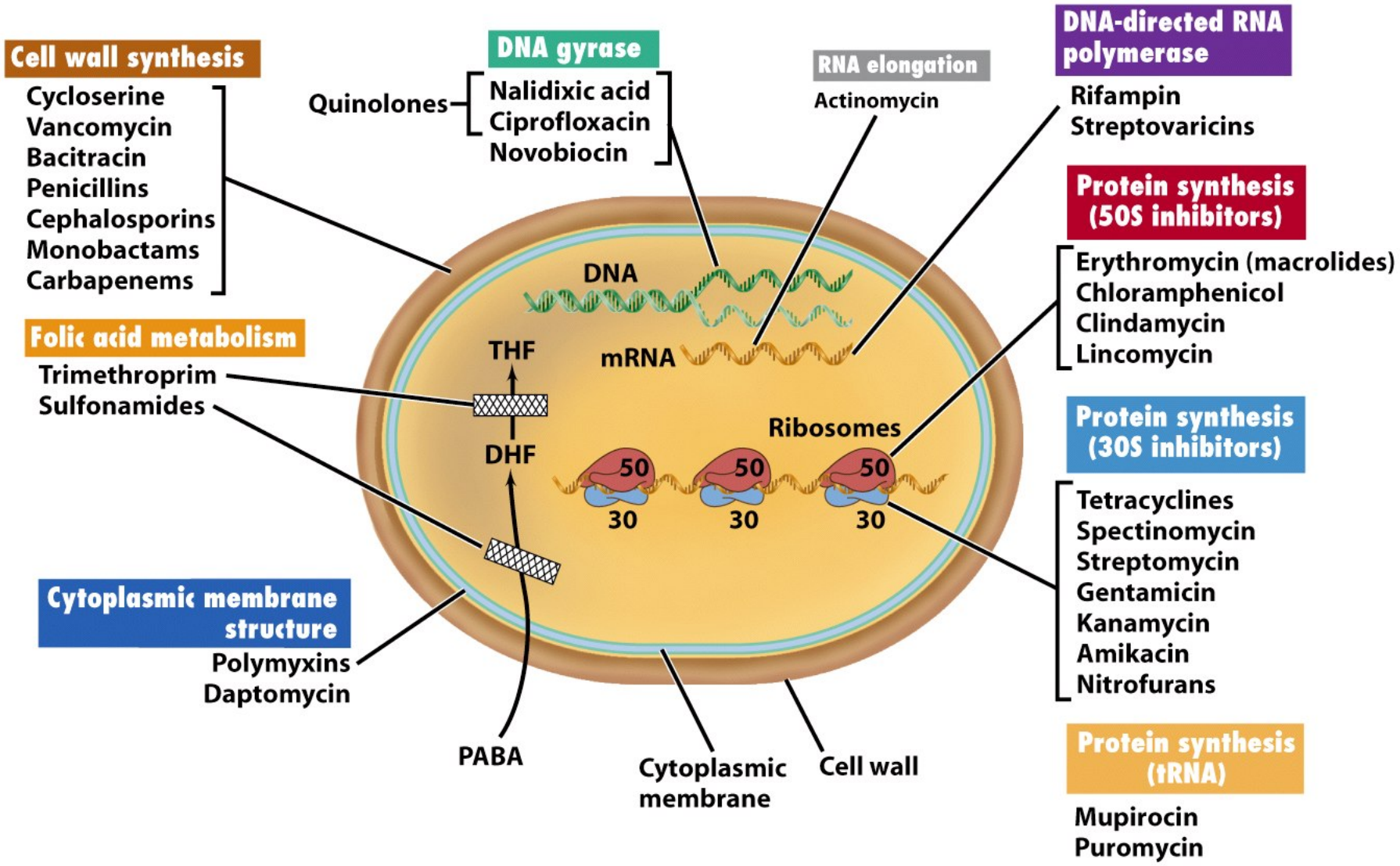


Figure 20-14 Brock Biology of Microorganisms 11/e

Antibiotics clinical use

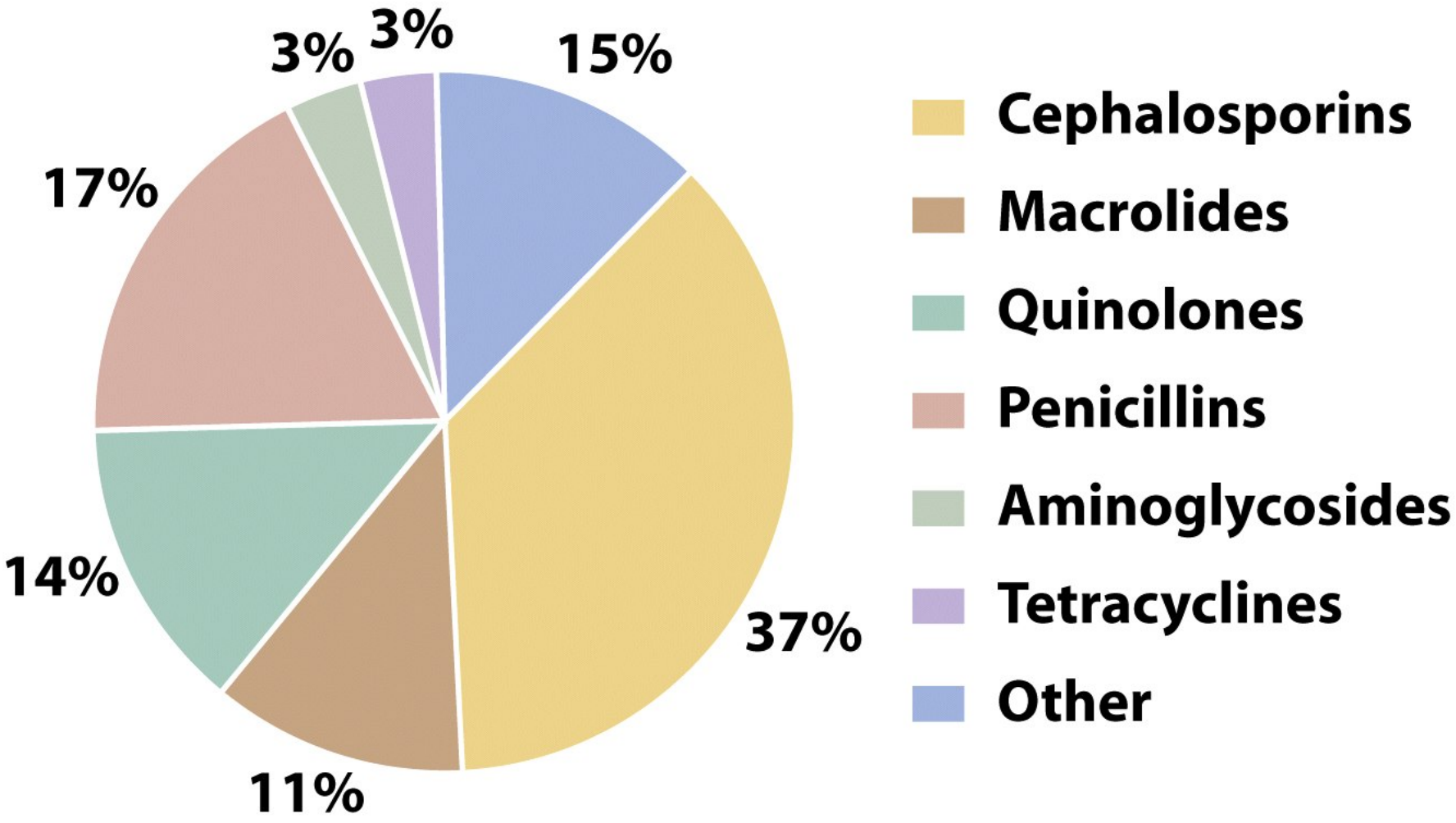


Figure 20-16 Brock Biology of Microorganisms 11/e

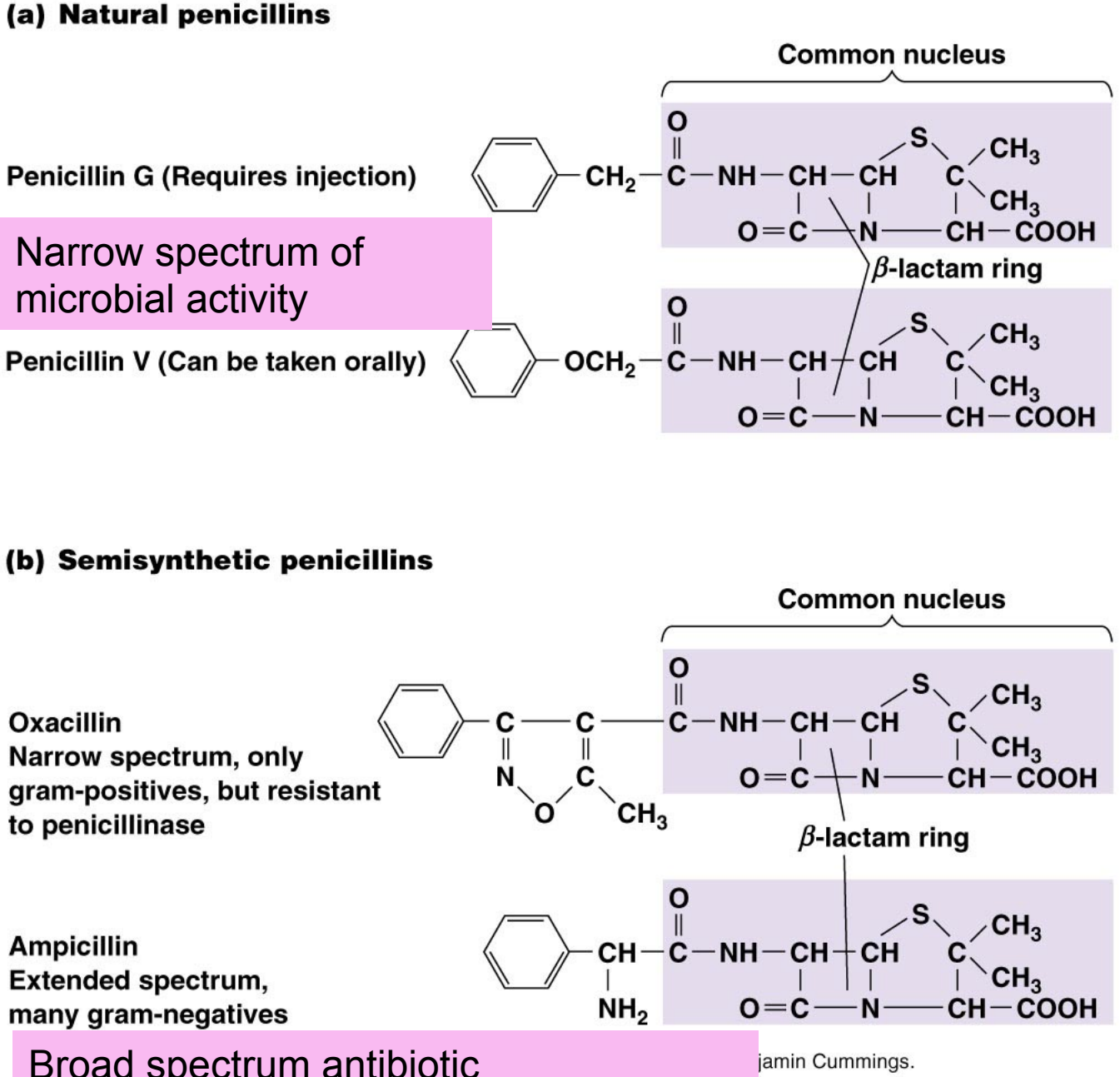


Figure 20.6 - Overview

Structure of peptidoglycanglycan tetrapeptide

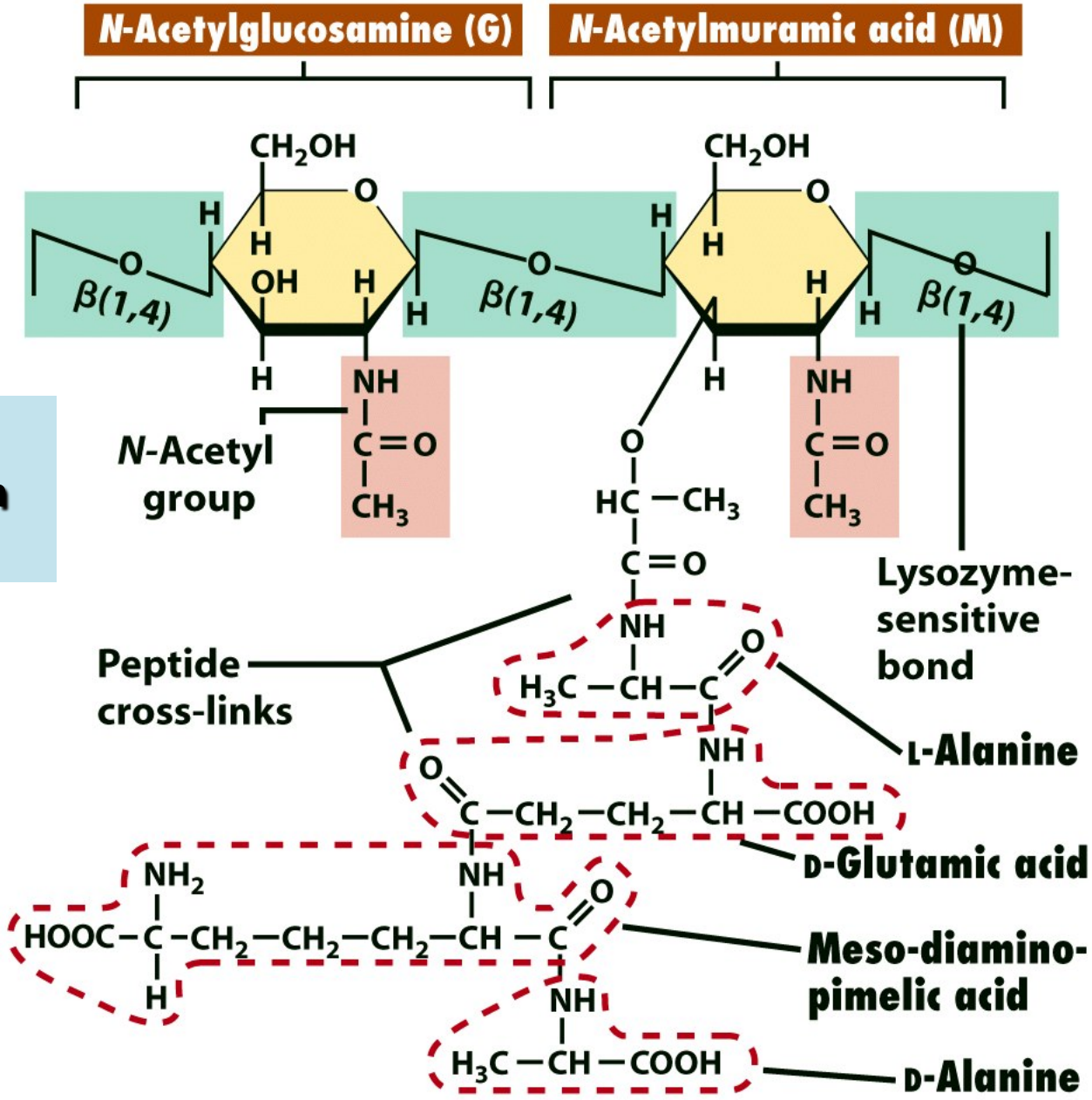


Figure 4-29 Brock Biology of Microorganisms 11/e

Peptidoglycan sheet in *Escherichia coli* and *Staphylococcus aureus*

Glycine interbridge in *S. aureus*

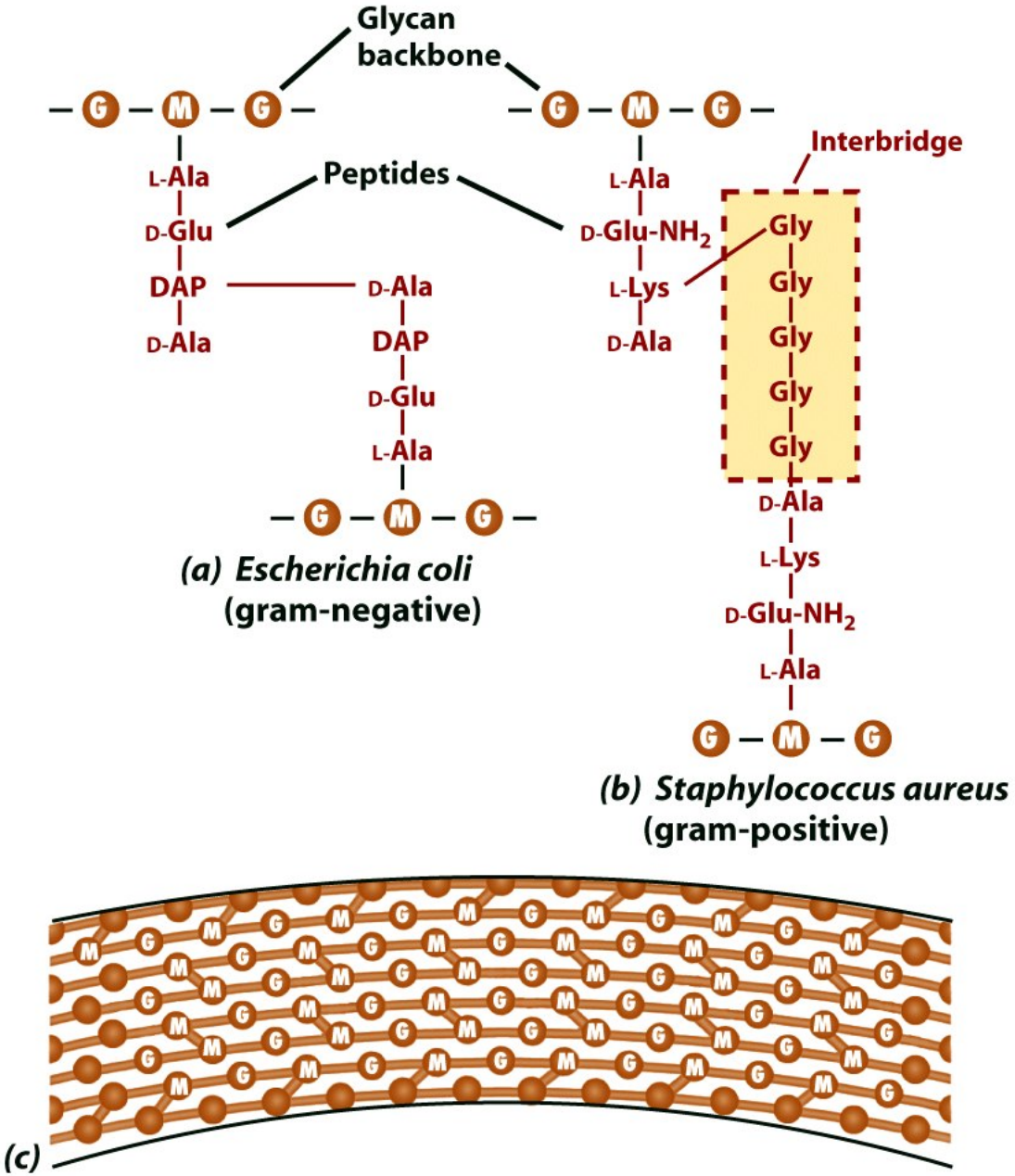
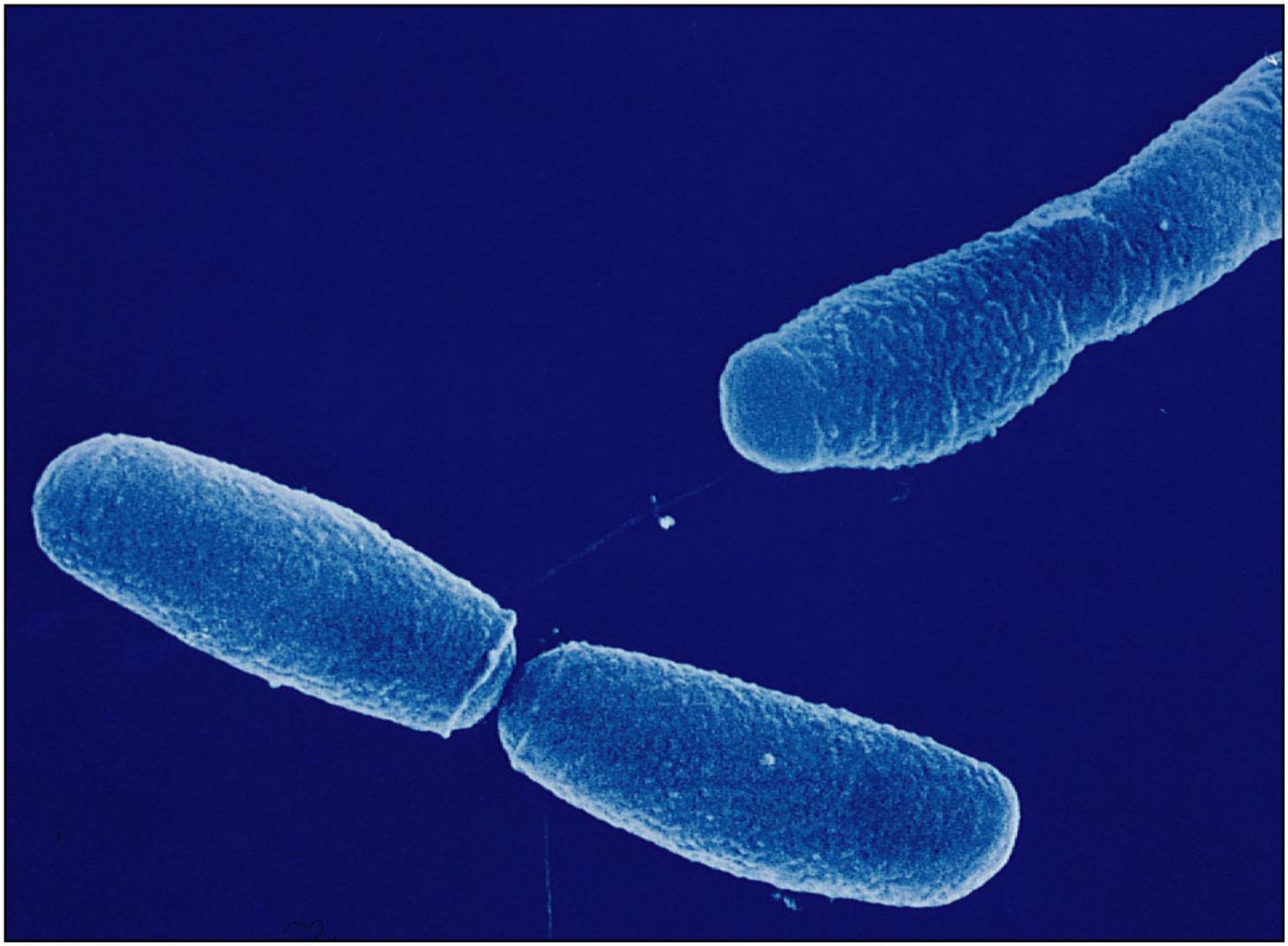
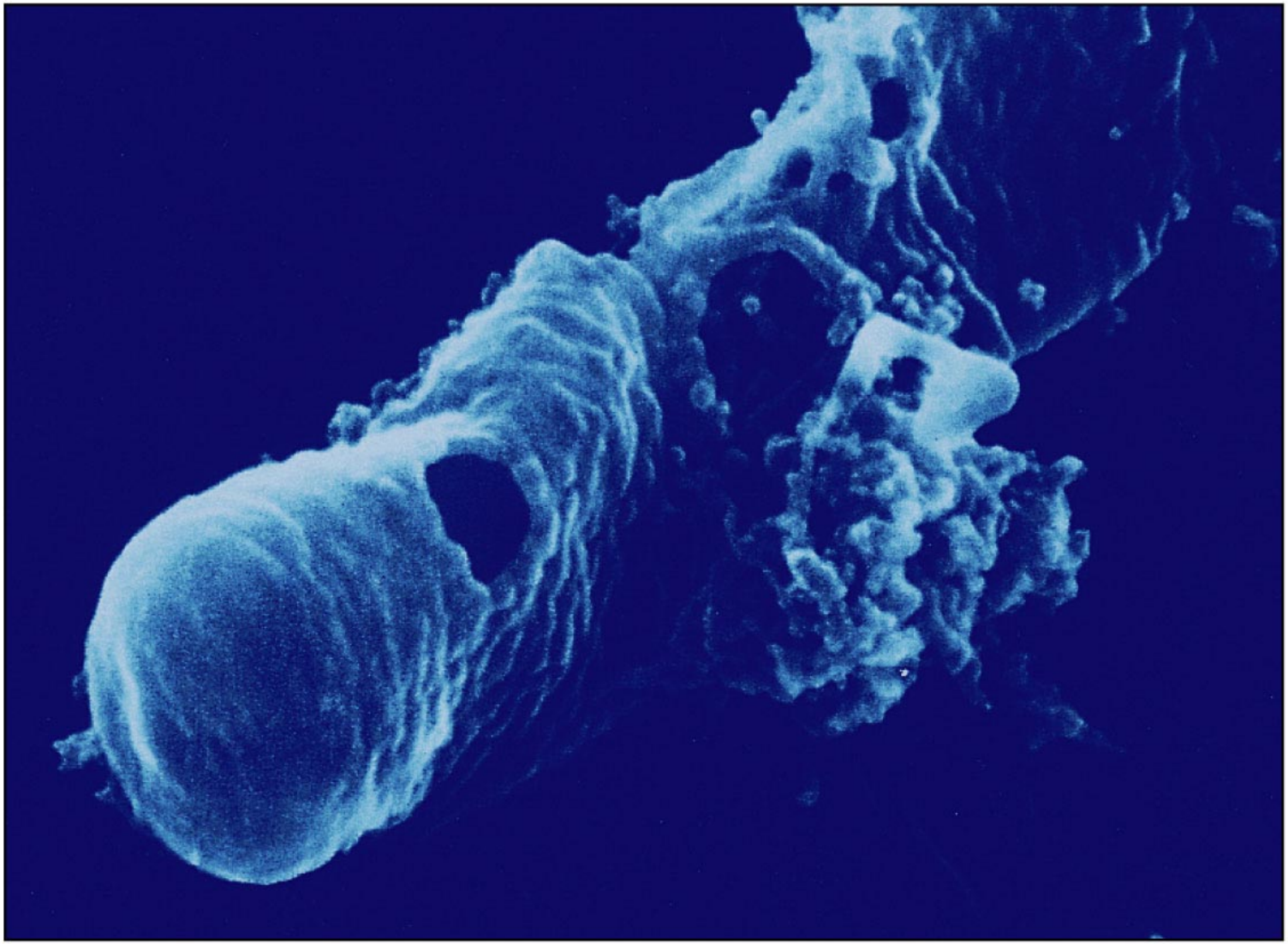


Figure 4-30 Brock Biology of Microorganisms 11/e



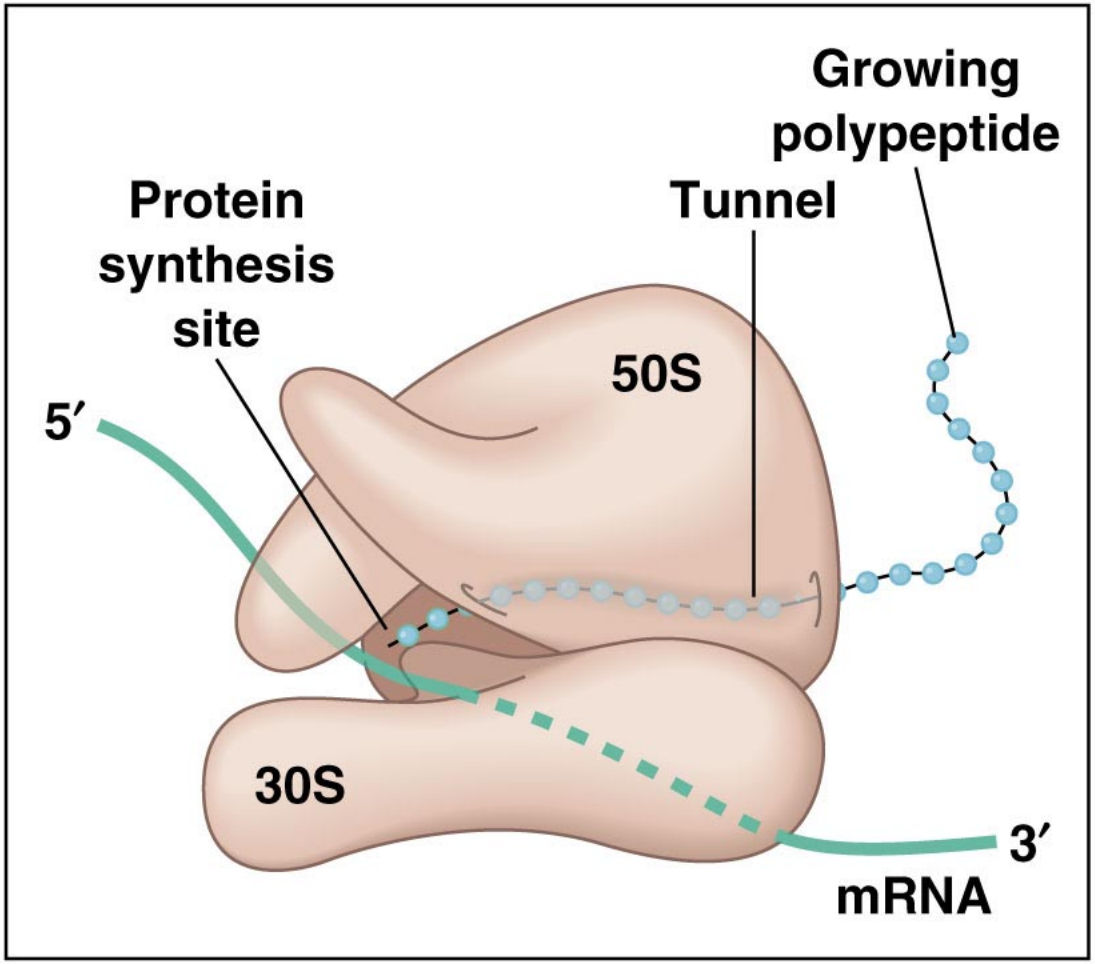
(a) Rod-shaped bacterium before penicillin.

Figure 20.3a



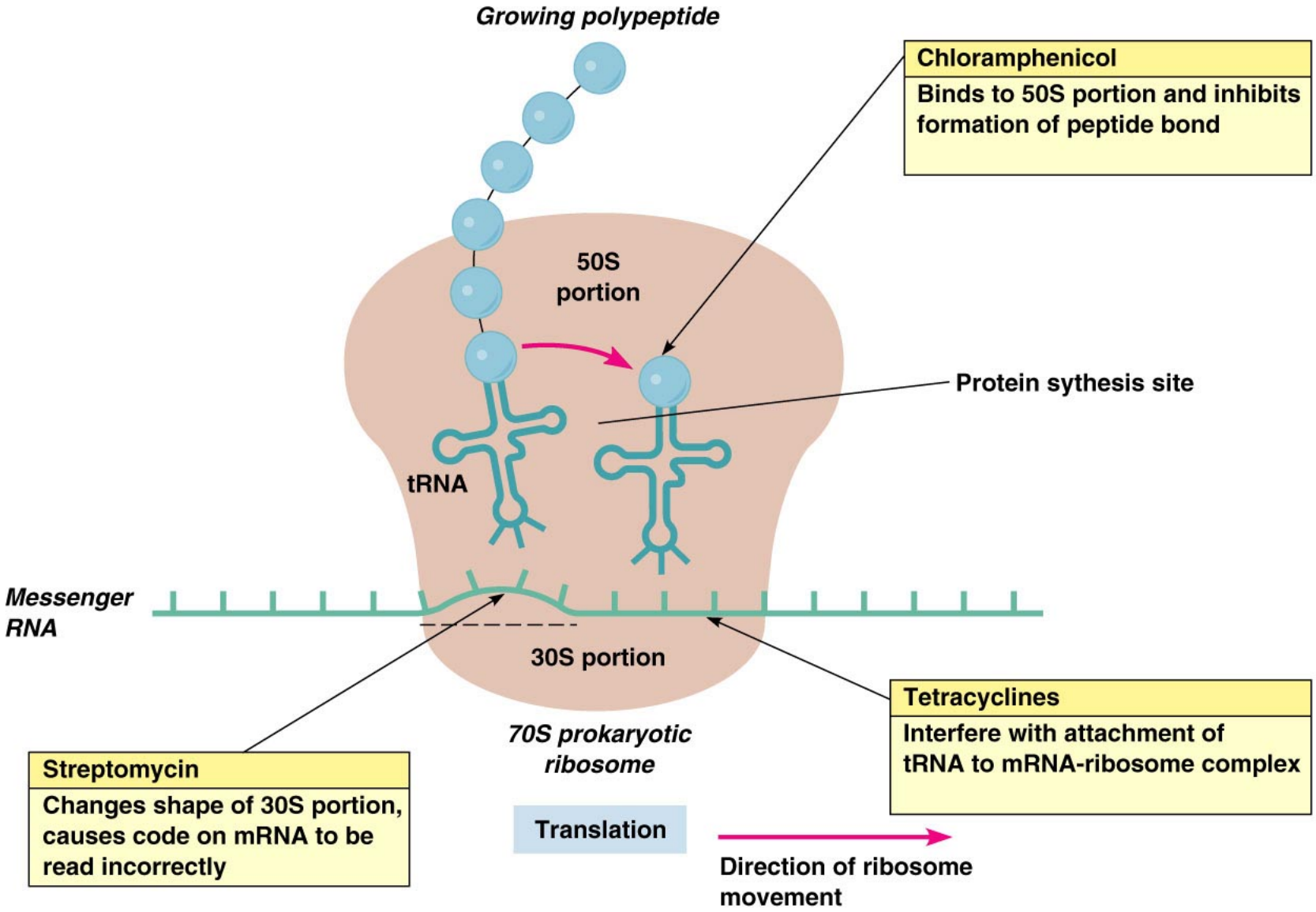
(b) The bacterial cell is lysing as penicillin weakens the cell wall.

Figure 20.3b



(a) Three-dimensional detail of the protein synthesis site showing the 30S and 50S subunit portions of the 70S prokaryotic ribosome.

Figure 20.4a



(b) In the diagram the black arrows indicate the different points at which chloramphenicol, the tetracyclines, and streptomycin exert their activities.

Figure 20.4b

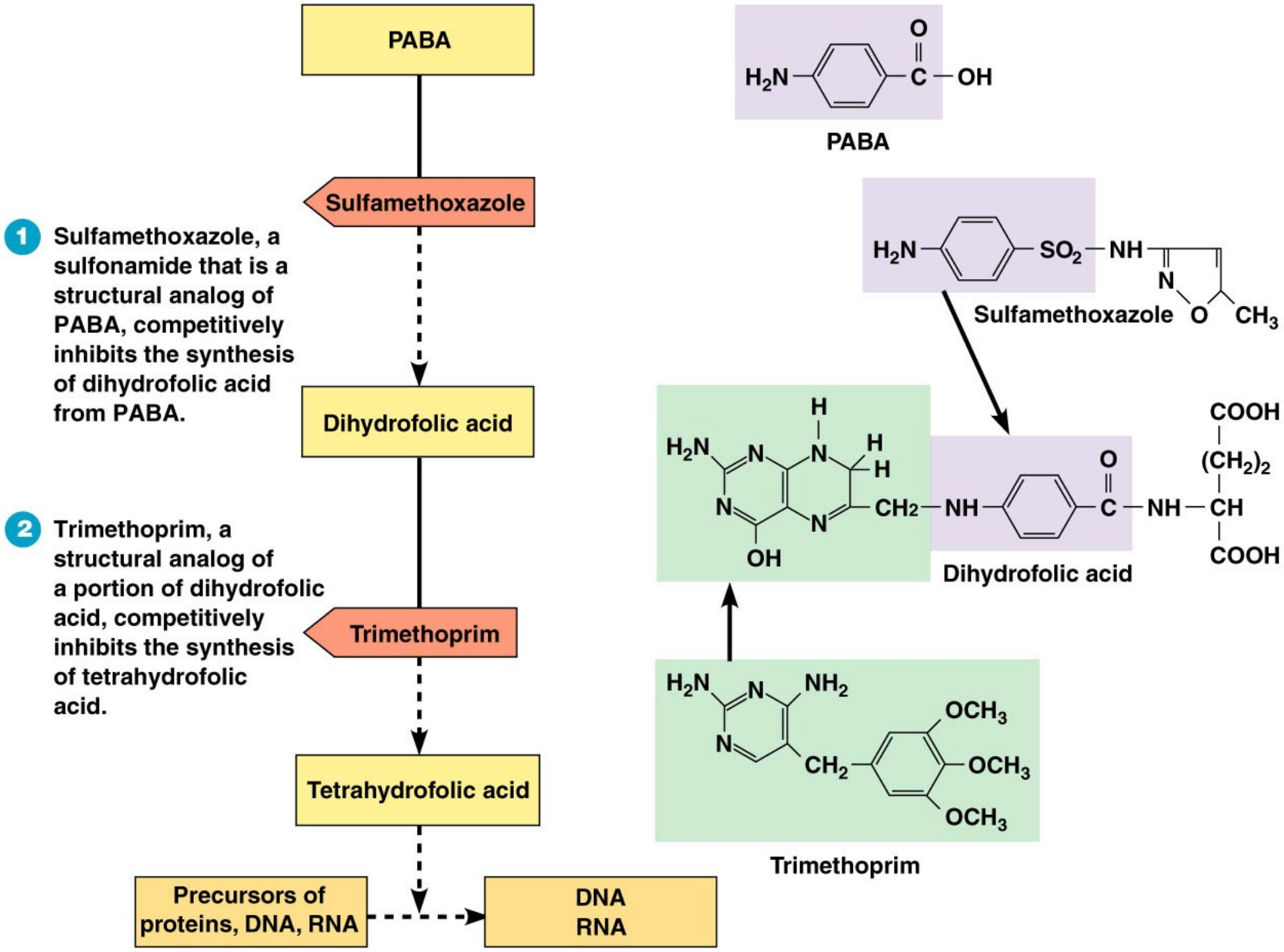
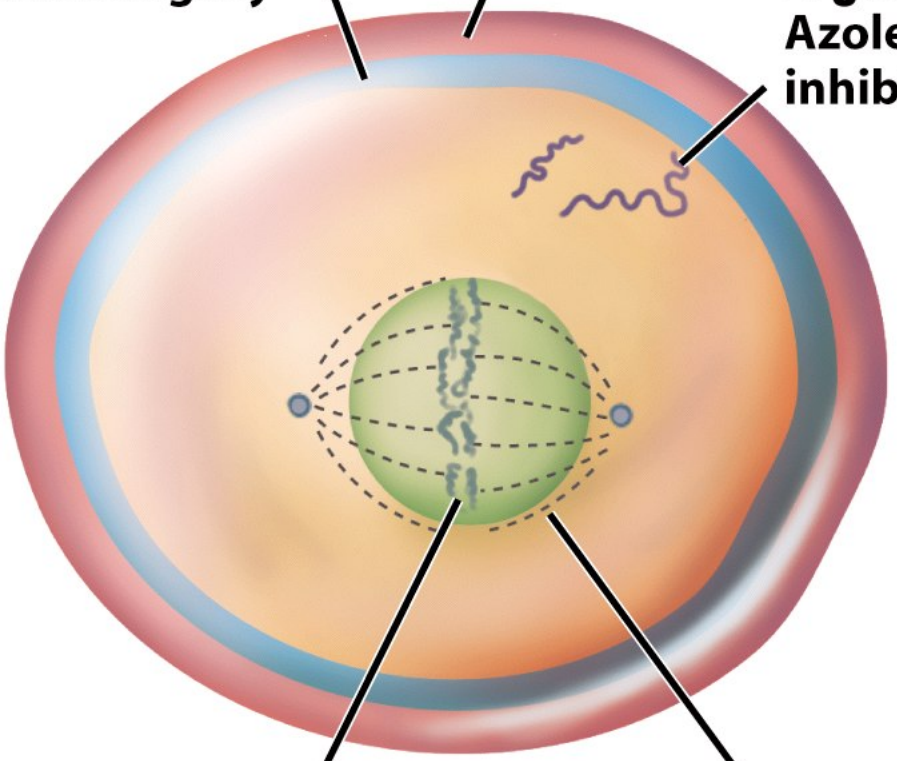


Figure 20.13 - Overview

Main fungal targets



Membrane functions:
Polyenes bind to ergosterol and disrupt membrane integrity

Cell wall synthesis:
Polyoxins inhibit chitin synthesis
Echinocandins inhibit glucan synthesis

Ergosterol synthesis:
Azoles and Allylamines inhibit synthesis

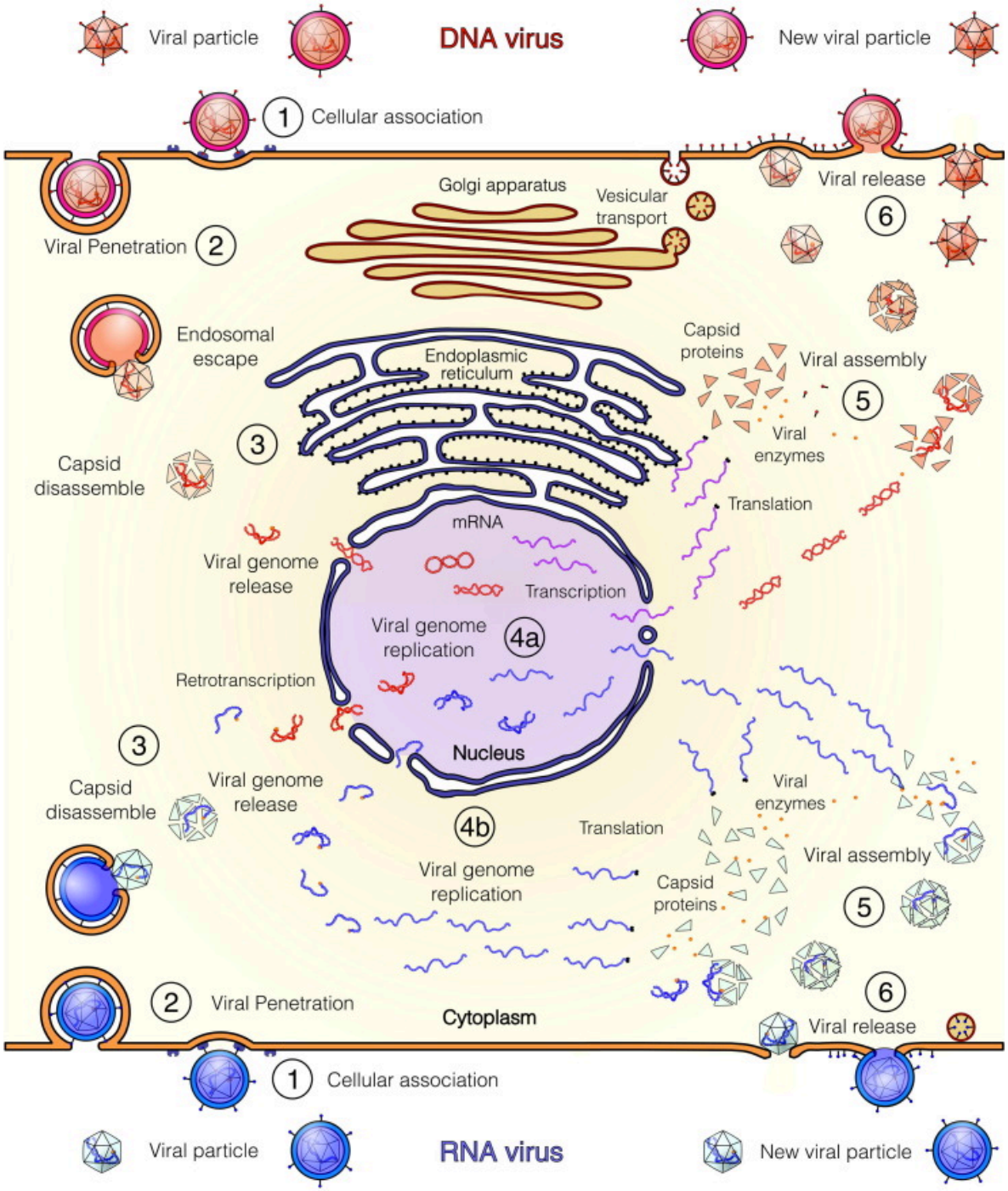
Nucleic acid synthesis:
5-Fluorocytosine is a nucleotide analog that inhibits nucleic acid synthesis

Microtubule formation:
Griseofulvin disrupts microtubule aggregation during mitosis

Figure 20-24 Brock Biology of Microorganisms 11/e

Injury of plasma membrane of a yeast caused by antifungal drug

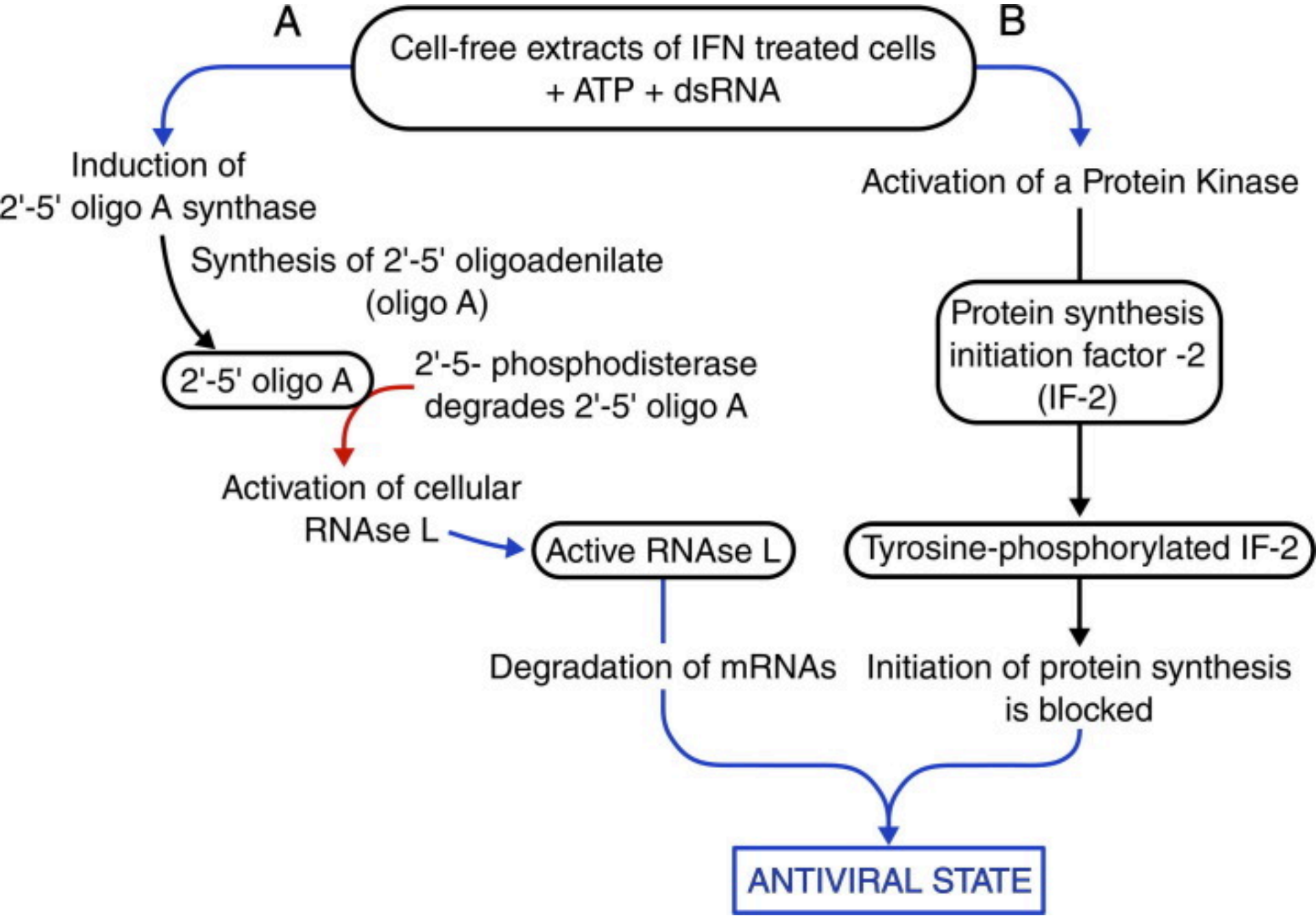
Figure 20.5

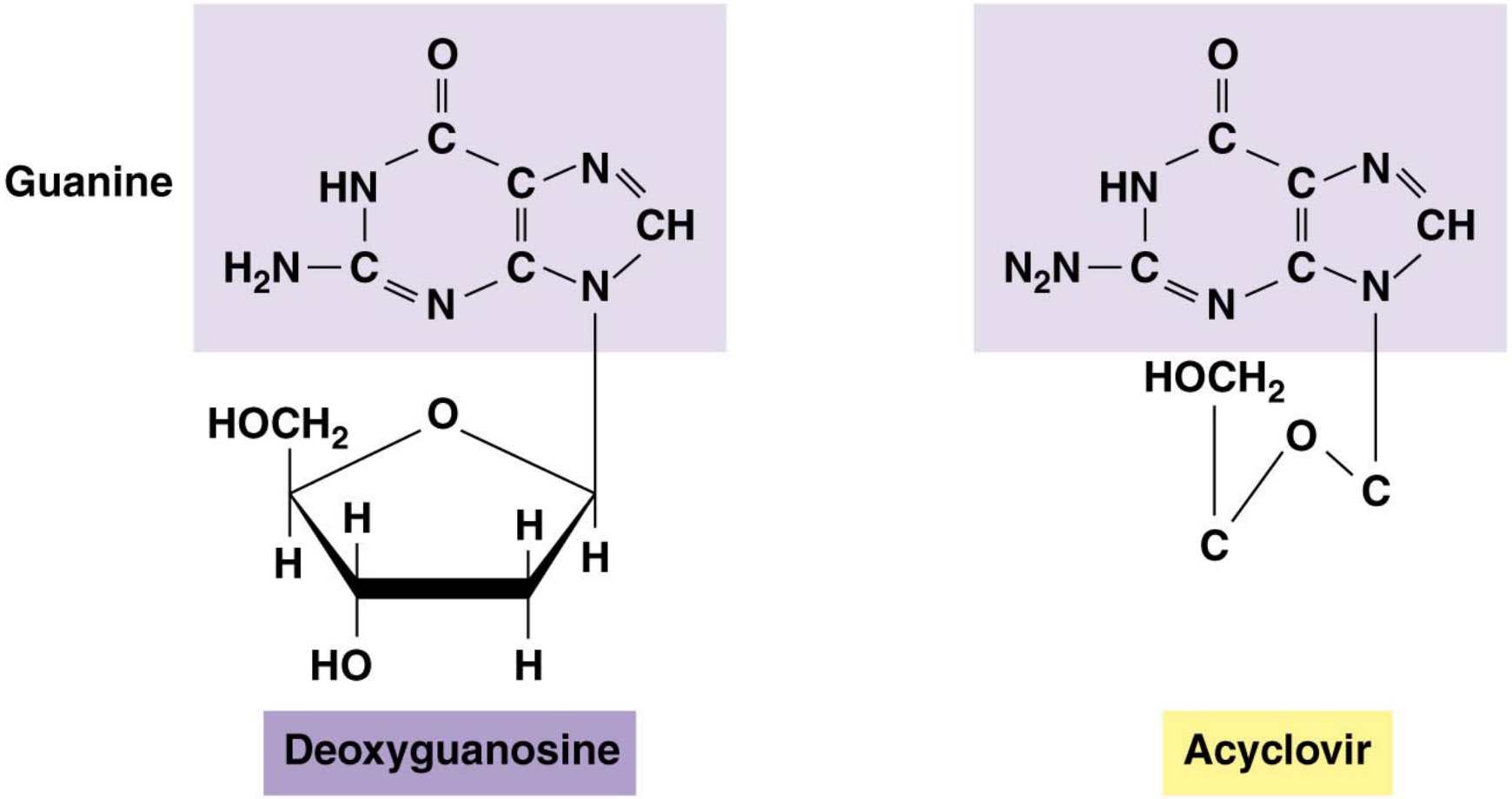


Schematic representation of viral life cycle. Top: DNA virus cycle (red), bottom: RNA virus cycle (blue). The numbers indicate the steps usually targeted by antiviral compounds: 1. Adsorption, 2. Internalization, 3. Viral capsid release and disaggregation, 4. DNA/ RNA replication (a: nucleus, b: cytoplasm), 5. Viral assembly and maturation, and 6. Release of mature virions.

Antivirals from bacteria

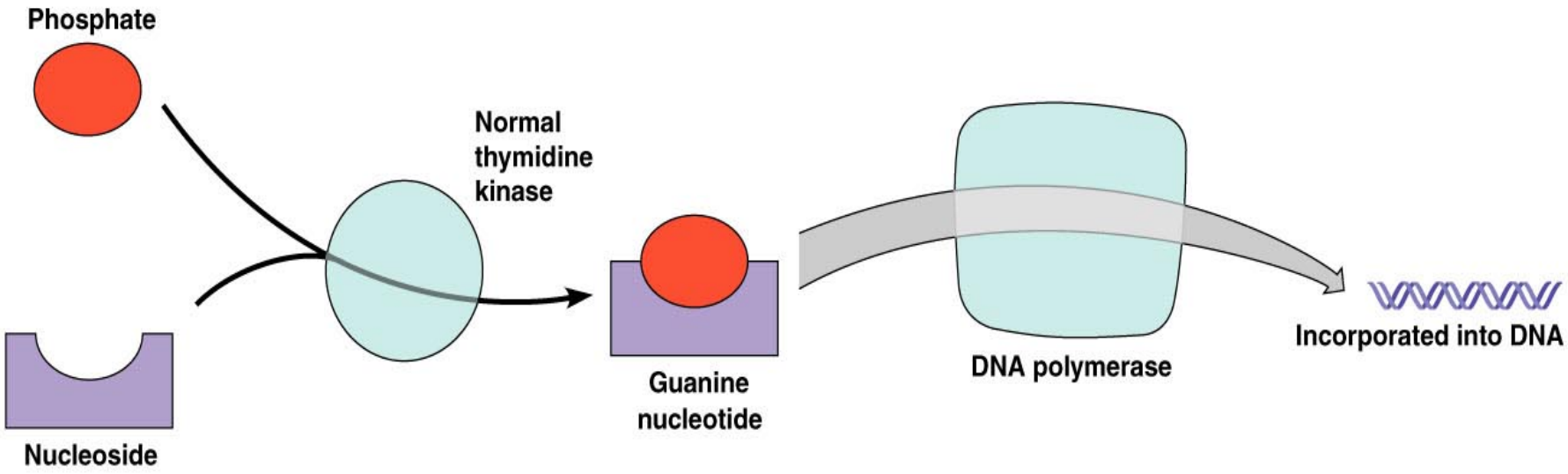
macrolide antibiotic *borrelidin* (CID: 6436801), produced by Sptreptomyces sp (recently rediscovered and named Streptomyces heilongjiangensis).
Ehrlichin (synthesized by Streptomyces lavendulae),
Abikoviromycin (CID: 6450263, produced by several species of Streptomyces),
Violarin (CID: 44256909, from Actinomyces violaceous),
Myxoviromycin (CID: 160703, a compound produced by Streptomyces sp, that targets orthomyxoviruses),
Virocidin (CID: 9989534),
Niromycins (CAS: 101997-22-2 and CAS: 101997-20-0, from actinomycetes), extracts from propionibacteria, and
Vivomycin (CID: 3037981, produced by Streptomyces C2989) .


A
Cell-free extracts of IFN treated cells
+ ATP + dsRNA
B
Induction of
2'-5' oligo A synthase
Synthesis of 2'-5' oligoadenilate
(oligo A)
2'-5' oligo A
2'-5- phosphodisterase
degrades 2'-5' oligo A
Activation of cellular
RNAse L
Active RNAse L
Degradation of mRNAs
Activation of a Protein Kinase
Protein synthesis
initiation factor -2
(IF-2)
Tyrosine-phosphorylated IF-2
Initiation of protein synthesis
is blocked
ANTIVIRAL STATE



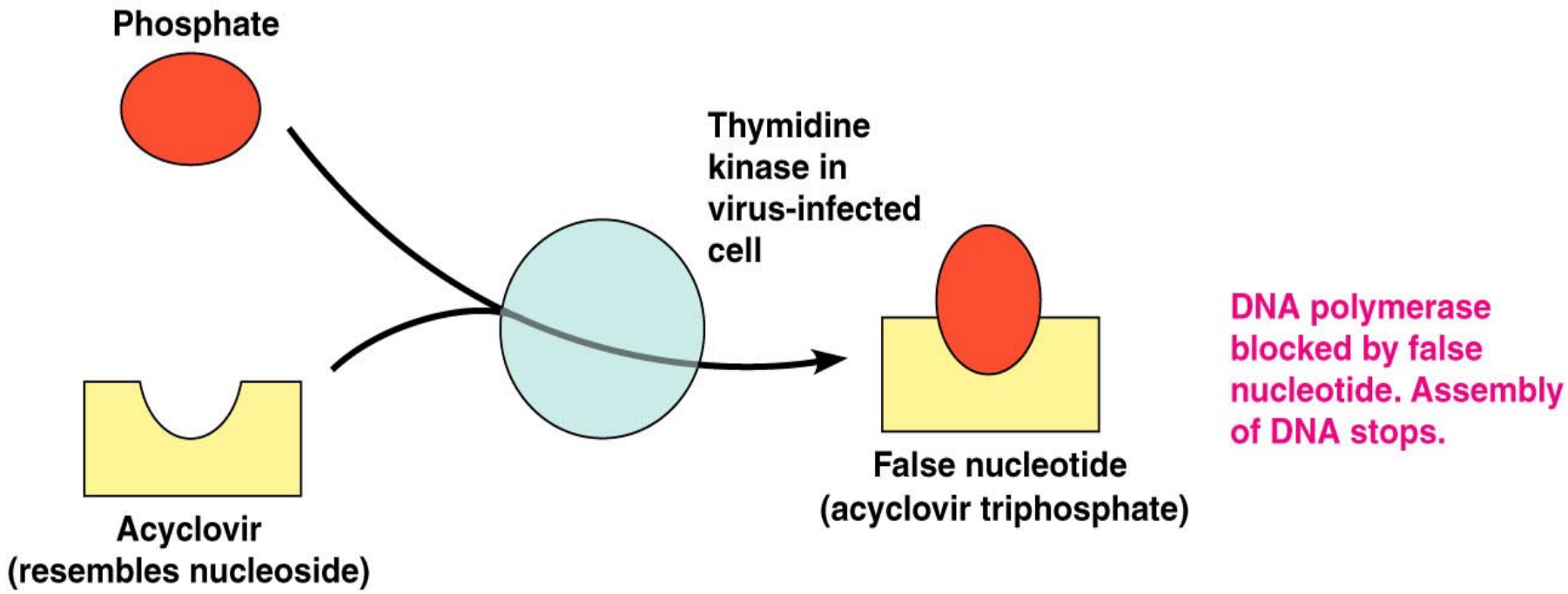
(a) Acyclovir structurally resembles the nucleoside deoxyguanosine.

Figure 20.16a



(b) The enzyme thymidine kinase combines phosphates with nucleosides to form nucleotides, which are then incorporated into DNA.

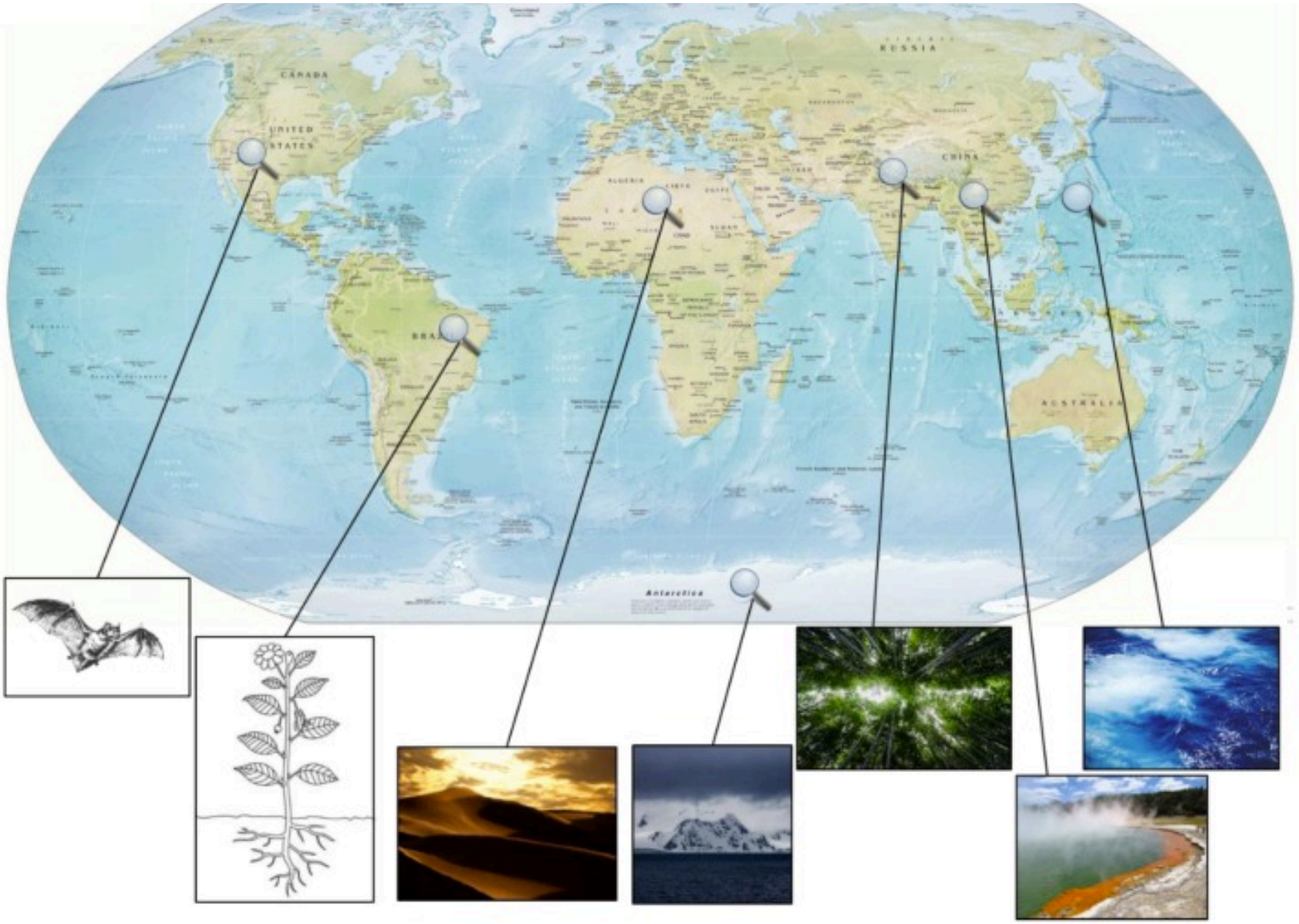
Figure 20.16b



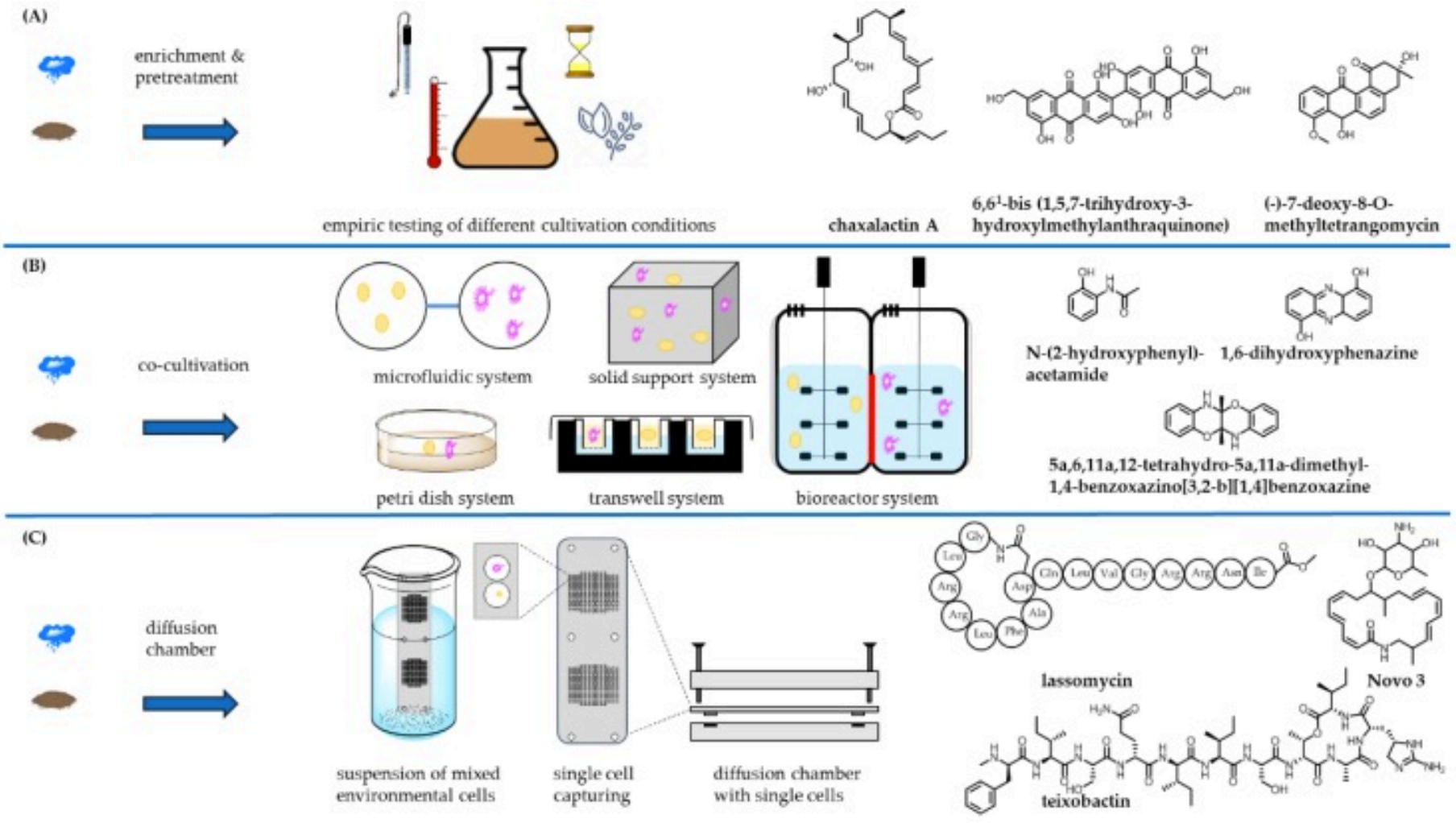
(c) Acyclovir has no effect on a cell not infected by a virus, that is, with normal thymidine kinase. In a virally infected cell, the thymidine kinase is altered and converts the acyclovir (which resembles the nucleoside deoxyguanosine) into a false nucleotide—which blocks DNA synthesis by DNA polymerase.

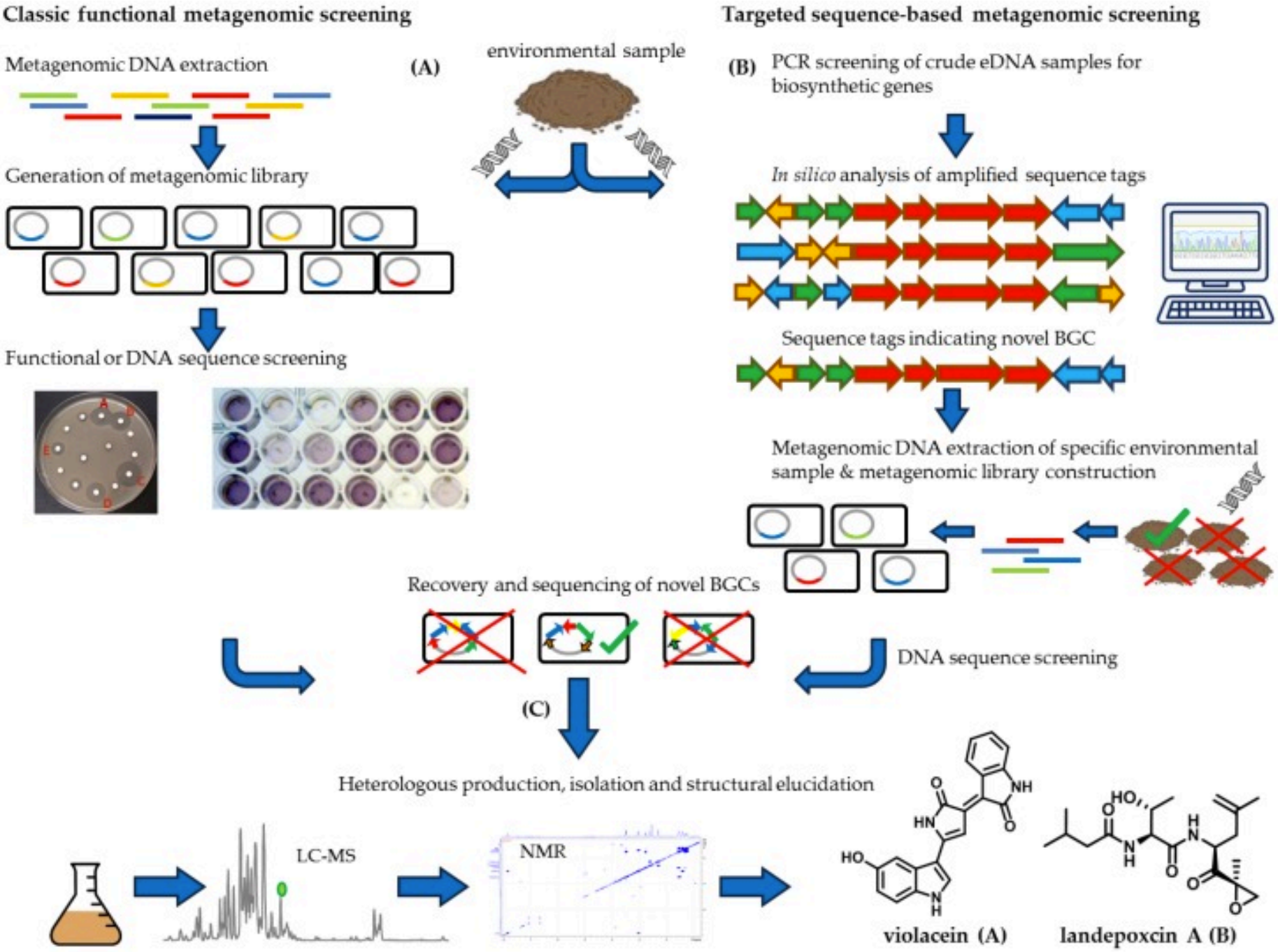
Figure 20.16c

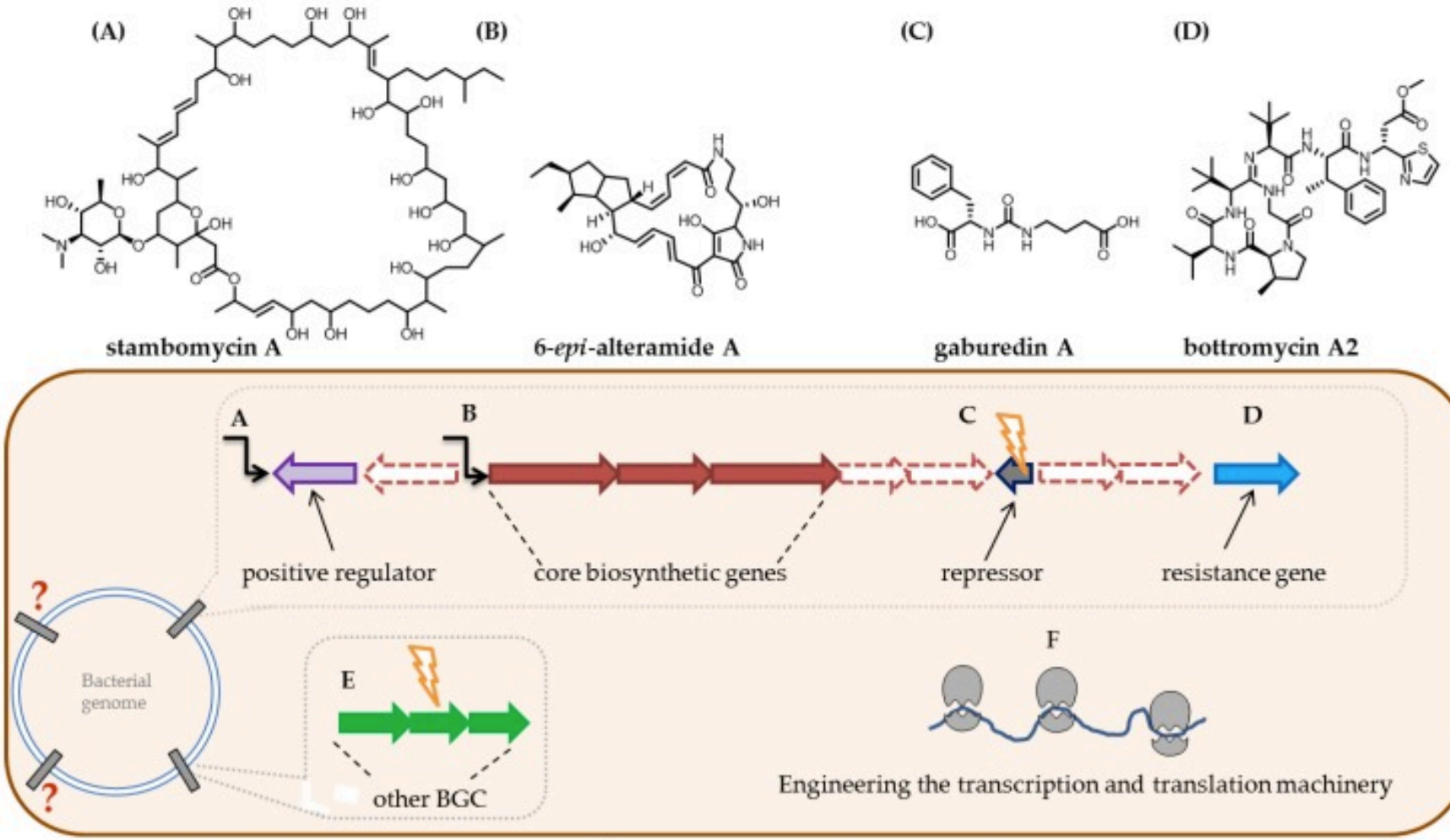
Underexploited habitats of actinobacteria attracted more attention for microbial natural product discovery. Currently, oceans, deserts, mountains and Antarctica ranges together with hot springs and endophytes and symbionts are focuses of the search for new bioactive compounds.

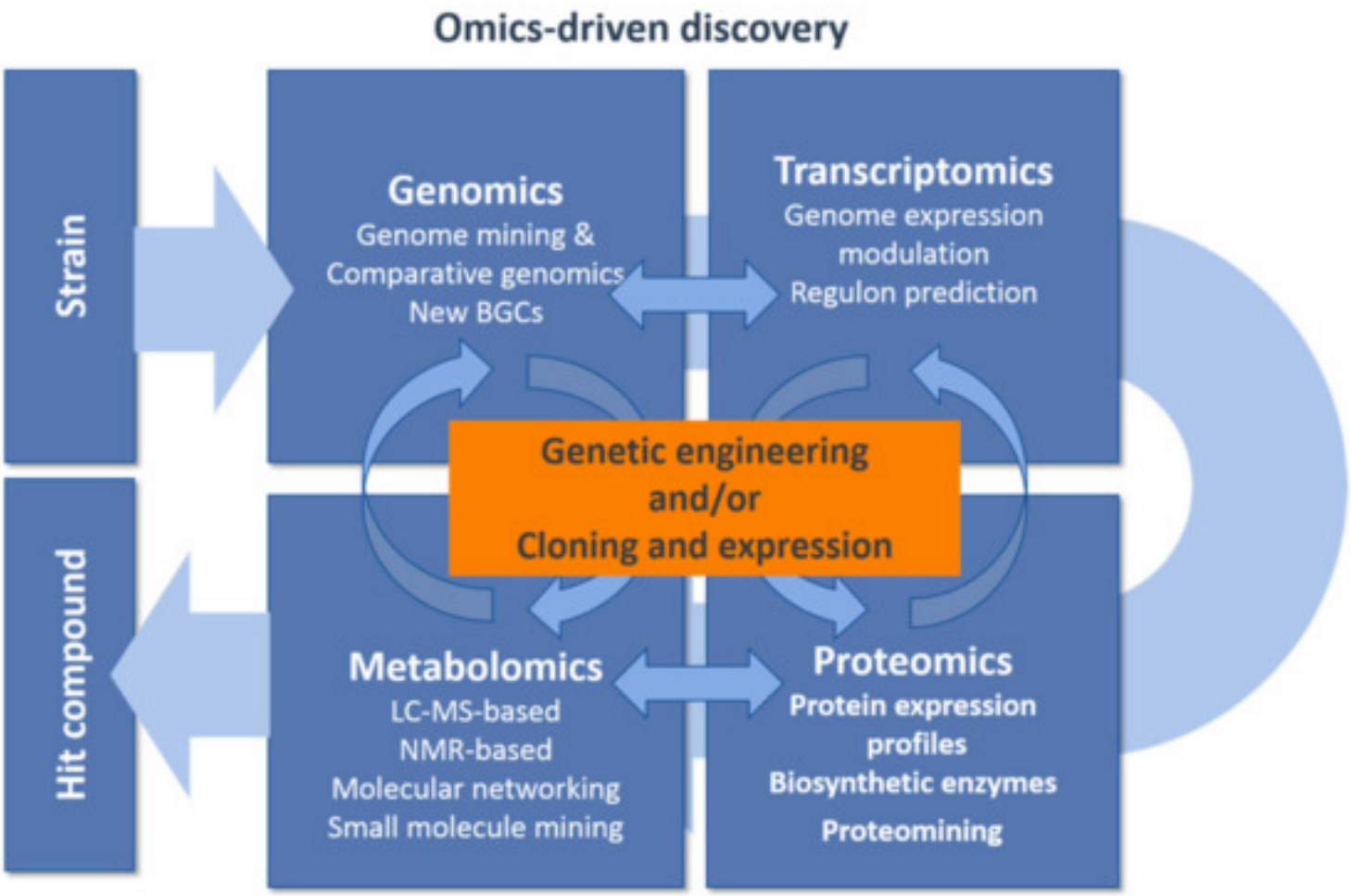


Isolation strategies. (**A**) Soil sample or marine sample undergoes enrichment and/or pretreatment to increase the chance to isolate new species and/or reduce undesirable background from previously isolated strains. Methods for fermentation varies in incubation time, media composition, additives, pH and temperature to enable growth of desirable strains.. (**B**) Sample is co-cultivated with other microorganisms to promote culturable isolates or to stimulate the secondary metabolism. (**C**) Sample is used to create a suspension of mixed environmental cells. The isolation chip (iChip) plate is immersed into this suspension to capture (on average) a single cell.




Classic functional metagenomic screening
Targeted sequence-based metagenomic screening
Metagenomic DNA extraction
(A)
environmental sample
(B) PCR screening of crude eDNA samples for biosynthetic genes
Generation of metagenomic library
In silico analysis of amplified sequence tags
Functional or DNA sequence screening
Sequence tags indicating novel BGC
Metagenomic DNA extraction of specific environmental sample & metagenomic library construction
Recovery and sequencing of novel BGCs
DNA sequence screening
(C)
Heterologous production, isolation and structural elucidation
LC-MS
NMR
violacein (A)
landepoxcin A (B)


(A)
(B)
(C)
(D)
stambomycin A
6-epi-alteramide A
gaburedin A
bottromycin A2
A
B
C
D
positive regulator
core biosynthetic genes
repressor
resistance gene
Bacterial genome
E
other BGC
F
Engineering the transcription and translation machinery
(E)
(F)
amexanthomycin A
piperidamycin A


Omics-driven discovery
Strain
Hit compound
Genomics
Genome mining &
Comparative genomics
New BGCs
Transcriptomics
Genome expression
modulation
Regulon prediction
Genetic engineering
and/or
Cloning and expression
Metabolomics
LC-MS-based
NMR-based
Molecular networking
Small molecule mining
Proteomics
Protein expression
profiles
Biosynthetic enzymes
Proteomining

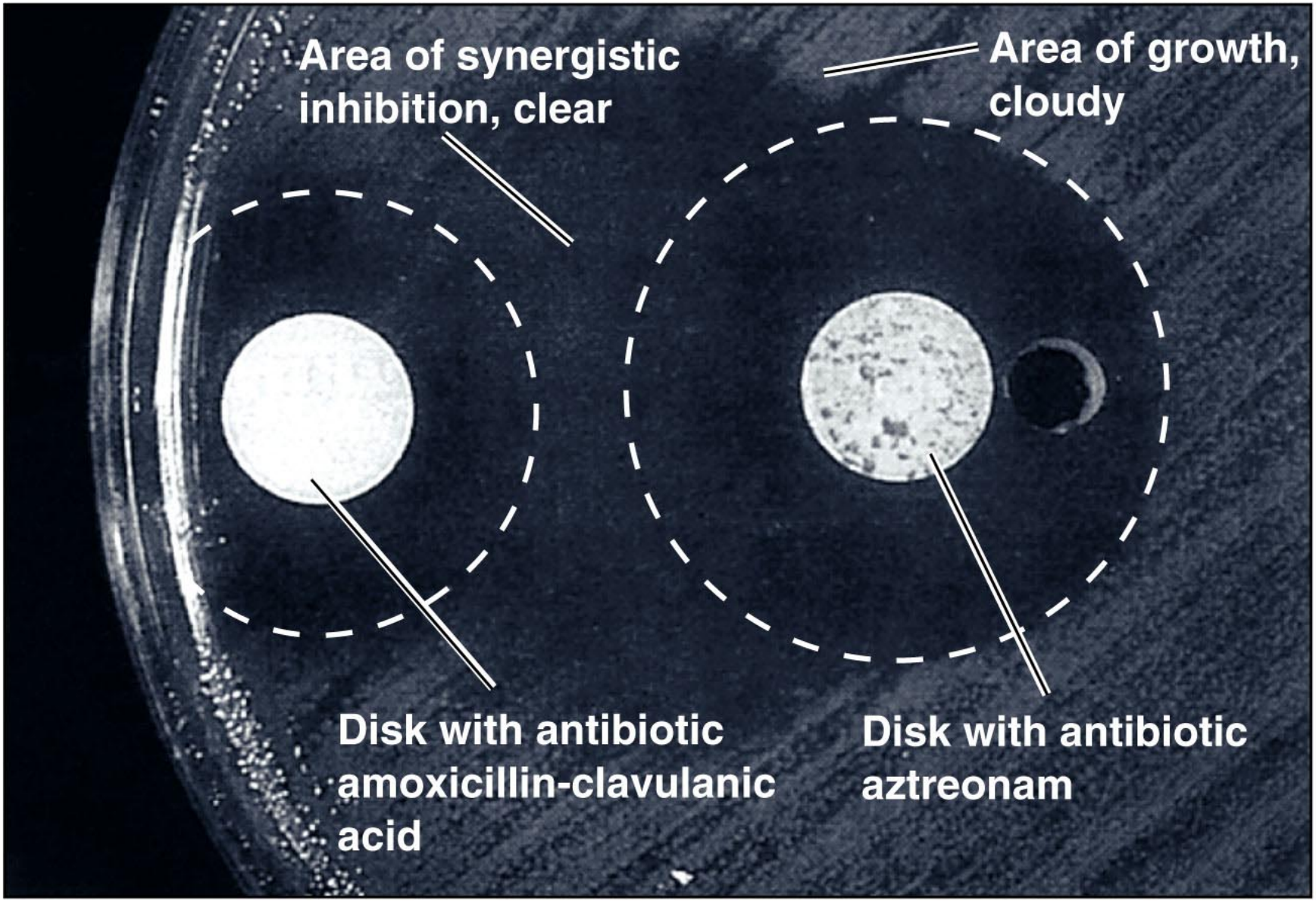


Figure 20.22

1. Reversible attachment of planktonic cells. (seconds)
2. First colonizers become irreversibly attached. (second, minutes)
3. Growth and cell division. (hours, days)
4. Production of EPS and formation of water channels. (hours, days)
5. Attachment of secondary colonizers and dispersion of microbes to new sites. (days, months)