Basic Water Microbiology

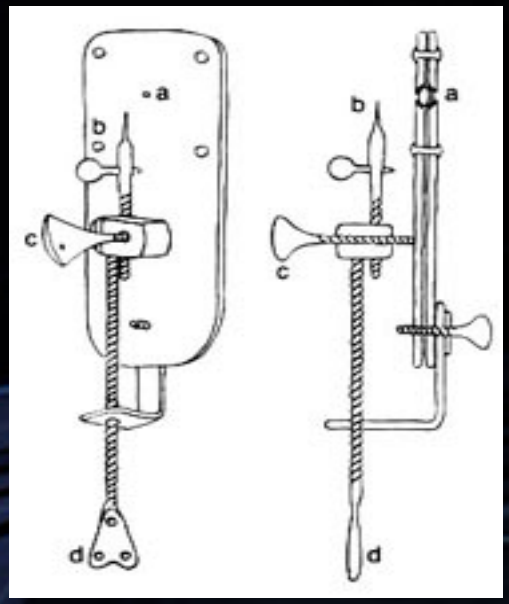
Discovery of the Microbial World

"I have had several gentlewomen in my house, who were keen on seeing the little eels in vinegar; but some of them were so disgusted at the spectacle, that they vowed never to use vinegar again.

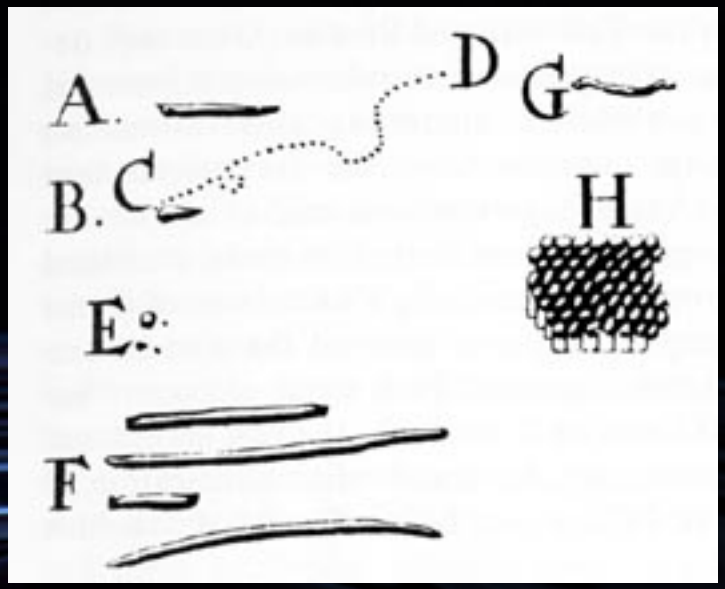
But what if one should tell such people in future that there are more animals living in the scum on the teeth in a man's mouth, than there are men in a whole kingdom?"

Antonj van Leeuwenhoek
1632-1723

Leeuwenhoek's Microscope

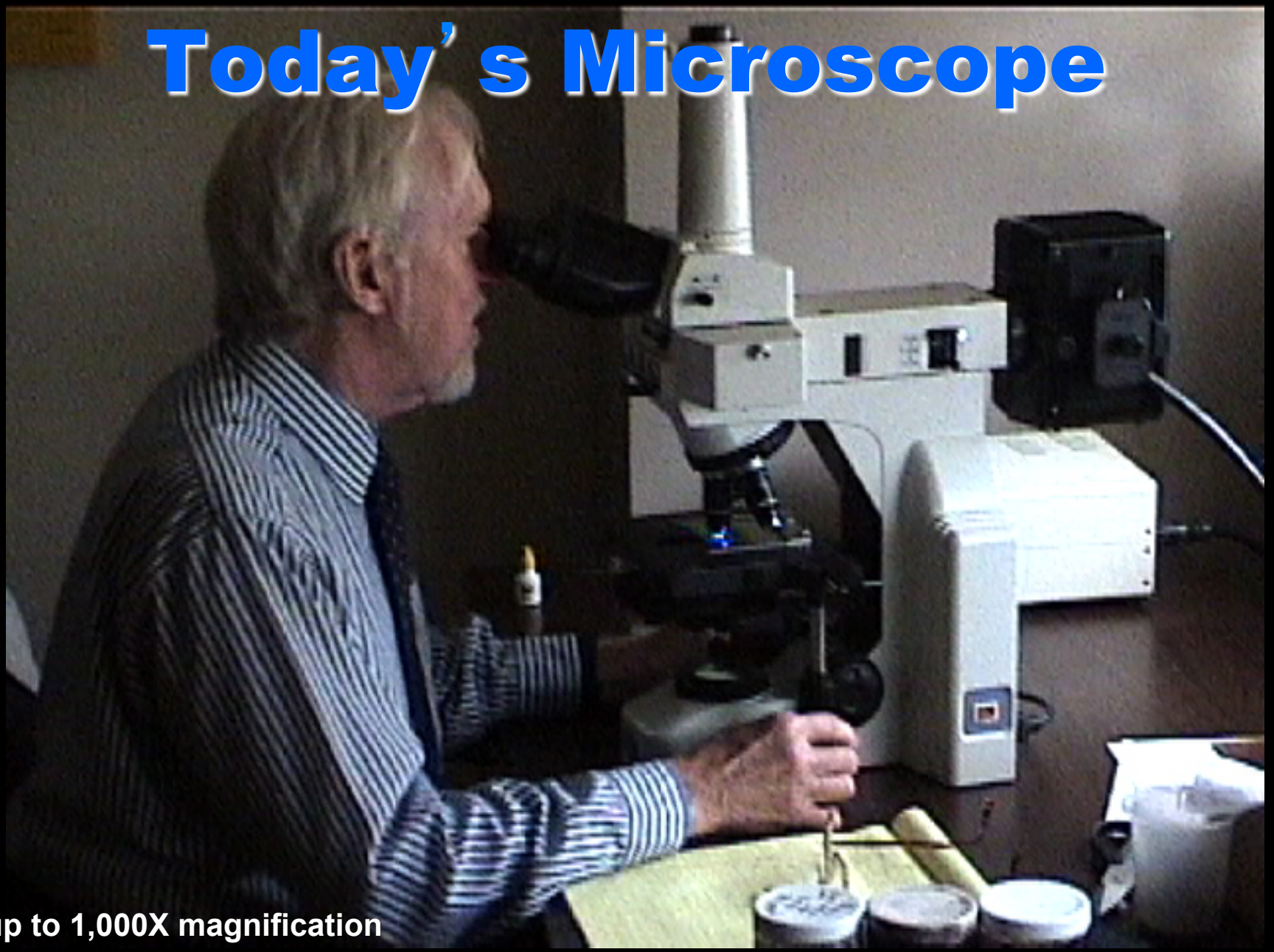


50 to 300X magnification

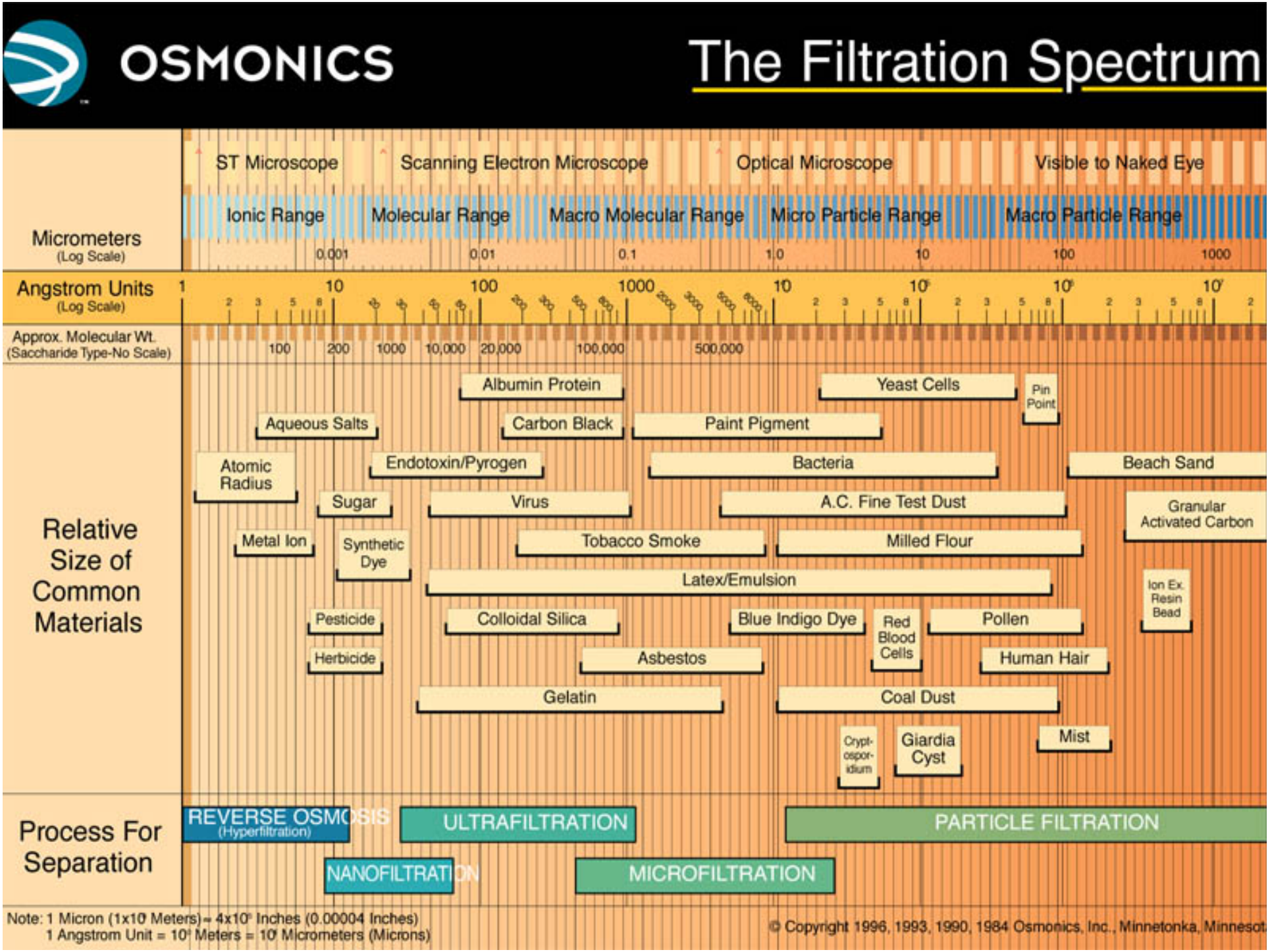


"animalcules," or little animals

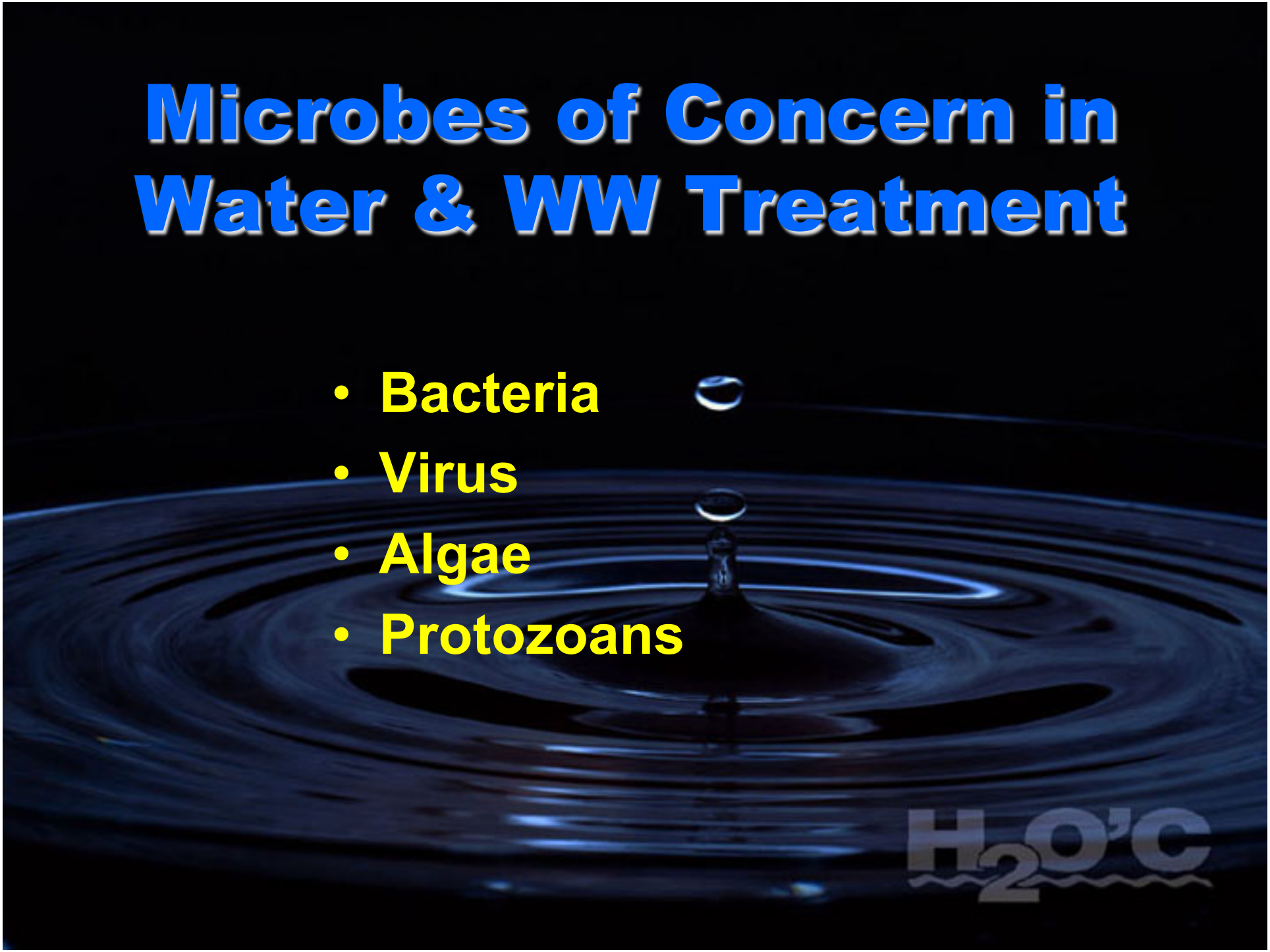
Today's Microscope



up to 1,000X magnification


OSMONICS
The Filtration Spectrum
ST Microscope
Scanning Electron Microscope
Optical Microscope
Visible to Naked Eye
Ionic Range
Molecular Range
Macro Molecular Range
Micro Particle Range
Macro Particle Range
Micrometers (Log Scale)
0.001
0.01
0.1
1.0
10
100
1000
Angstrom Units (Log Scale)
Approx. Molecular Wt. (Saccharide Type-No Scale)
100
200
1000
10,000
20,000
100,000
500,000
Relative Size of Common Materials
Albumin Protein
Yeast Cells
Pin Point
Aqueous Salts
Carbon Black
Paint Pigment
Atomic Radius
Endotoxin/Pyrogen
Bacteria
Beach Sand
Sugar
Virus
A.C. Fine Test Dust
Granular Activated Carbon
Metal Ion
Synthetic Dye
Tobacco Smoke
Milled Flour
Latex/Emulsion
Ion Ex. Resin Bead
Pesticide
Colloidal Silica
Blue Indigo Dye
Red Blood Cells
Pollen
Herbicide
Asbestos
Human Hair
Gelatin
Coal Dust
Crypt-ospor-idium
Giardia Cyst
Mist
Process For Separation
REVERSE OSMOSIS (Hyperfiltration)
ULTRAFILTRATION
PARTICLE FILTRATION
NANOFILTRATION
MICROFILTRATION
Note: 1 Micron (1x10⁻⁶ Meters) = 4x10⁻⁵ Inches (0.00004 Inches)
1 Angstrom Unit = 10⁻¹⁰ Meters = 10⁻⁴ Micrometers (Microns)
© Copyright 1996, 1993, 1990, 1984 Osmonics, Inc., Minnetonka, Minnesot

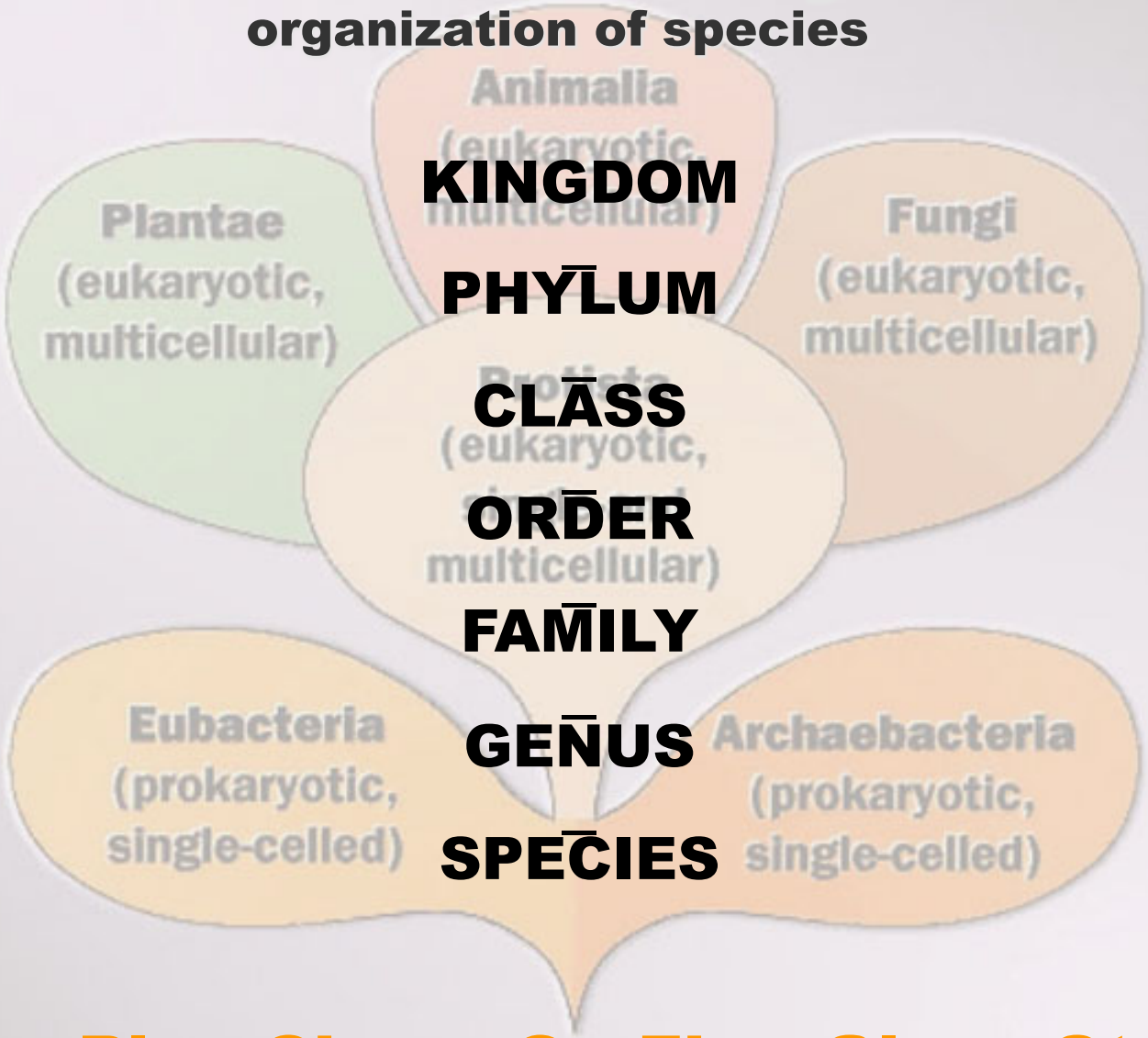
Microbes of Concern in Water & WW Treatment



- Bacteria
- Virus
- Algae
- Protozoans

Taxonomy

organization of species



KINGDOM

PHYLUM

CLASS

ORDER

FAMILY

GENUS

SPECIES

Kings Play Chess On Fine Glass Stools

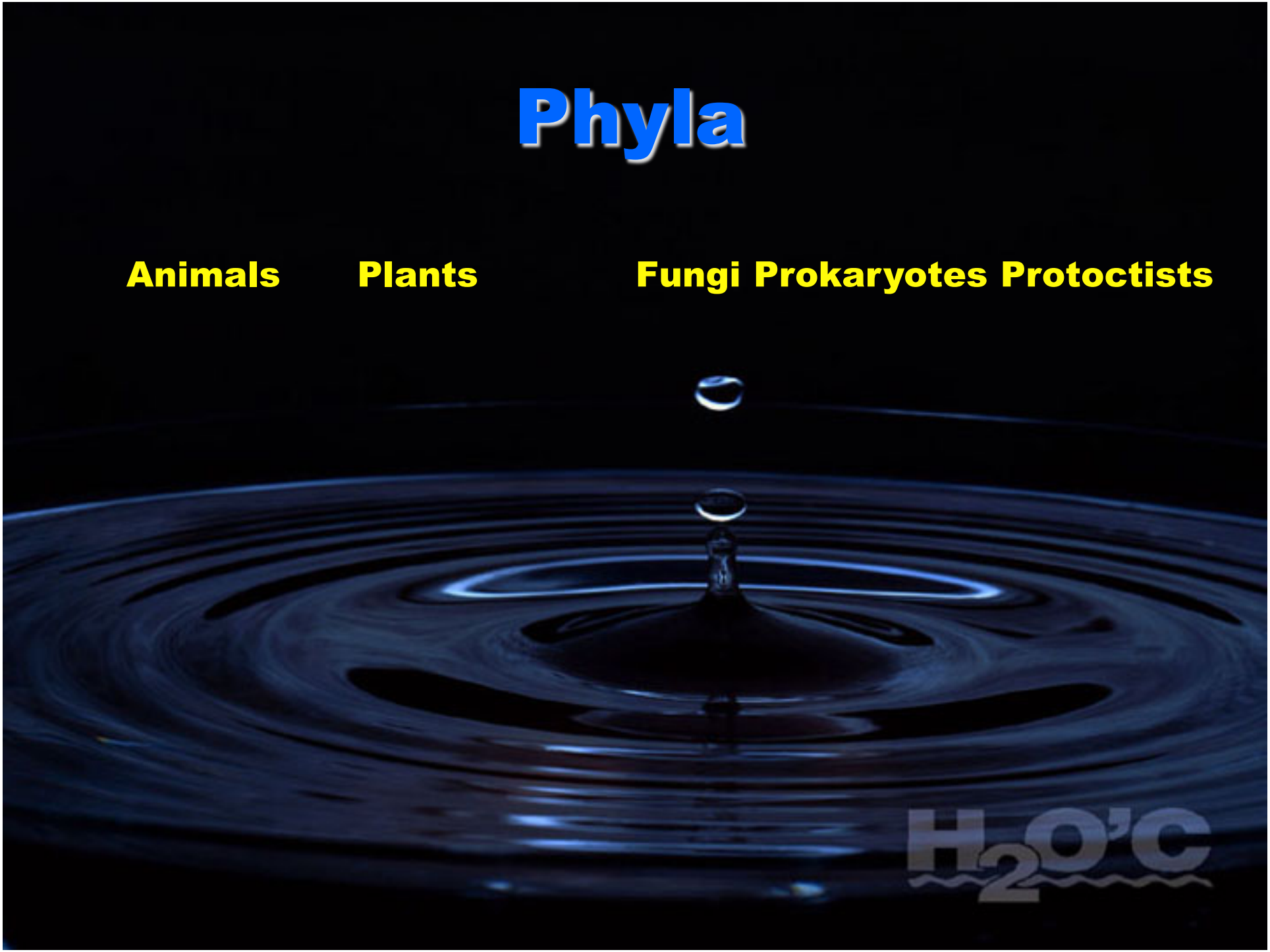
Five Kingdoms

Animals **Plants** **Fungi** **Monera** (bacteria) **Protoctists** (algae, protozoans)

Viruses are not typically considered to be living organisms due to their inability to replicate without a host cell

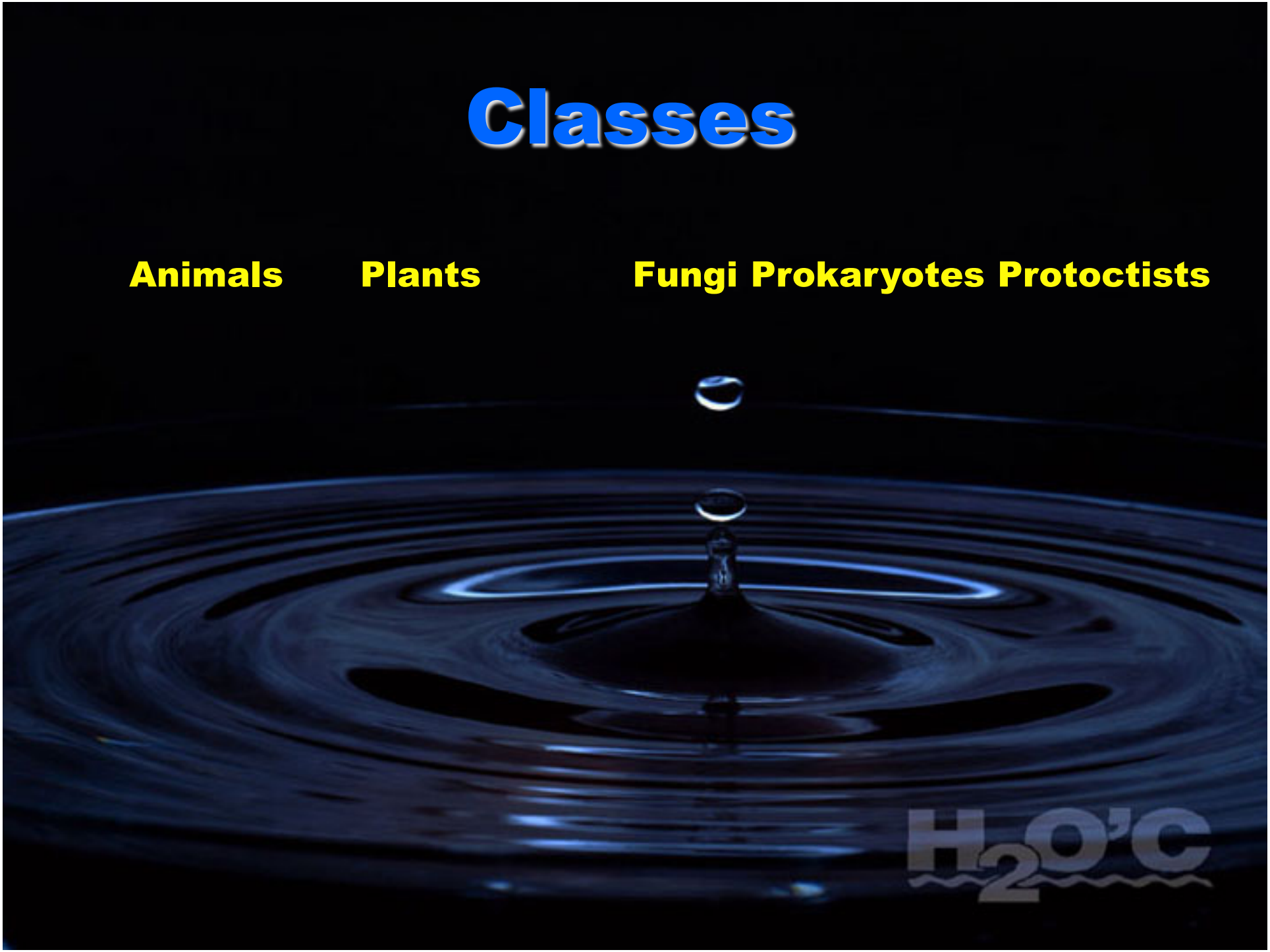
Phyla

Animals Plants Fungi Prokaryotes Protoctists



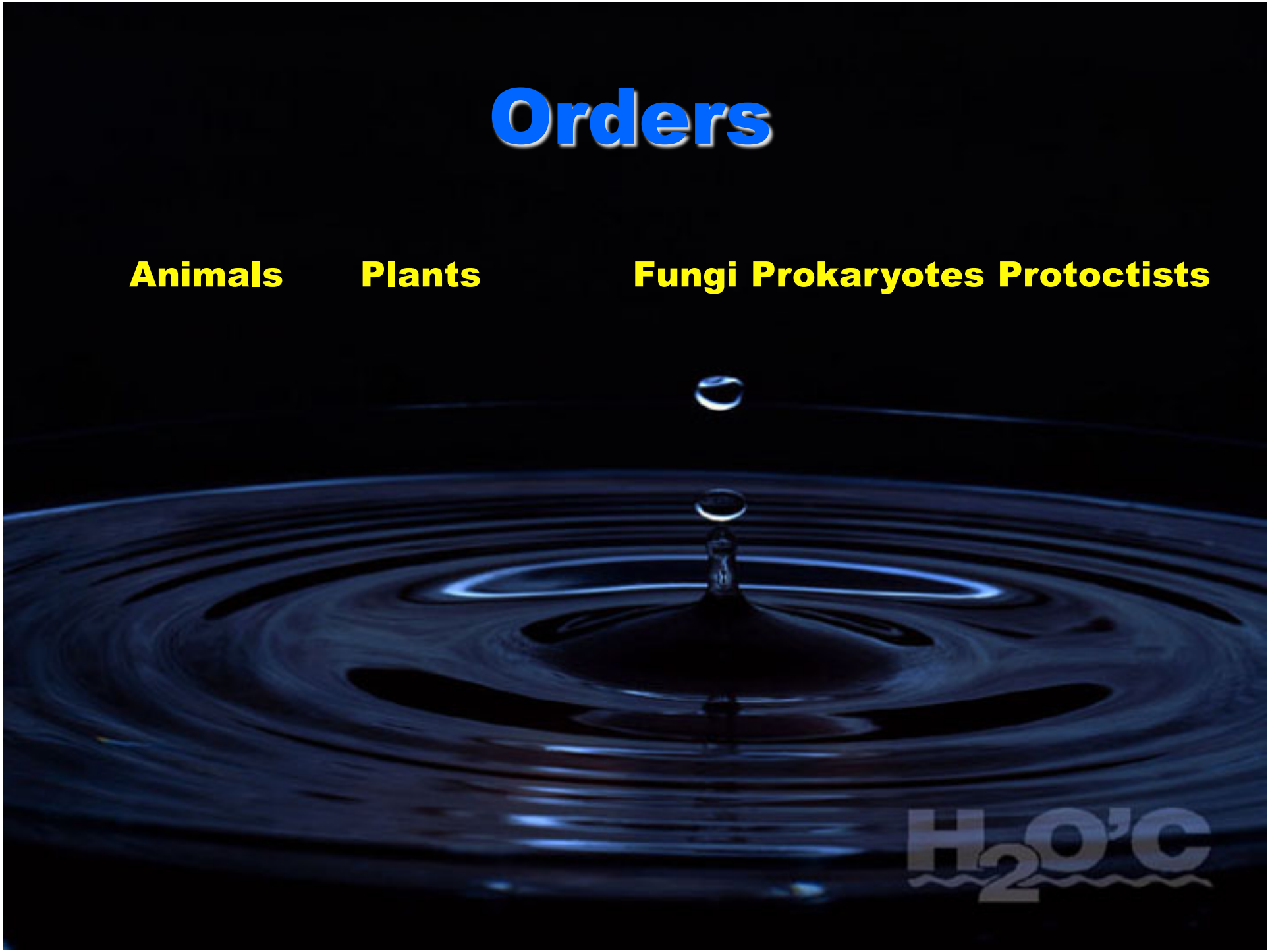
Classes

Animals **Plants** **Fungi** **Prokaryotes** **Protoctists**



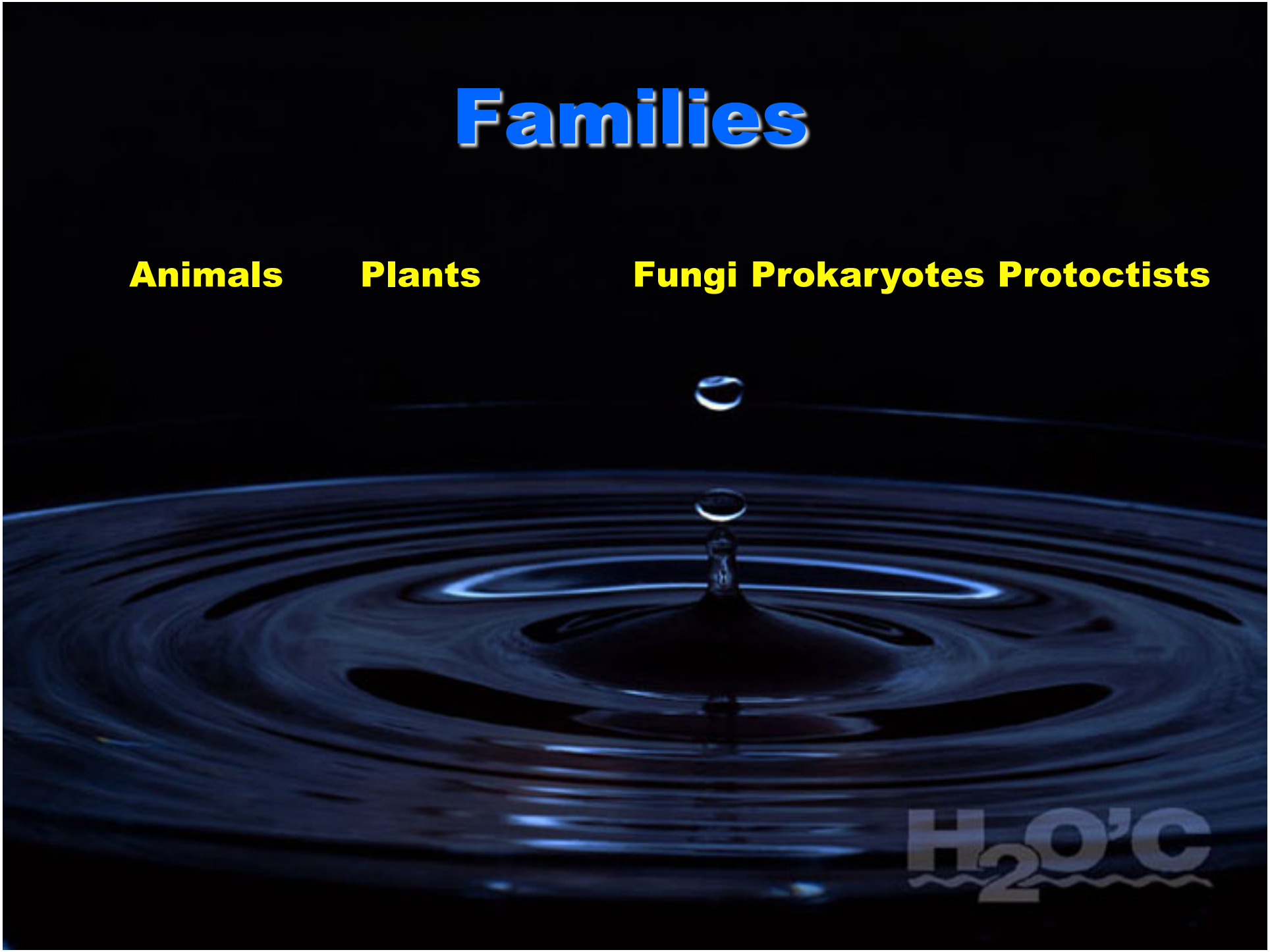
Orders

Animals **Plants** **Fungi** **Prokaryotes** **Protoctists**



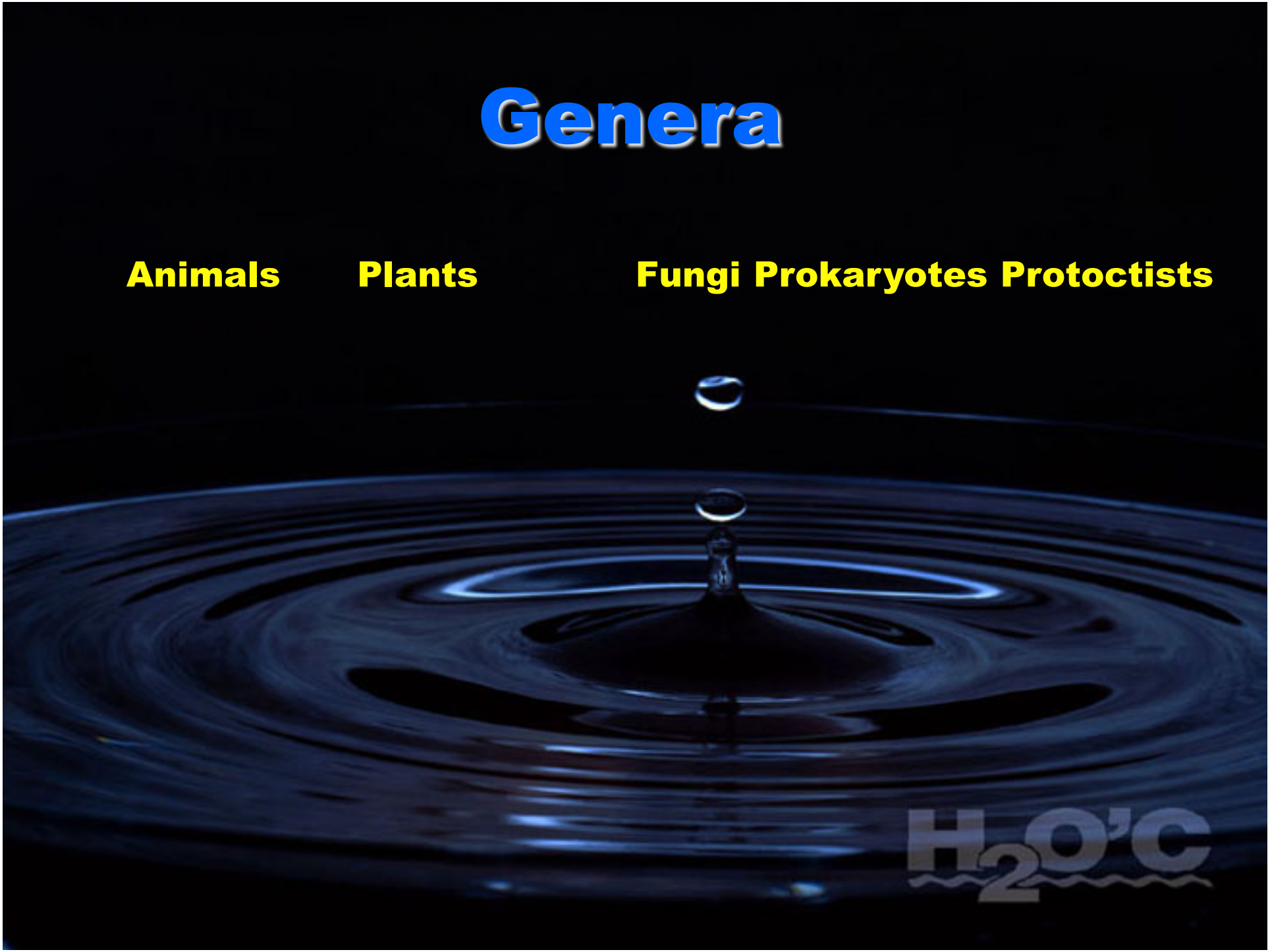
Families

Animals **Plants** **Fungi** **Prokaryotes** **Protoctists**



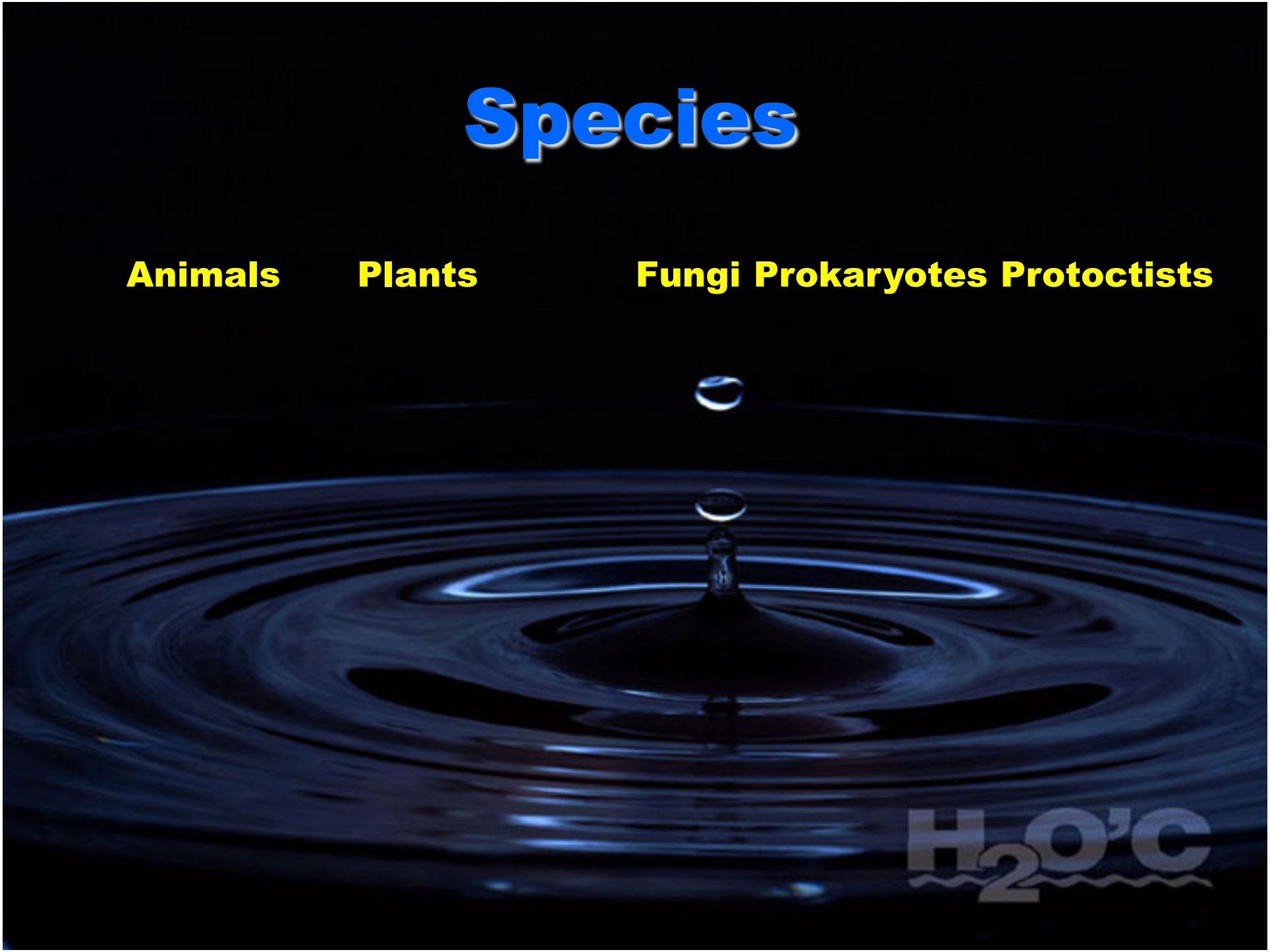
Genera

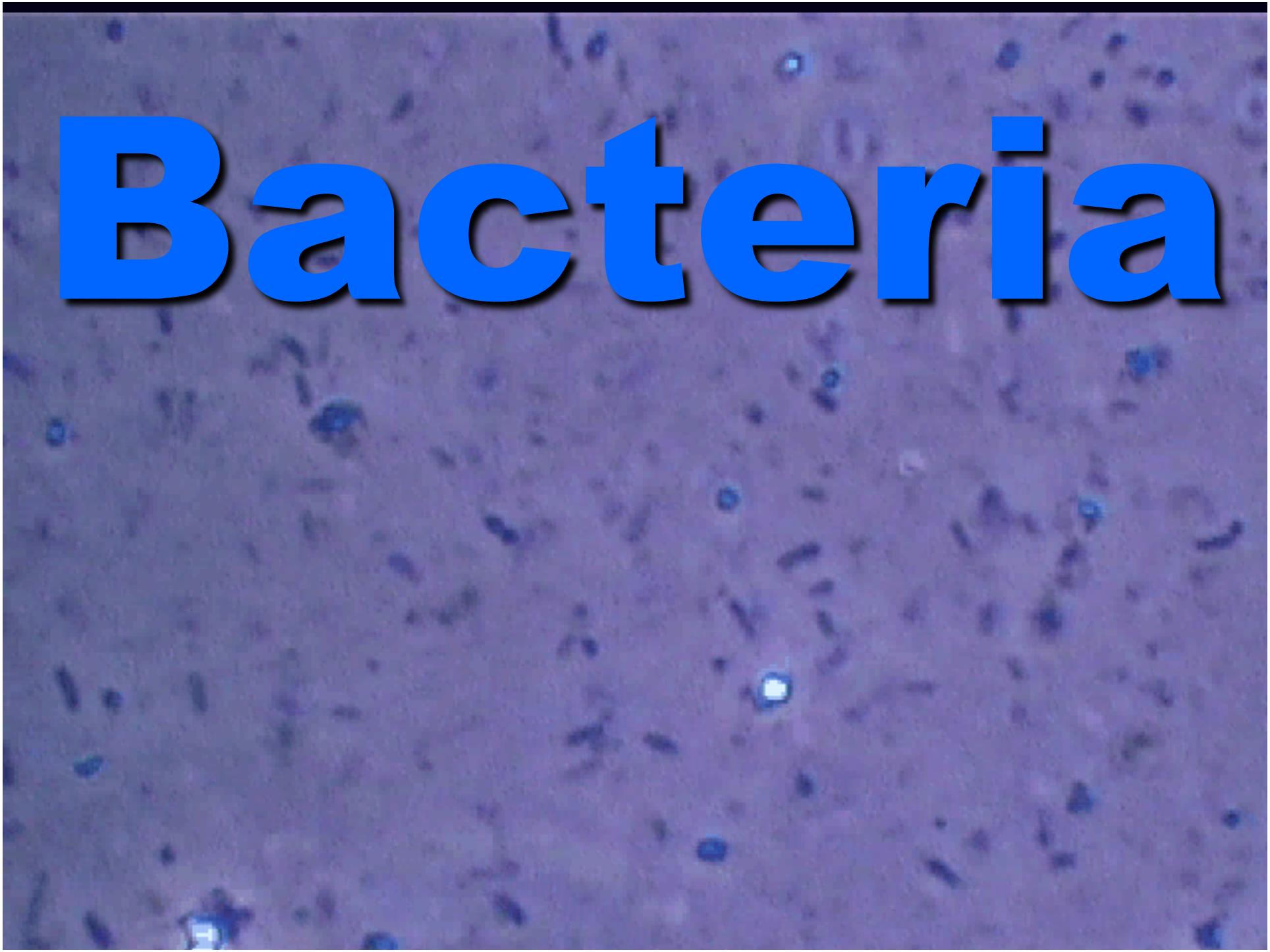
Animals Plants Fungi Prokaryotes Protoctists



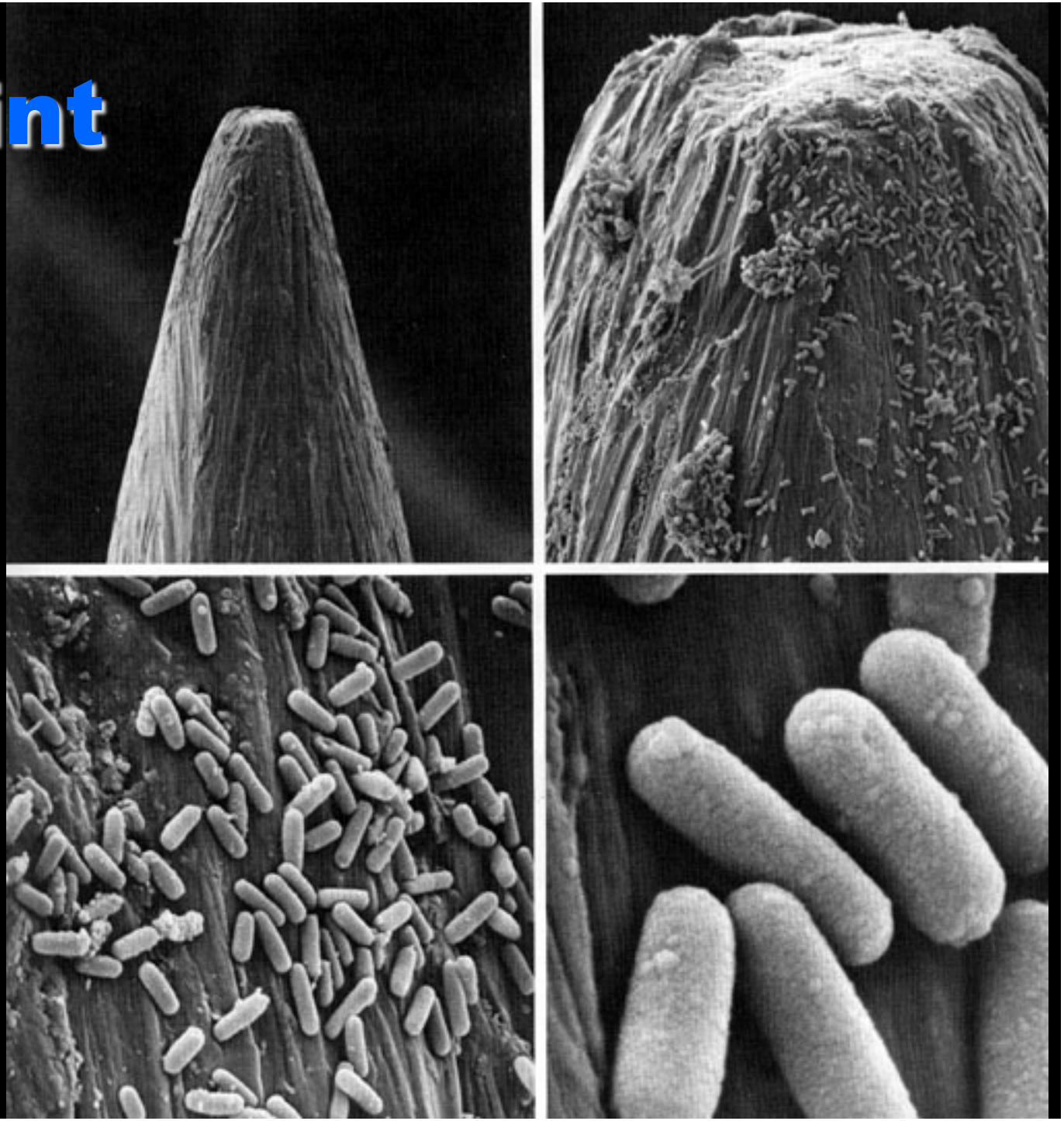
Species

Animals **Plants** **Fungi Prokaryotes Protoctists**




Bacteria

Pin Point



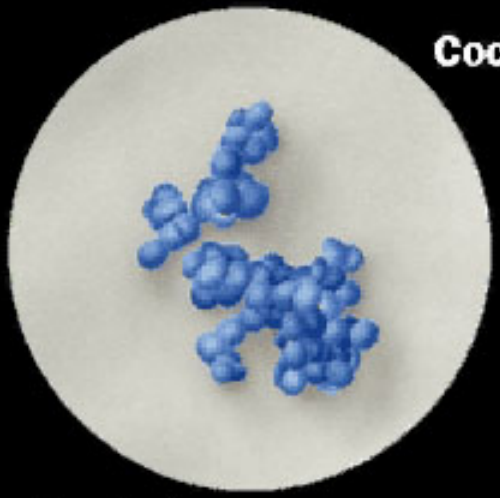
Classification of Bacteria

Bergey's Manual

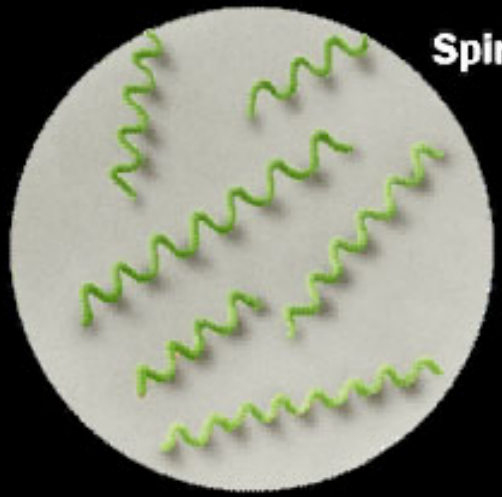
- Archaebacteria
  - Aerobic
  - Anaerobic
- Eubacteria
- Autotrophic bacteria
  - Phototrophic
    - Purple bacteria
    - Green bacteria
  - Chemotrophic
    - Nitrifiers
    - Sulfur oxidizers
    - Fe / Mn oxidizers
    - Methane oxiders
- Heterotrophic bacteria
  - Gram-negative
    - Aerobic
    - Facultatively anaerobic
    - Anaerobic
  - Gram-positive
  - Mycobacteria
- Bacteria with complex structures
  - Actinomycetes
  - Stalked and budding bacteria
  - Sheathed bacteria
  - Gliding and creeping bacteria
  - Spirochaetes
  - Mycoplasmas

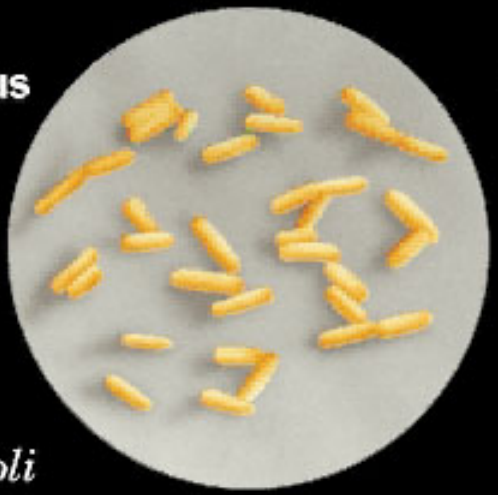
Morphology

Fancy Word for 'Shape'

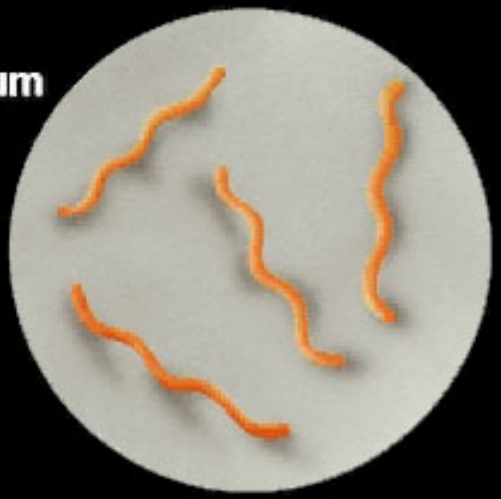


Staphylococcus aureus

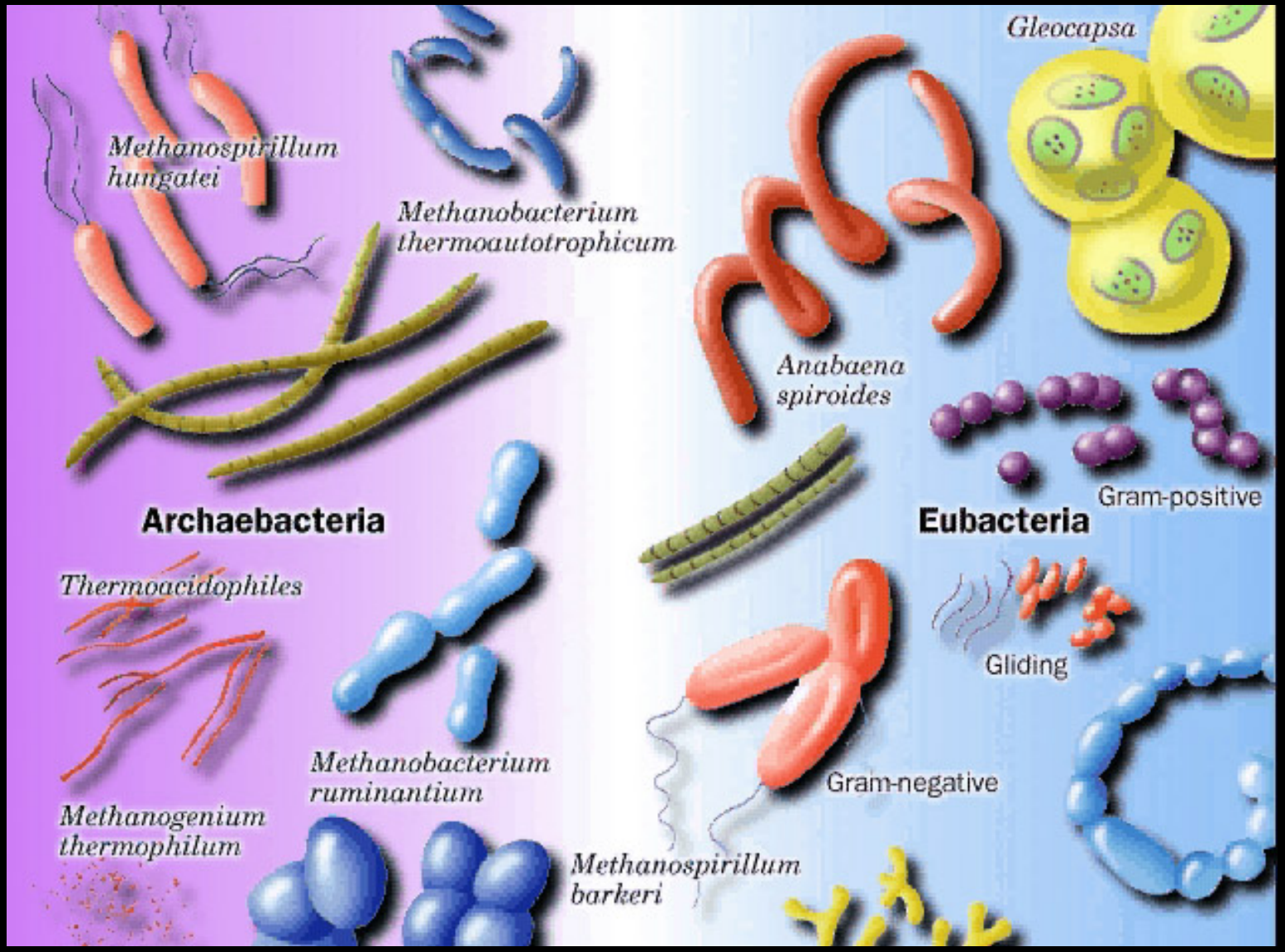




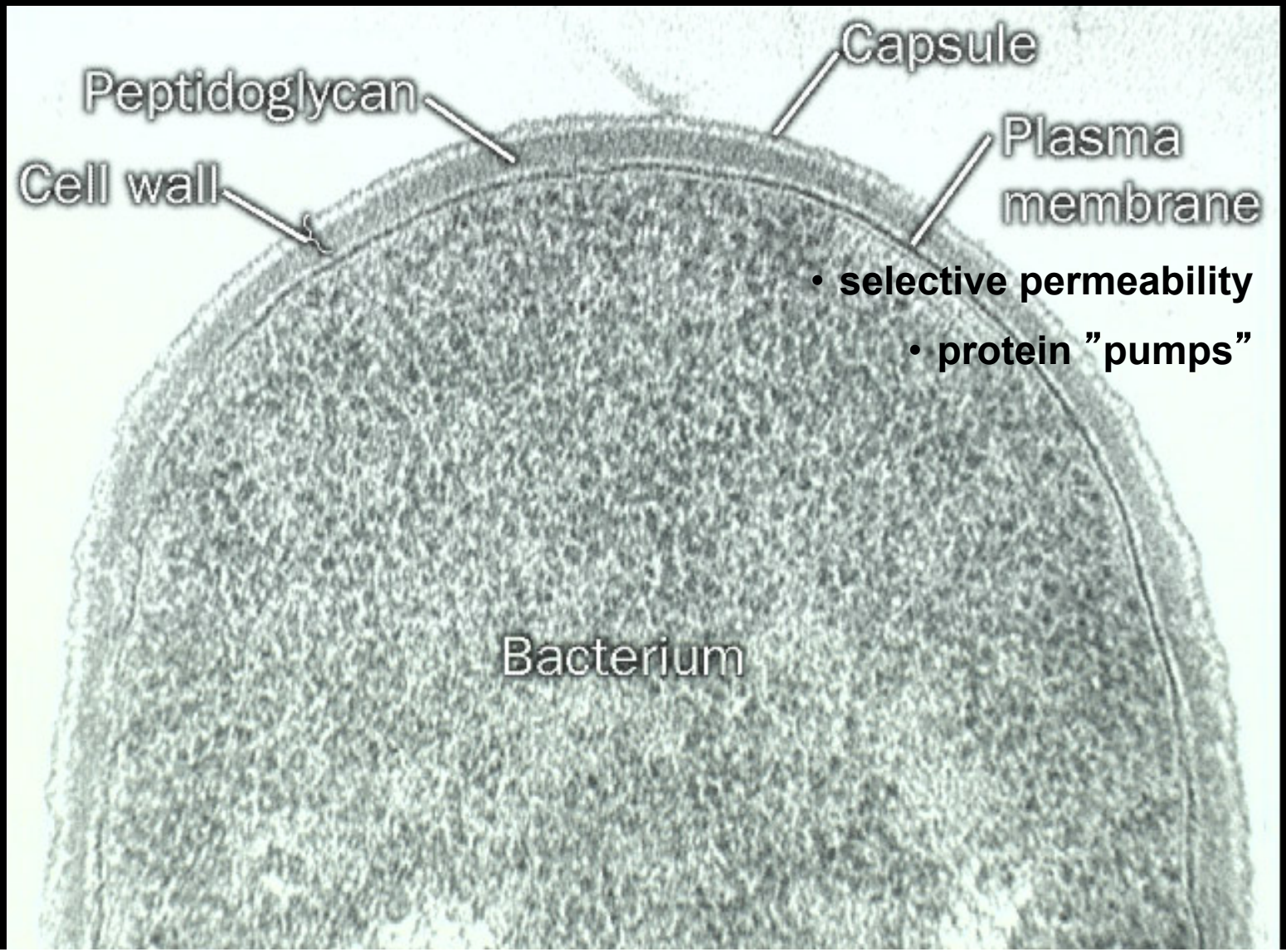
E. coli



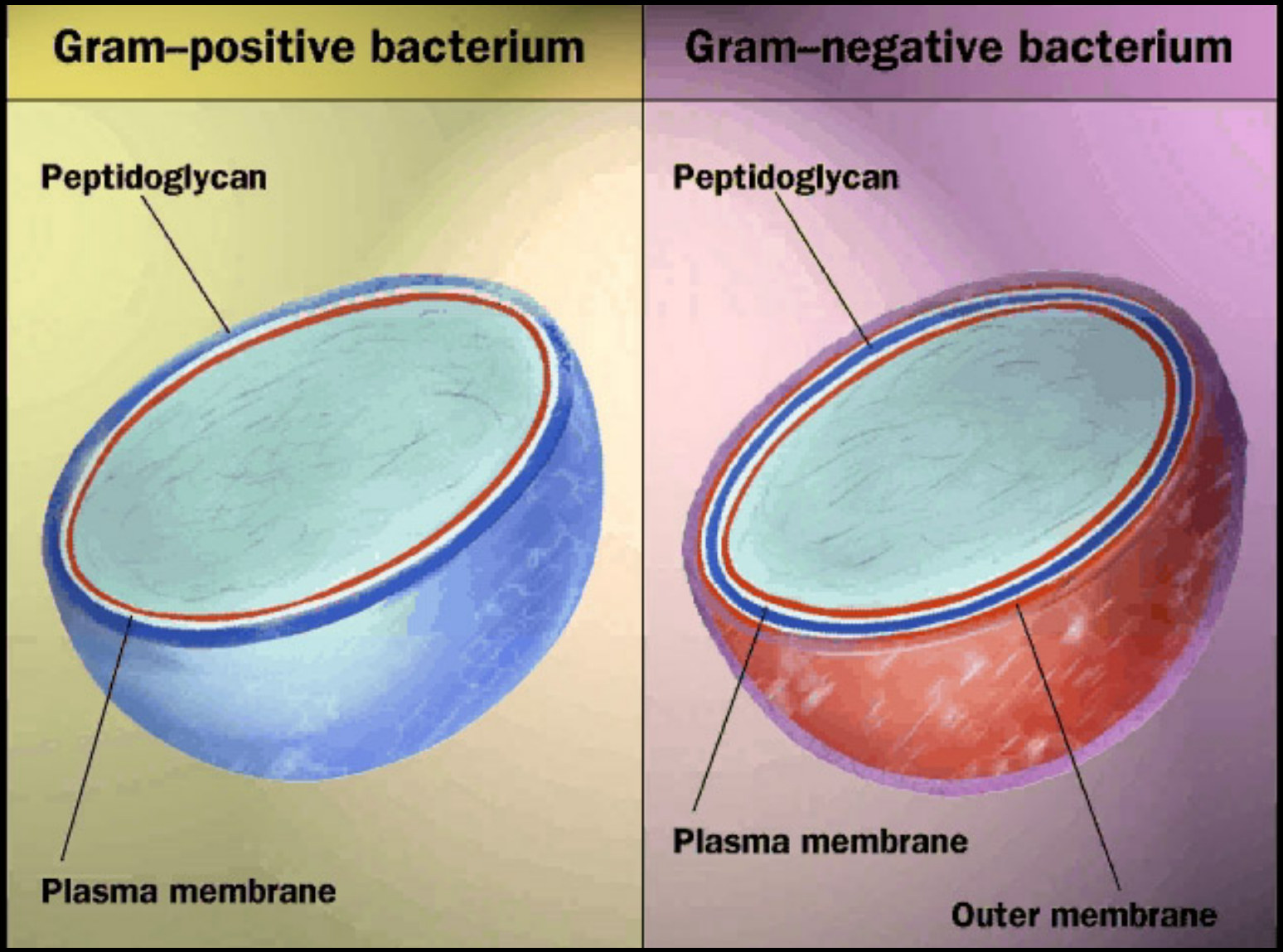
Types of Bacteria



Cell Wall


Capsule
Peptidoglycan
Plasma membrane
Cell wall
• selective permeability
• protein ”pumps”
Bacterium

Gram-positive/negative



Food

Autotrophs (self-nourishing)

require water, CO_2 , inorganic salts, energy source

Heterotrophs

saprophytic—absorb nutrients through cell membrane

holozoic—eat, digest, and absorb particulate food

Oxygen

Aerobes

utilize oxygen in respiration

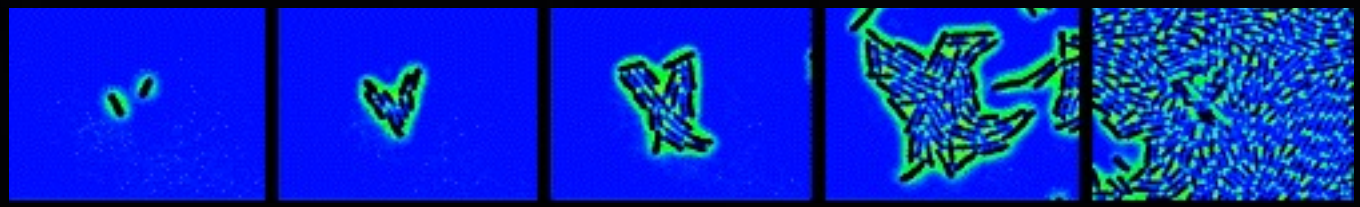
Obligate Anerobes

quickly killed by oxygen

Facultative Anerobes

can take it or leave it

Reproduction



Asexually (fission)

a cell divides into two cells

Kinetics (speed)

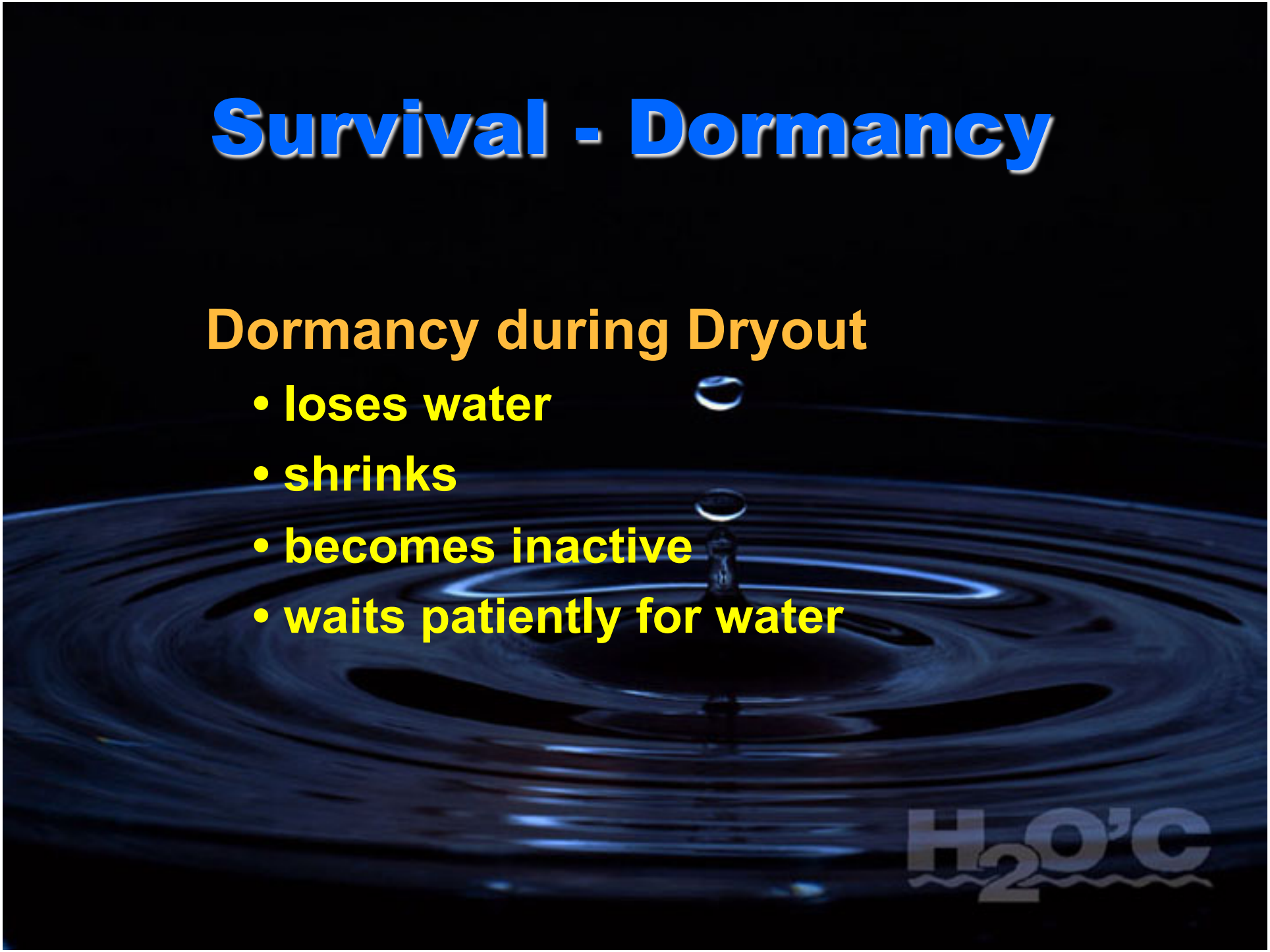
Cells can divide every 20 minutes

One cell ⊥ 8 hours ⊥ 12,000,000 cells

Inhibitors

- lack of food
- accumulation of waste products

Survival - Dormancy

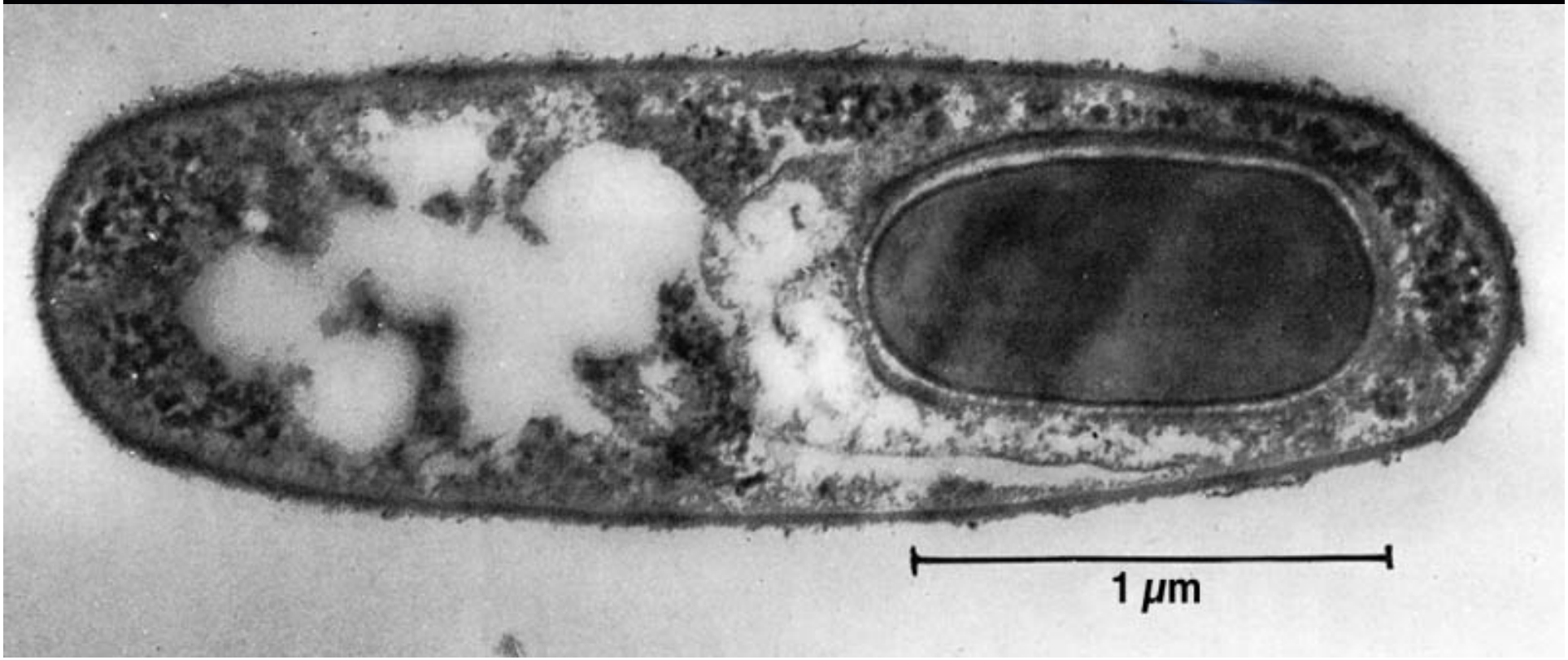


Dormancy during Dryout

- loses water
- shrinks
- becomes inactive
- waits patiently for water

Survival - Spores

- cell retreats in times of unfavorable conditions (dryness, temp, disinfectant)
- forms new, thicker cell wall within old one
- when favorable conditions reappear, spore absorbs water, breaks out of inner shell, returns to normal
- Anthrax bacilli can survive 30 years in spore form
- most pathogenic (disease-causing) bacteria do not form spores

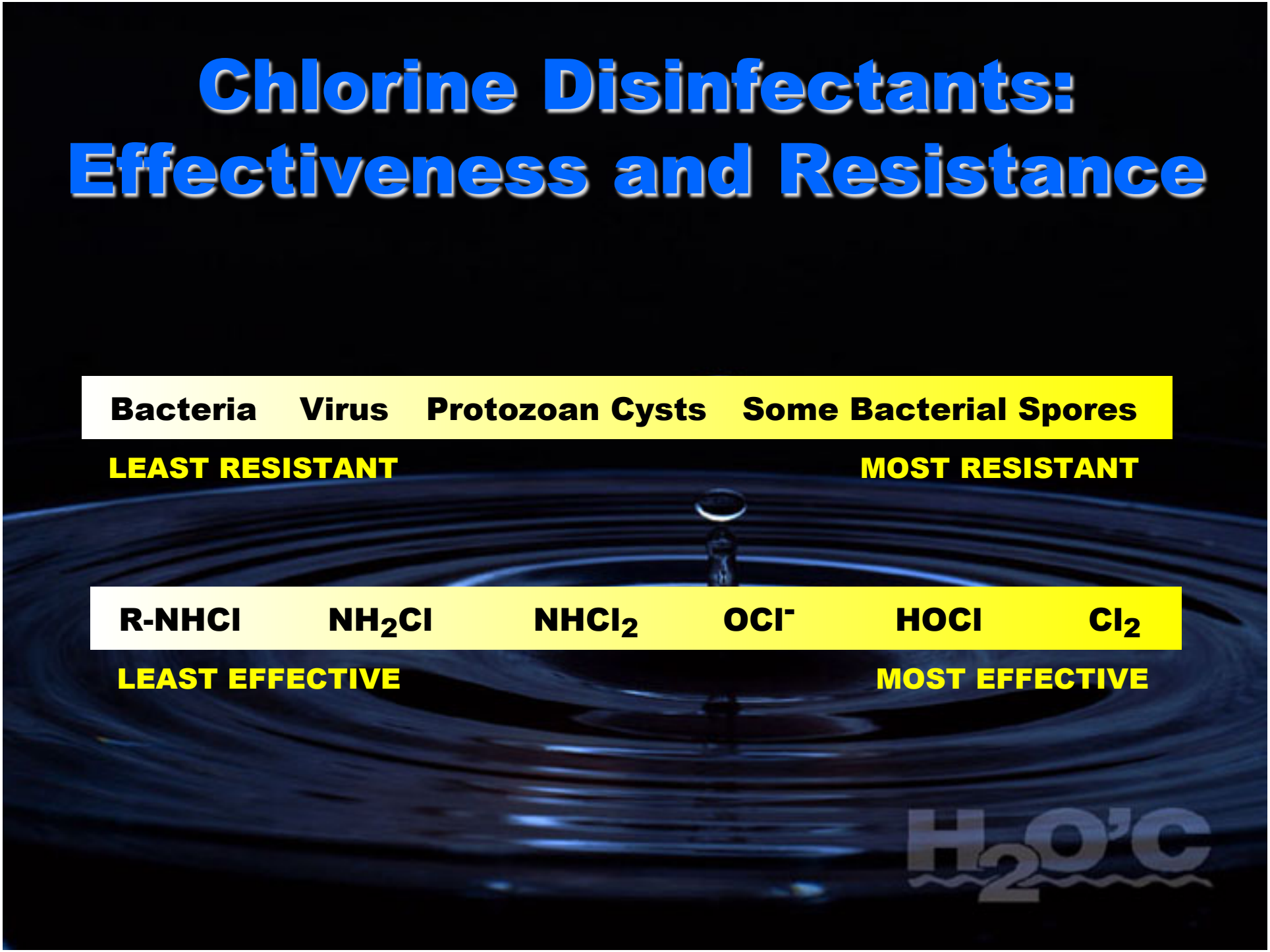


Ailments
Associated with Bacteria

typhoid fever	⊥	Bacillus typhosus
diarrhea	⊥	Escherichia coli
Legionnaire's disease	⊥	Legionella
Leptospirosis	⊥	Leptospirea
salmonellosis, paratyphoid	⊥	Salmonella
bacillary dysentary (Shigellosis)	⊥	Shigella
cholera	⊥	Vibrio cholerae
plague	⊥	Yersinia

There are over 3.000 species of bacteria;
only a handful are pathogenic (disease-causing)

Chlorine Disinfectants: Effectiveness and Resistance



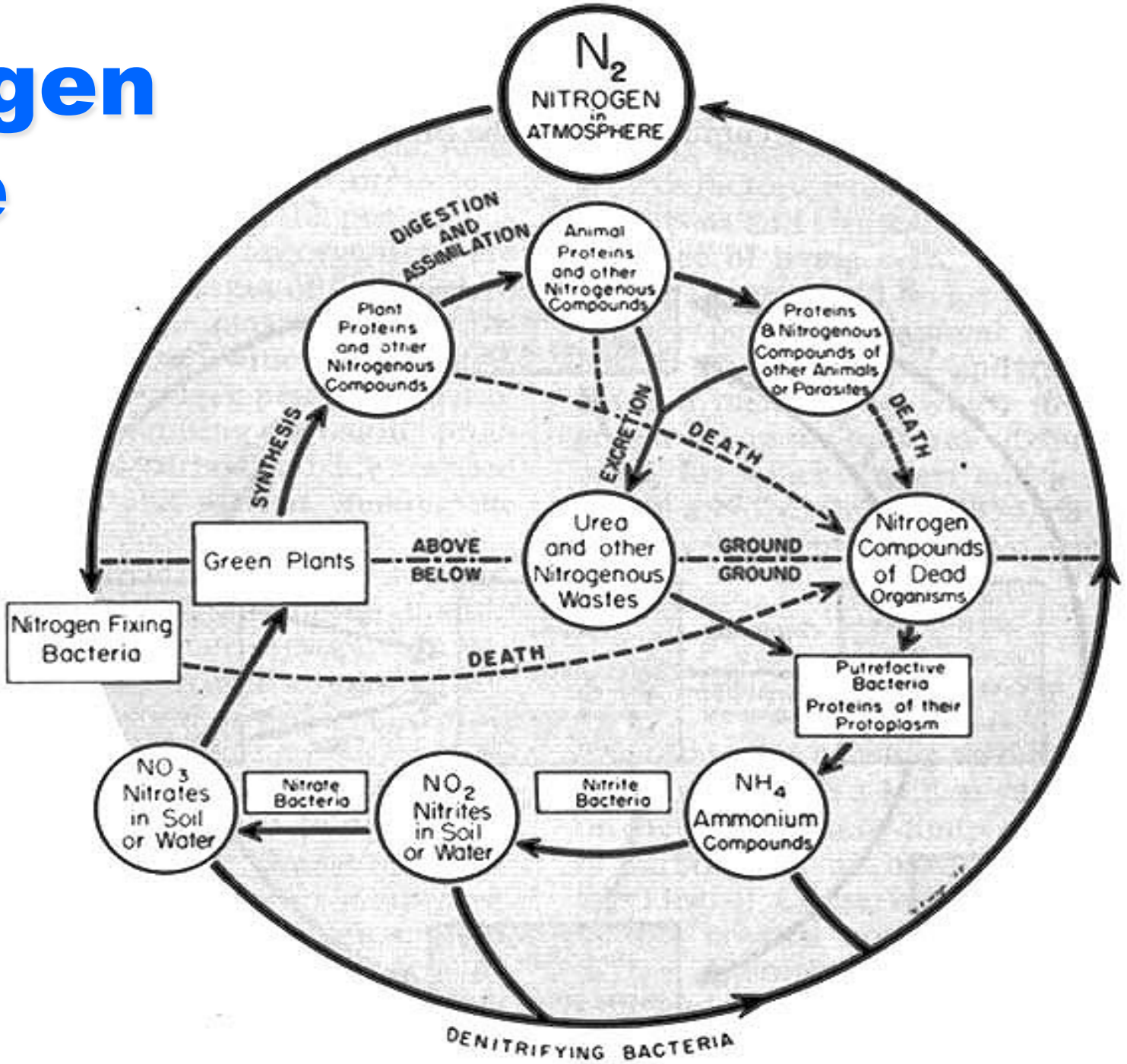
Bacterial Jobs

Fermentation: enzymatic anaerobic breakdown of carbohydrates

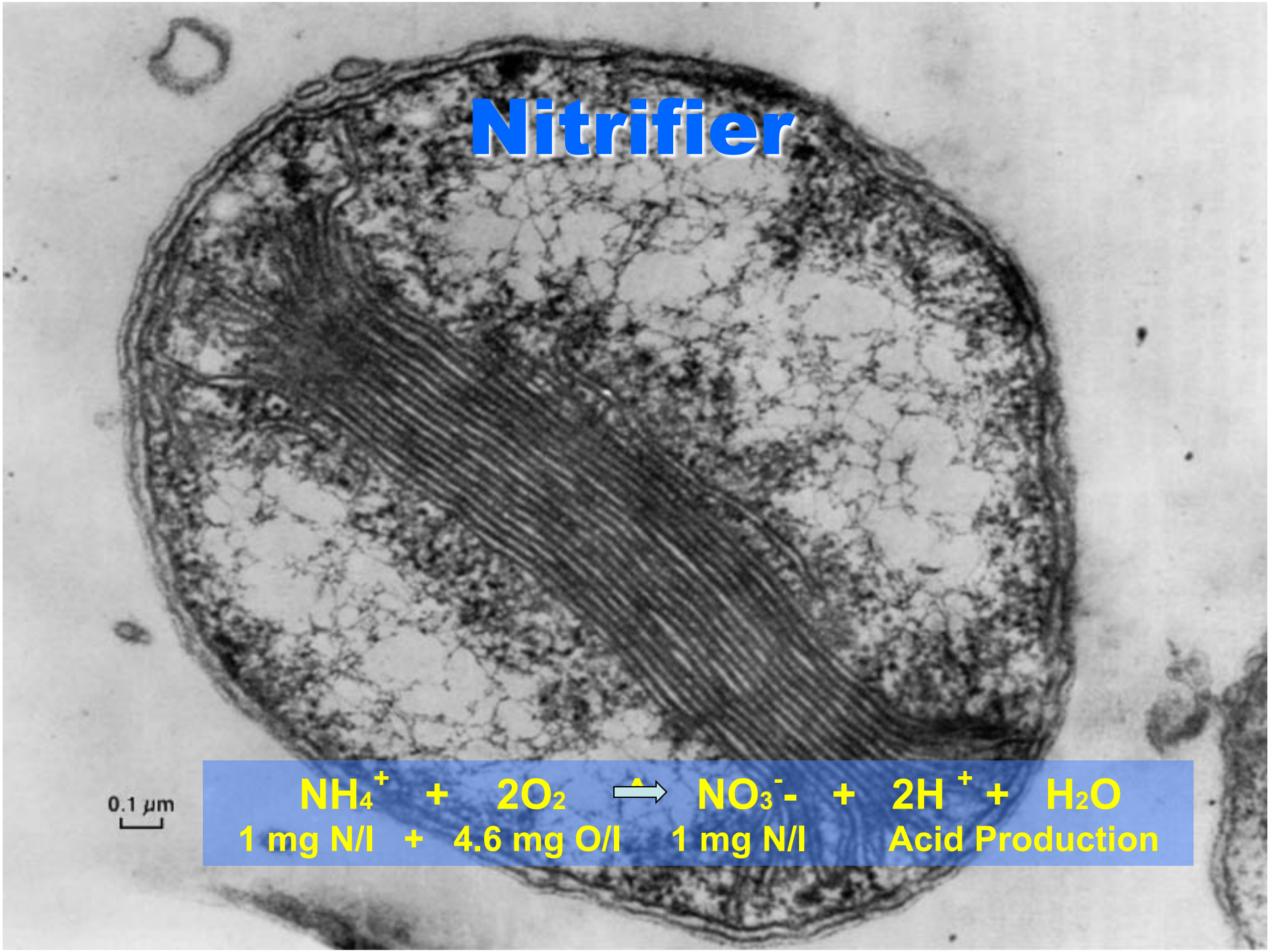
Putrefaction: enzymatic anaerobic breakdown of proteins and amino acids

Stench—nitrogen-and sulfur-containing compounds produced during putrefaction

Nitrogen Cycle



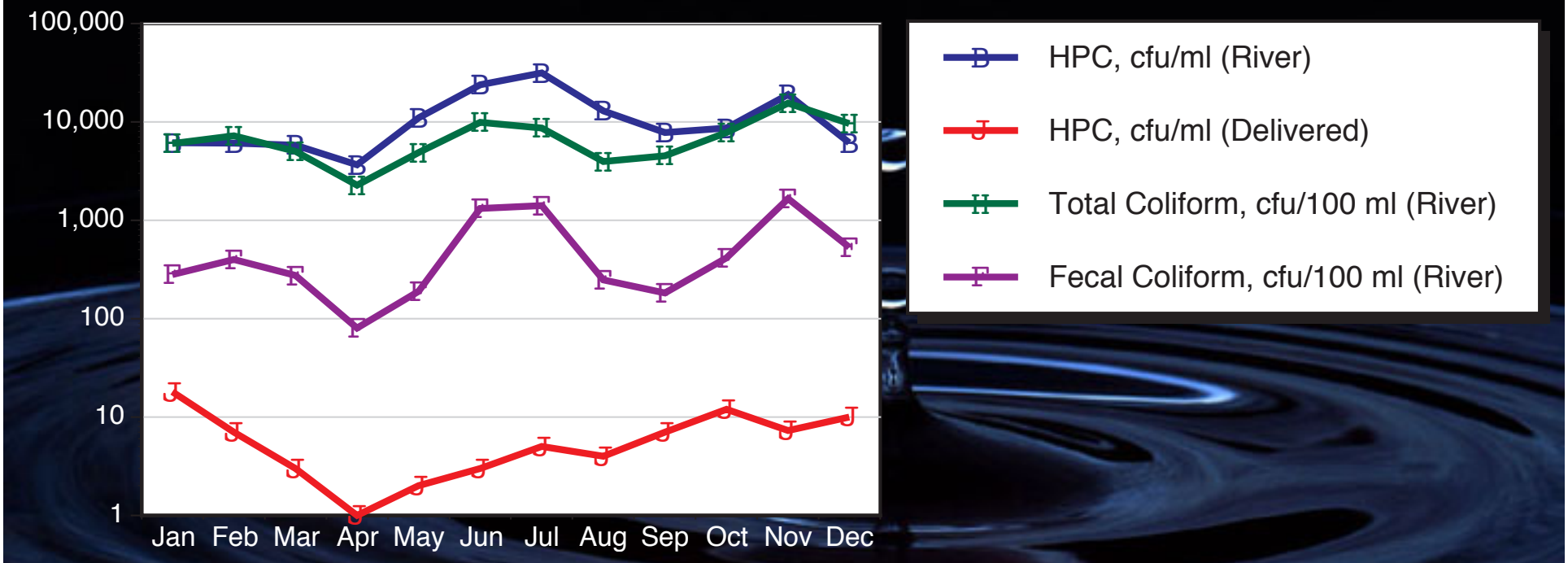
Nitrifier



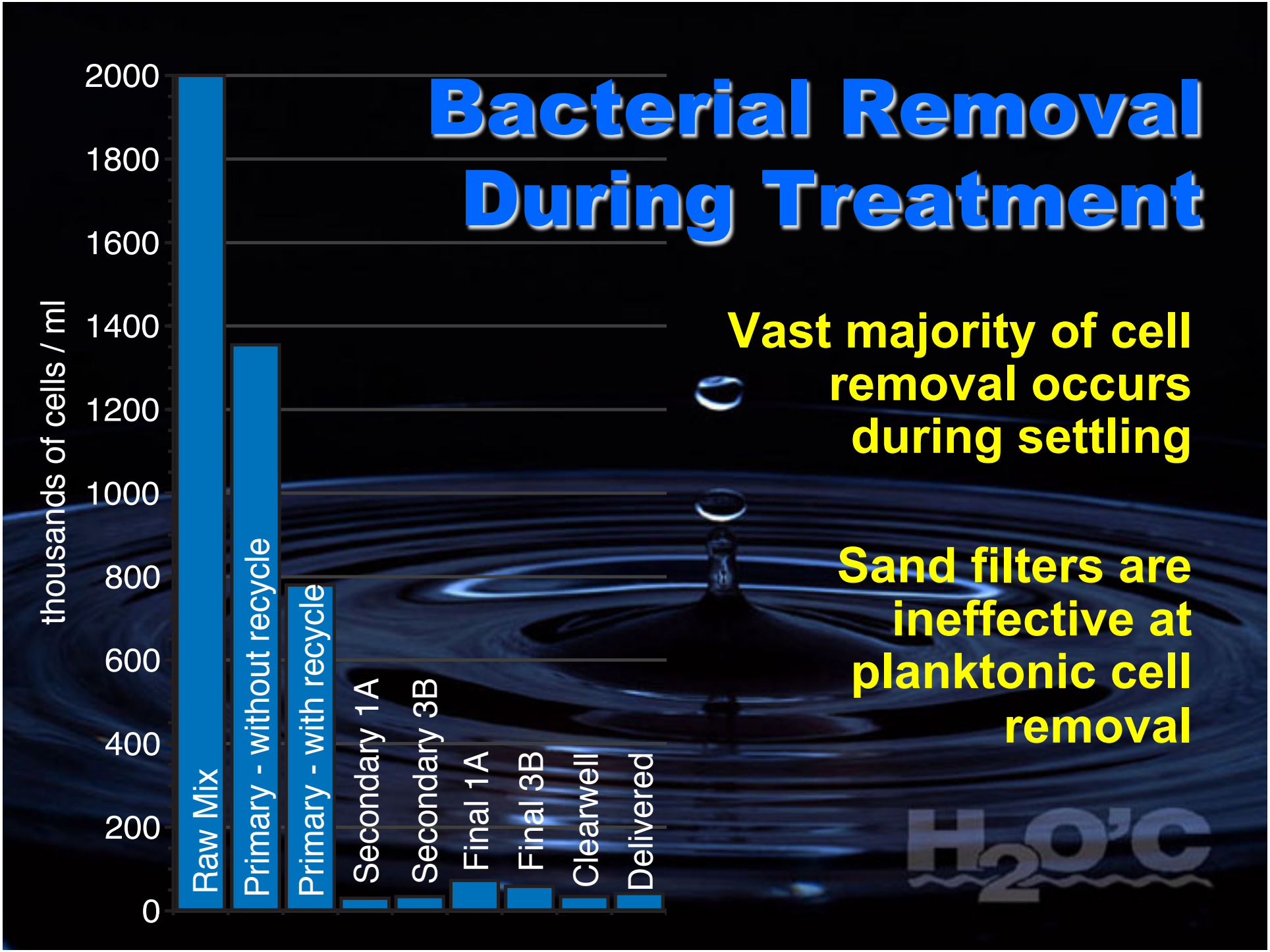
$$NH_4^+ + 2O_2 \Rightarrow NO_3^- + 2H^+ + H_2O$$

1 mg N/l + 4.6 mg O/l → 1 mg N/l + Acid Production

HPC & Coliform



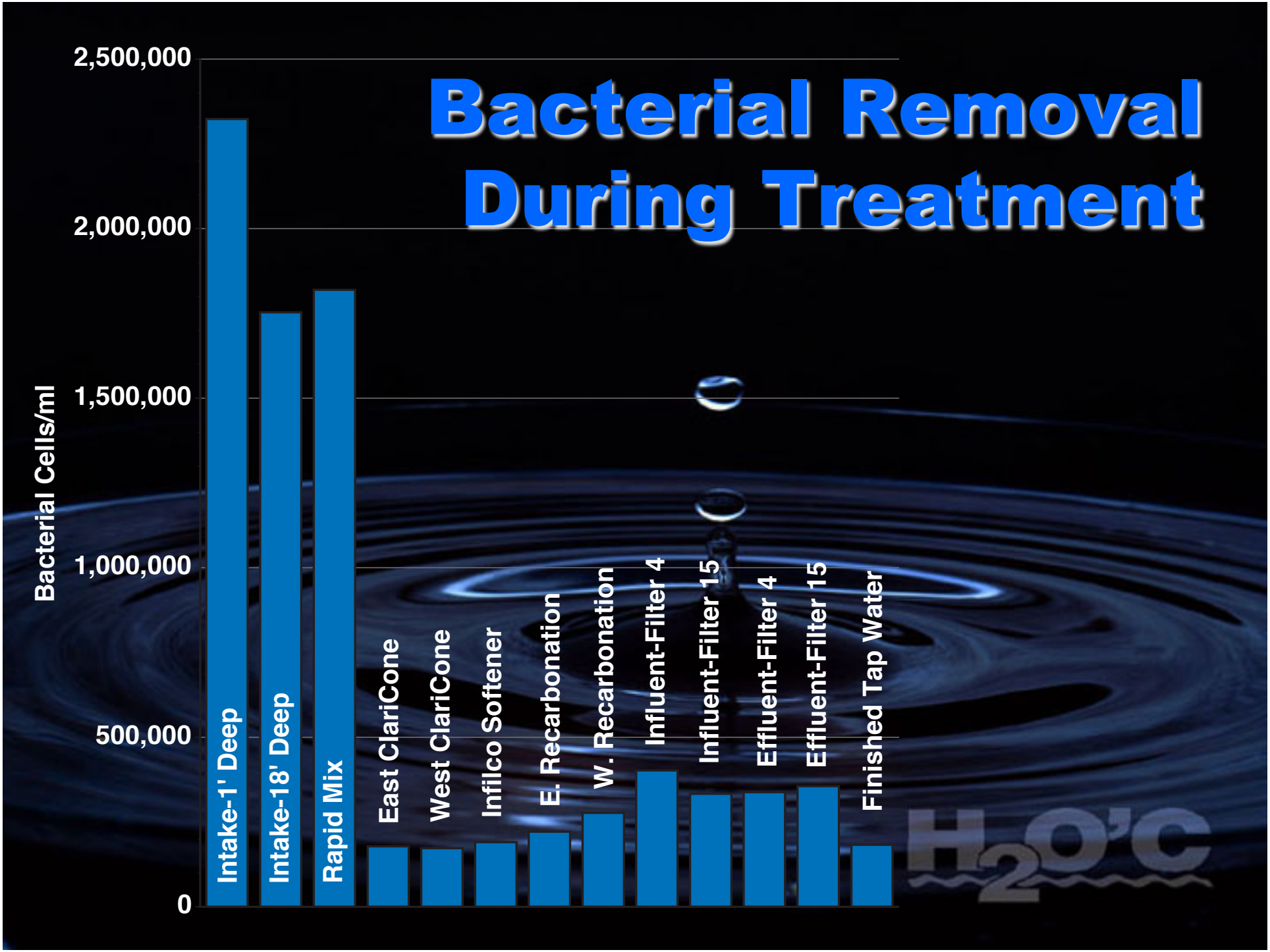
Bacterial Removal During Treatment



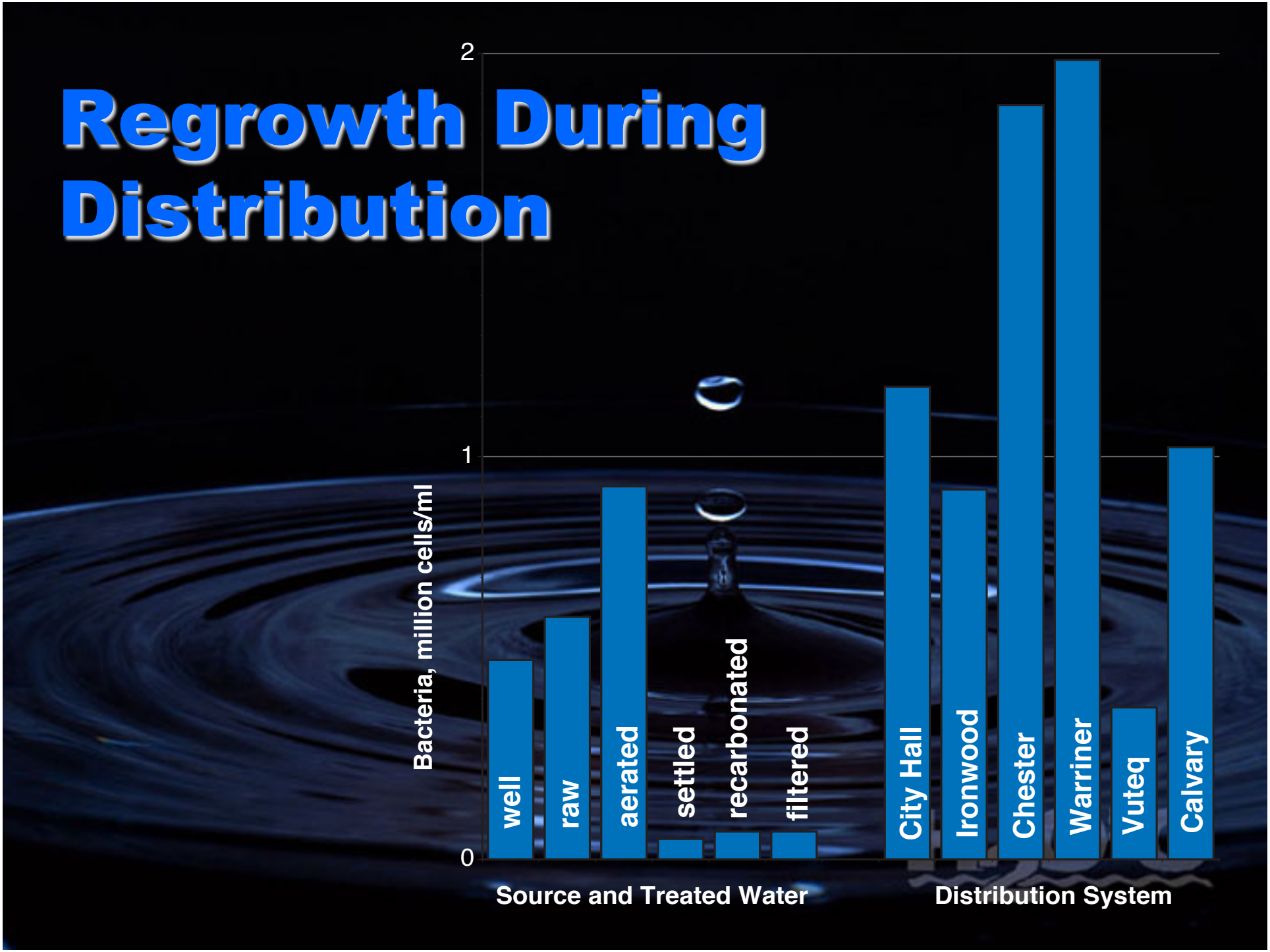
Vast majority of cell removal occurs during settling

Sand filters are ineffective at planktonic cell removal

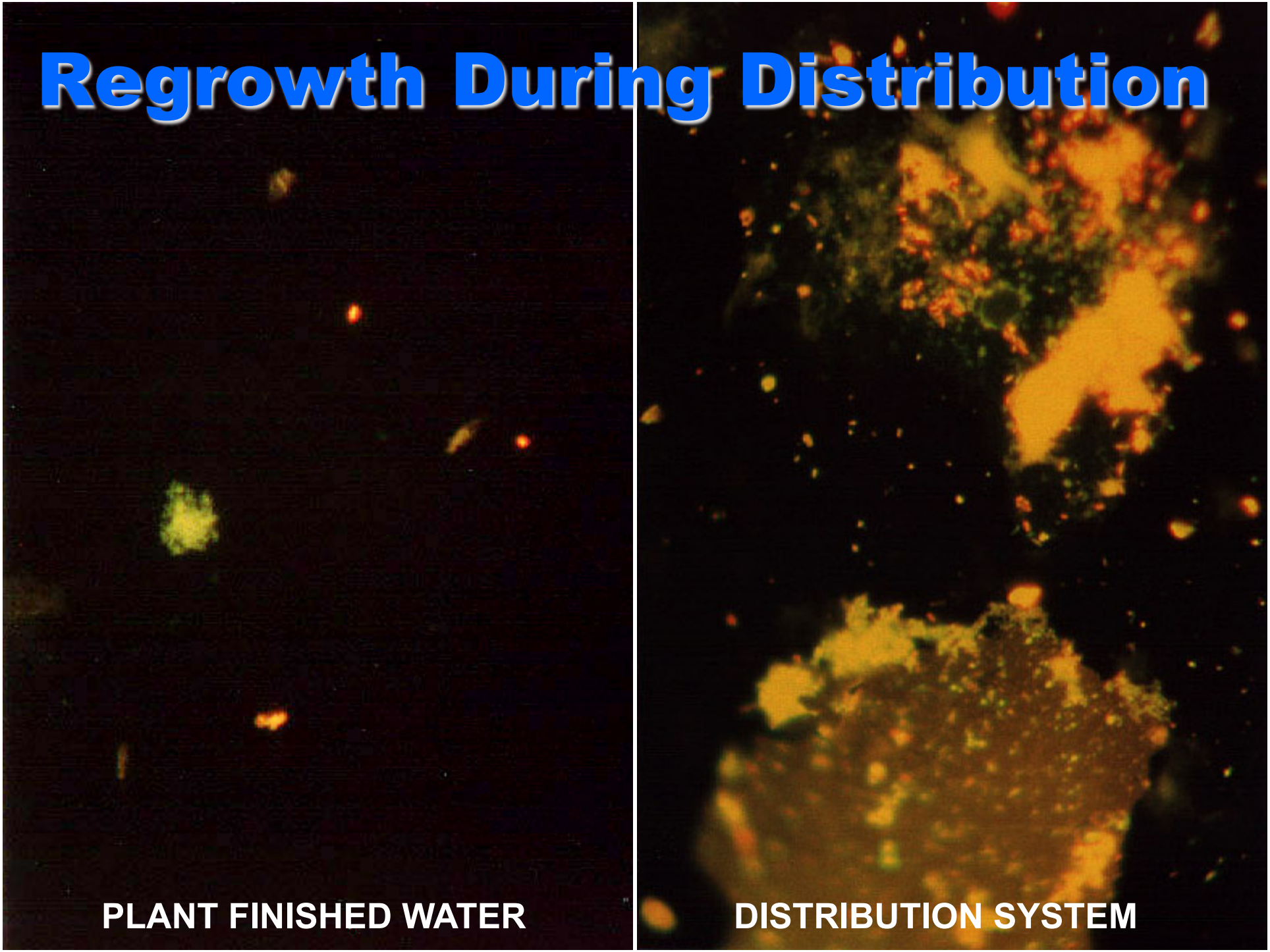
$H_2O'C$


Bacterial Removal During Treatment
Bacterial Cells/ml
2,500,000
2,000,000
1,500,000
1,000,000
500,000
0
Intake-1' Deep
Intake-18' Deep
Rapid Mix
East ClariCone
West ClariCone
Infilco Softener
E. Recarbonation
W. Recarbonation
Influent-Filter 4
Influent-Filter 15
Effluent-Filter 4
Effluent-Filter 15
Finished Tap Water
H2O'C

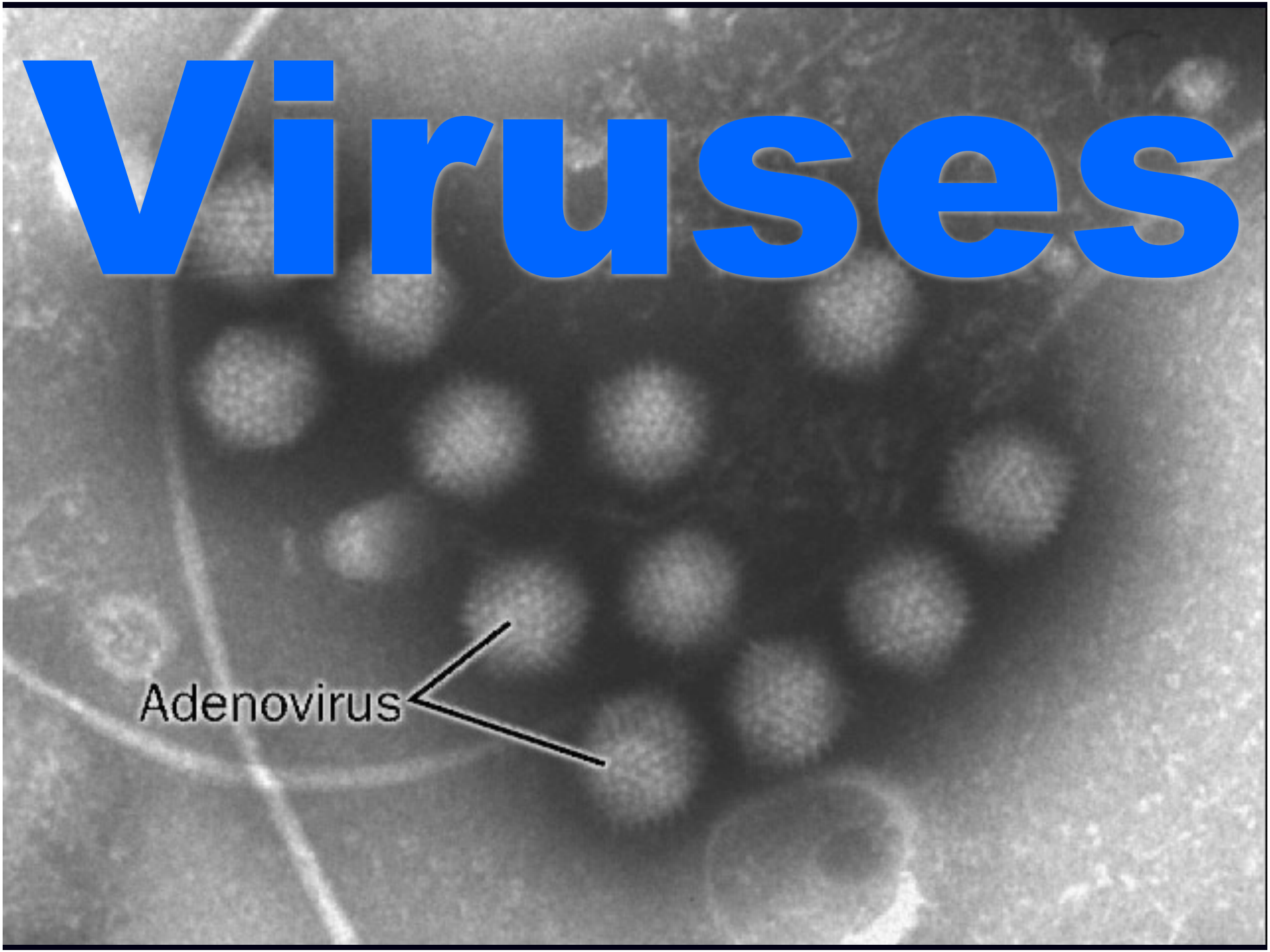
Regrowth During Distribution



Regrowth During Distribution



Viruses



Viruses

	Virus	Nucleic Acid Type	Shape	Size (nm) (or diameter × length)
Animal Viruses	Vaccinia	DNA		230 × 300
	Mumps	RNA		150 × 300
	Herpes	DNA		100 × 200
	Influenza	RNA		80 × 120
	Adenovirus	DNA		70 × 90
	Poliovirus	RNA		28
Plant Viruses	Wound tumor	RNA		55 × 60
	Tobacco mosaic	RNA		18 × 300
	Potato X	RNA		10 × 500
Bacterial Phages	T phage	DNA		65 × 200
	ϕX174	DNA		25

[a] Adapted from H. Lechevalier and D. Pramer (1971), *The Microbes*. J. B. Lippincott Co., Philadelphia, Pa., and R. W. Horne (1963), "The Structure of Viruses," *Scientific American*.

Viruses - Plant and Bacterial

Wound tumor	RNA		55 × 60
Tobacco mosaic	RNA		18 × 300
Potato X	RNA		10 × 500
T phage	DNA		65 × 200
ϕX174	DNA		25

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Viruses - Animal

Virus	*Nucleic Acid Type*	*Shape*	*Size (nm) (or diameter × length)*
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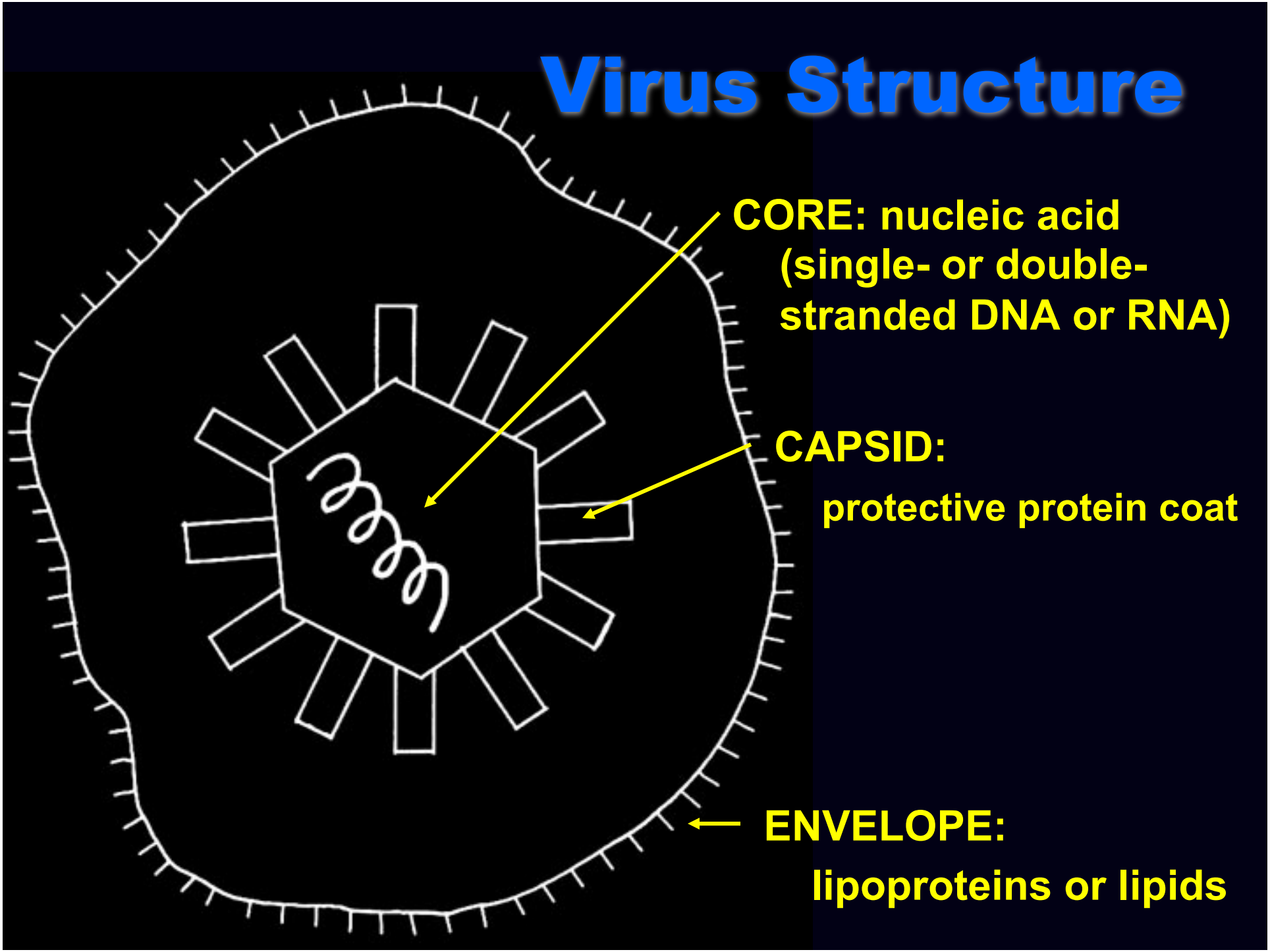
Herpes	DNA		100 × 200
Influenza	RNA		80 × 120
Adenovirus	DNA		70 × 90
Poliovirus	RNA		28

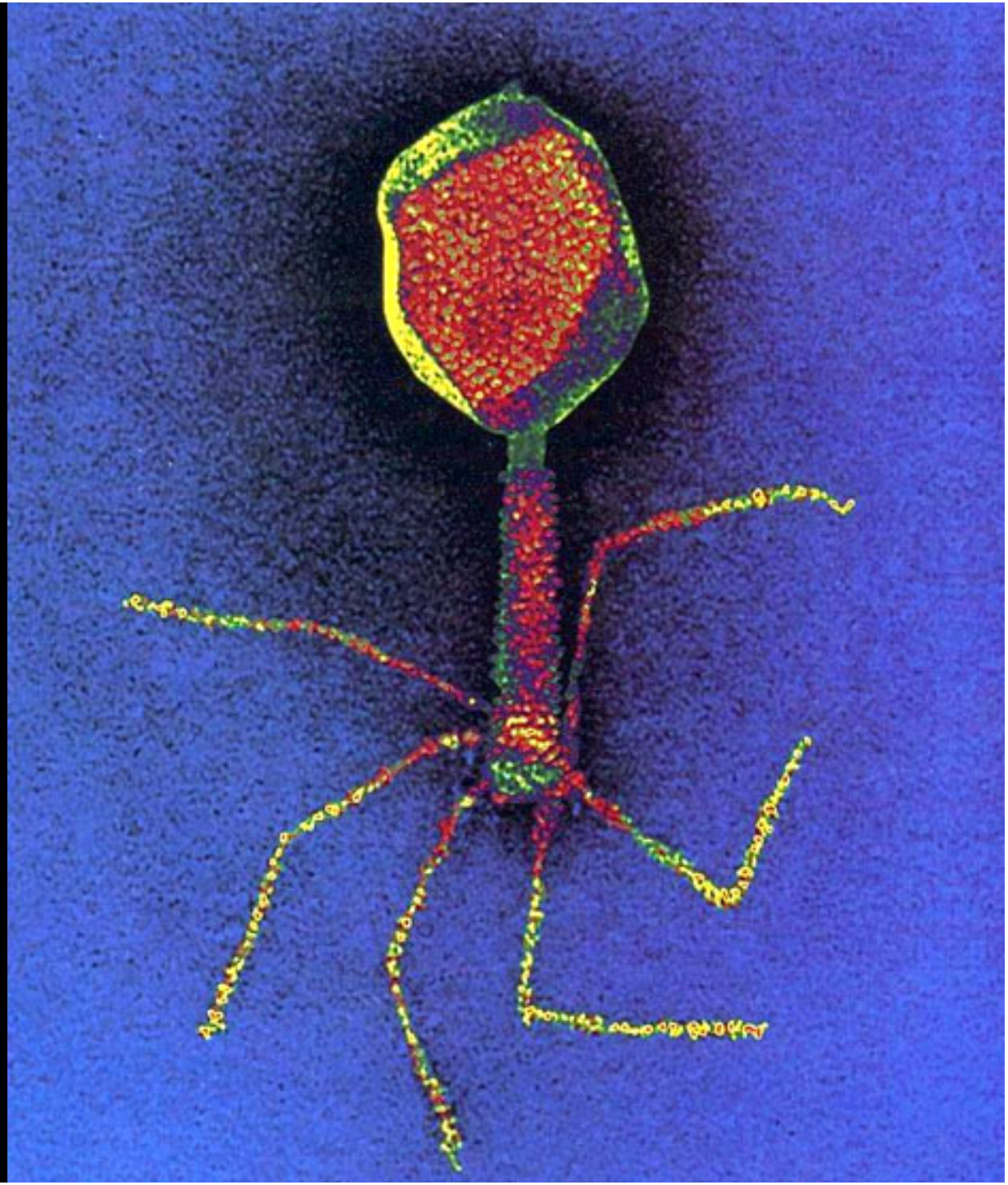
Virus Structure



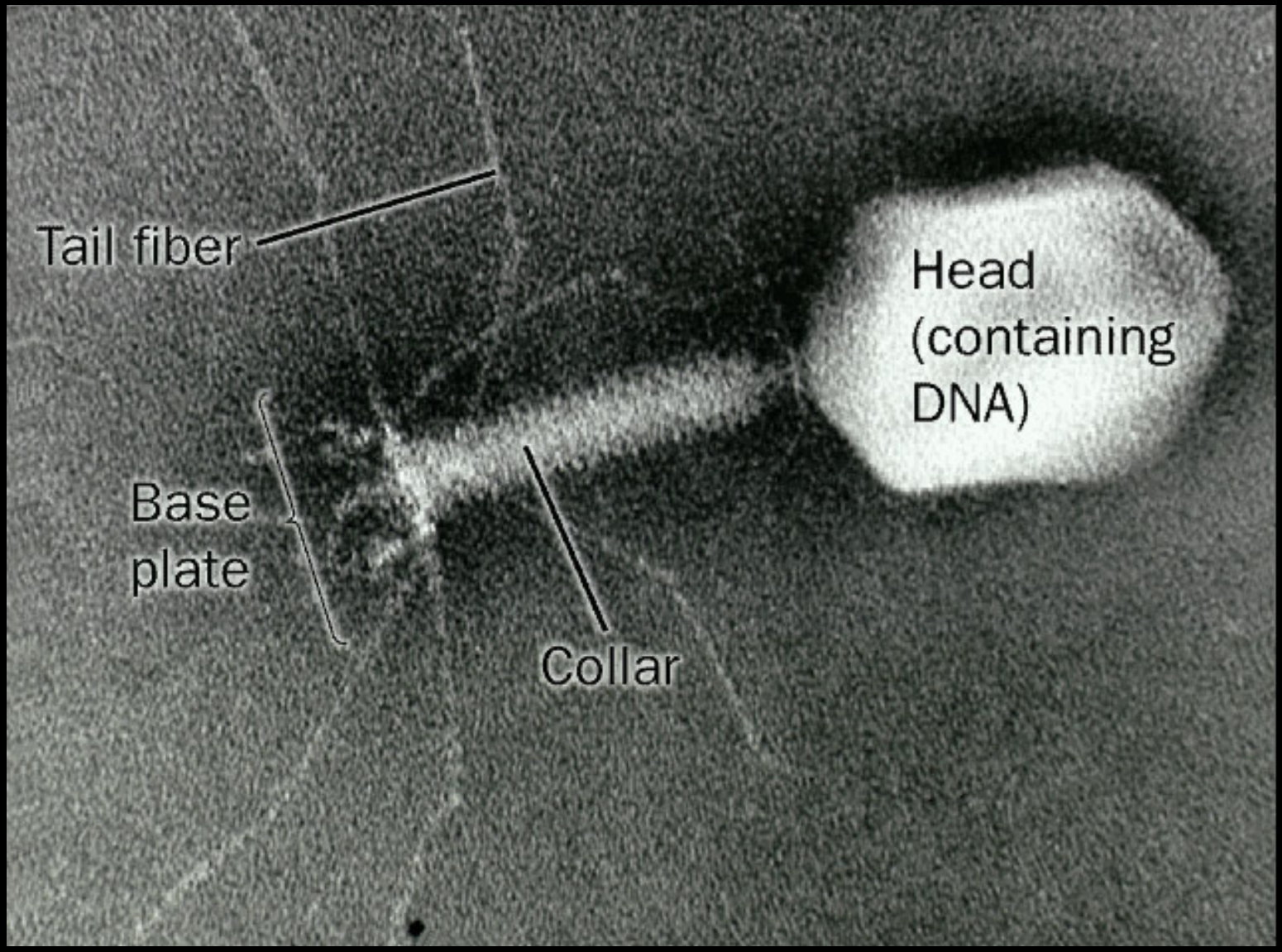
Phage

"one that eats"

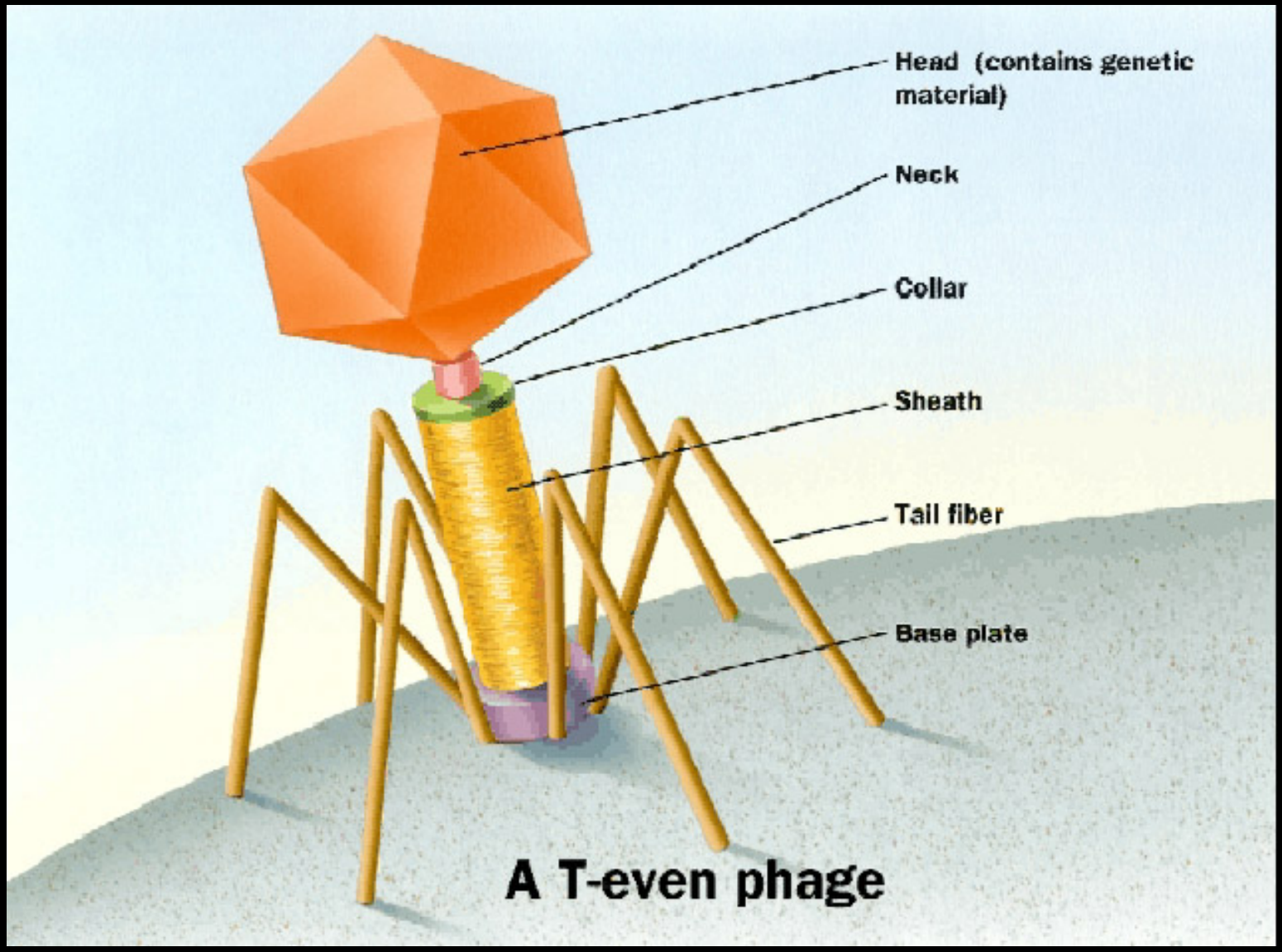
- Head contains DNA
- Legs attach to bacterium
- ≈0.2 μm long

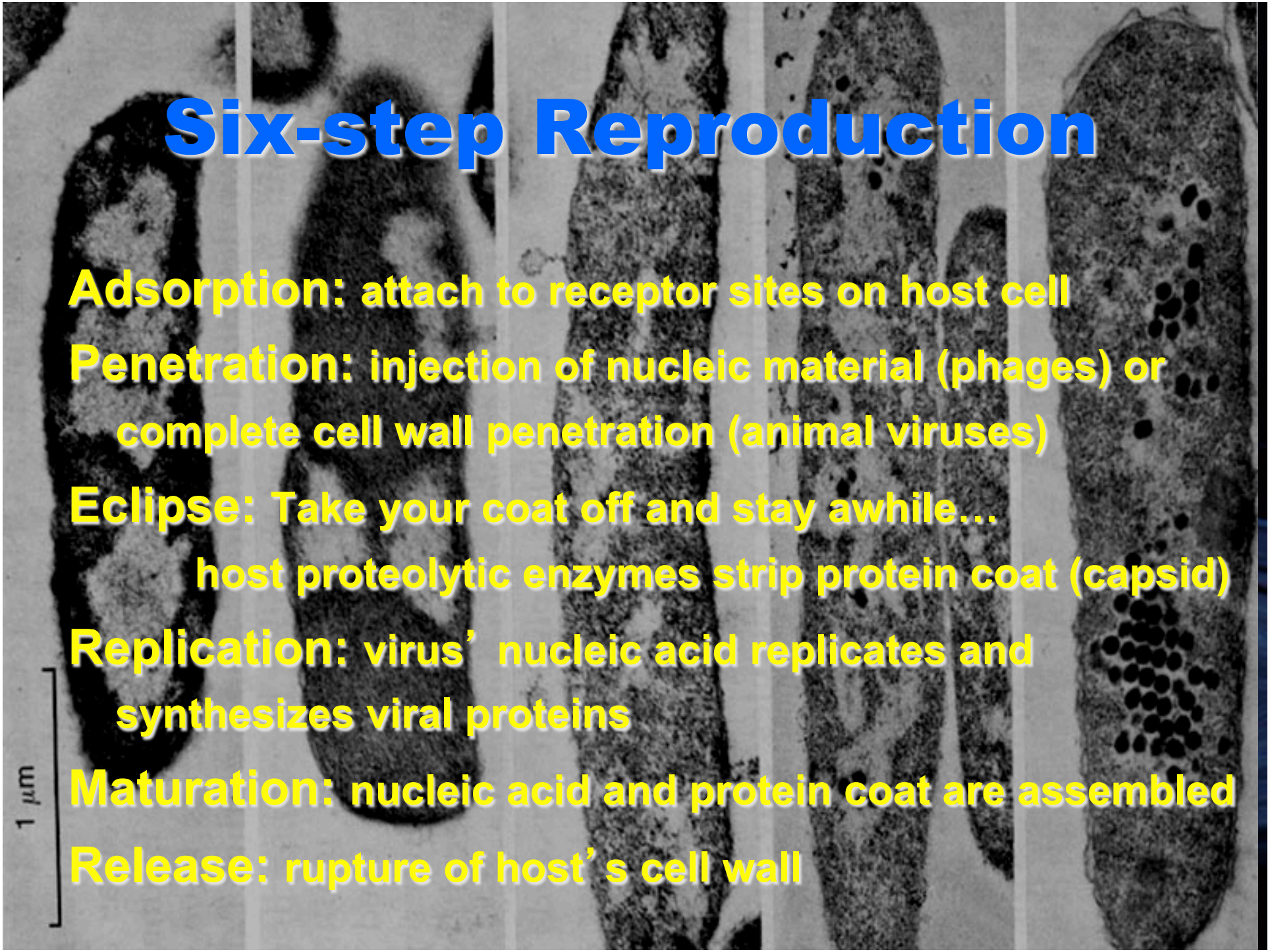


Bacteriophage Micrograph



Bacteriophage Illustration





Six-step Reproduction

Adsorption: attach to receptor sites on host cell

Penetration: injection of nucleic material (phages) or complete cell wall penetration (animal viruses)

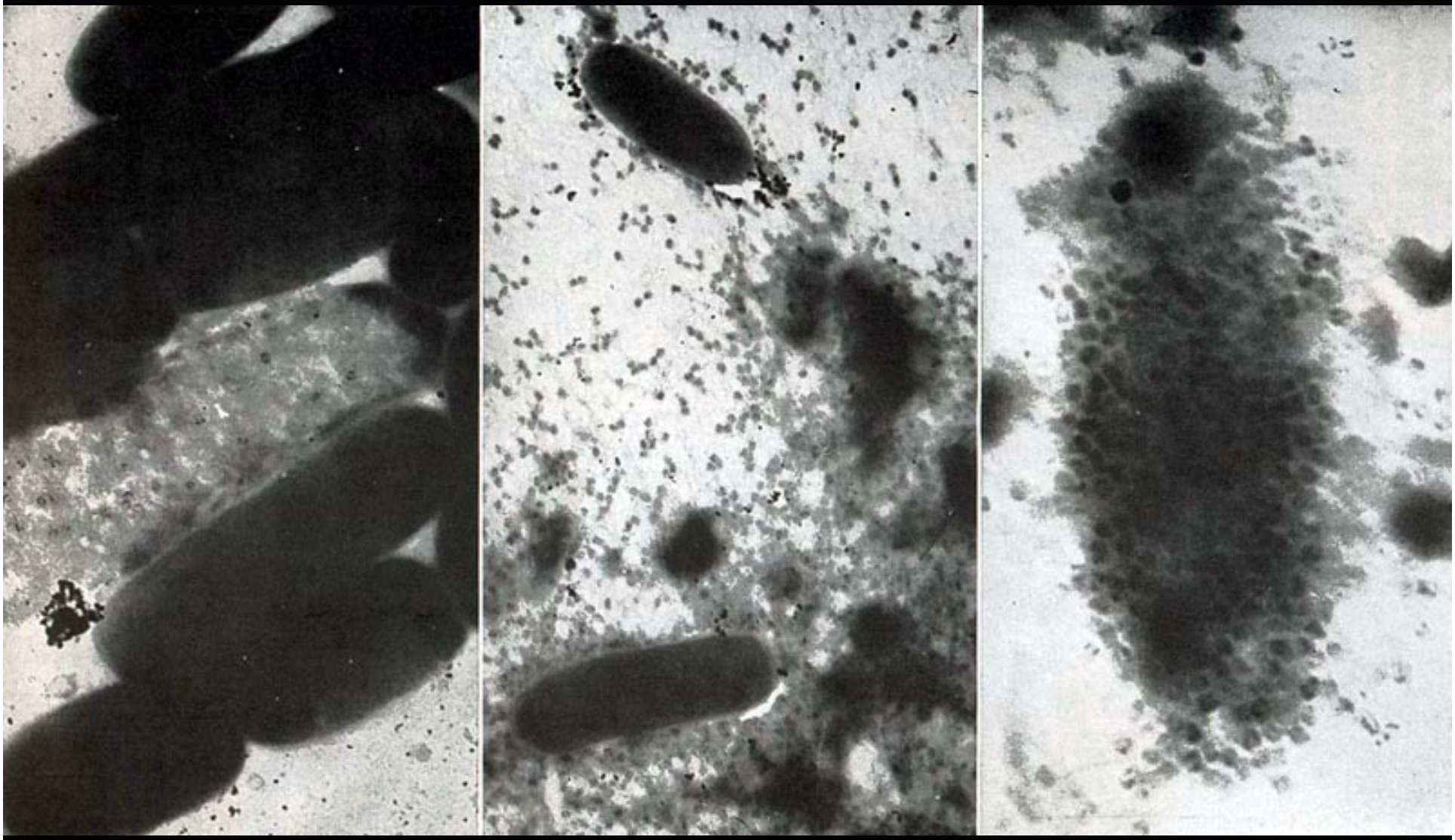
Eclipse: Take your coat off and stay awhile…
host proteolytic enzymes strip protein coat (capsid)

Replication: virus' nucleic acid replicates and synthesizes viral proteins

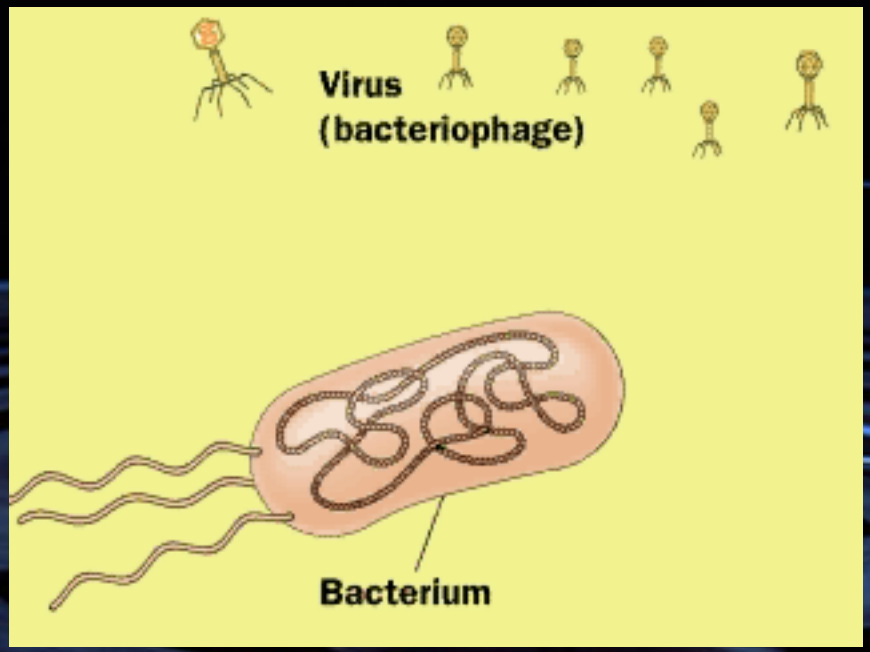
Maturation: nucleic acid and protein coat are assembled

Release: rupture of host's cell wall

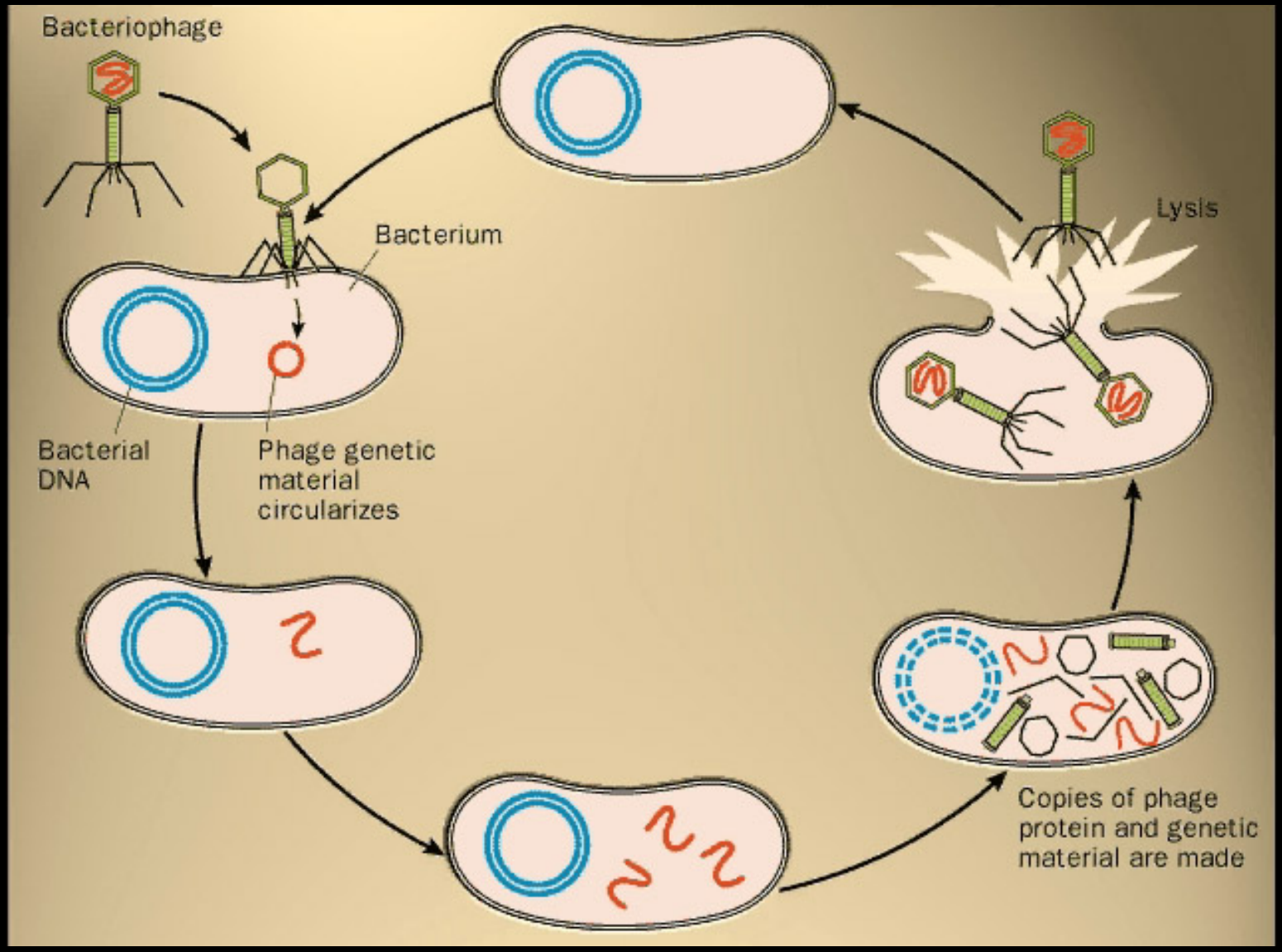
Bacteriophage Reproduction

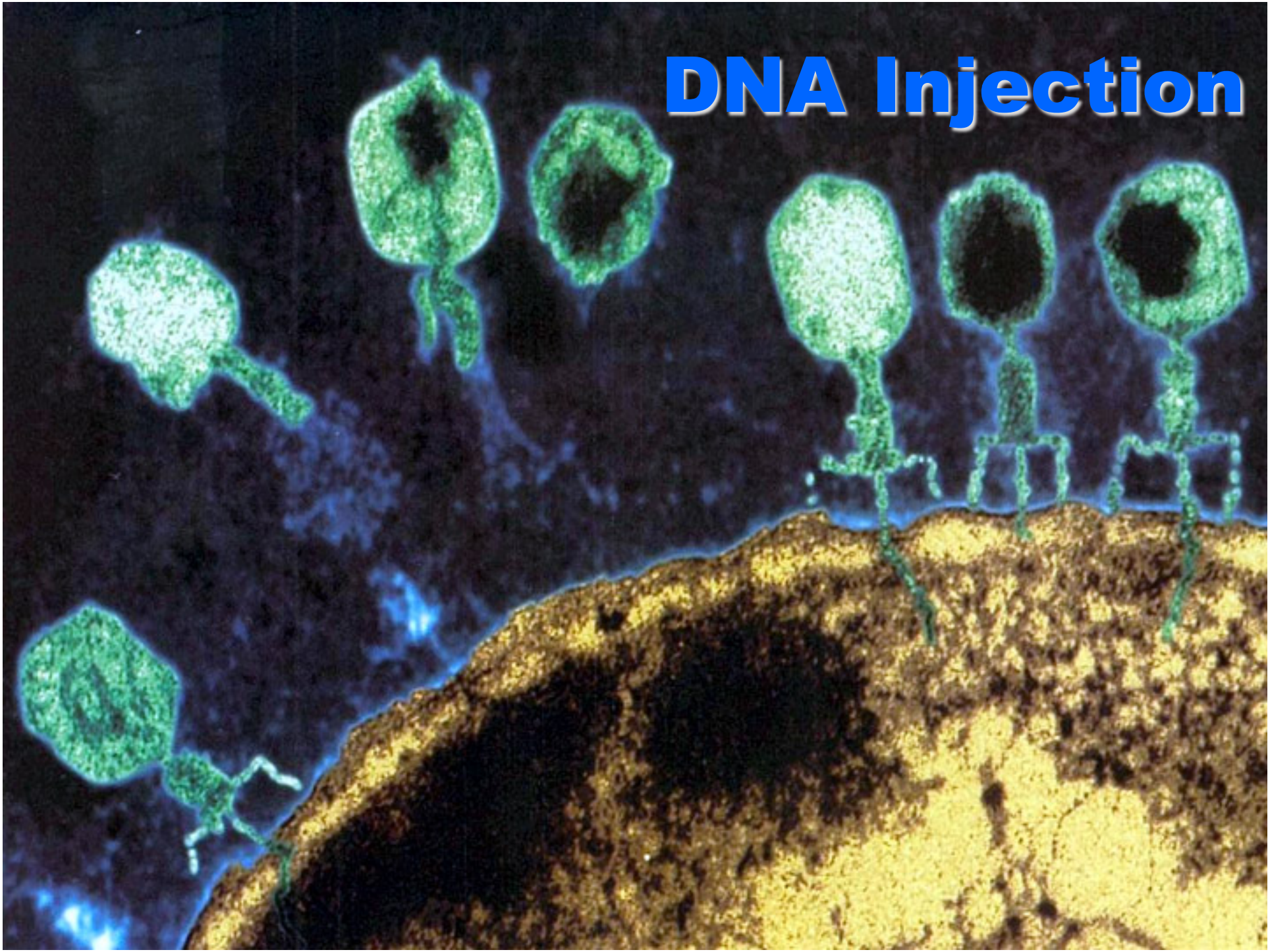


Phage Invaders

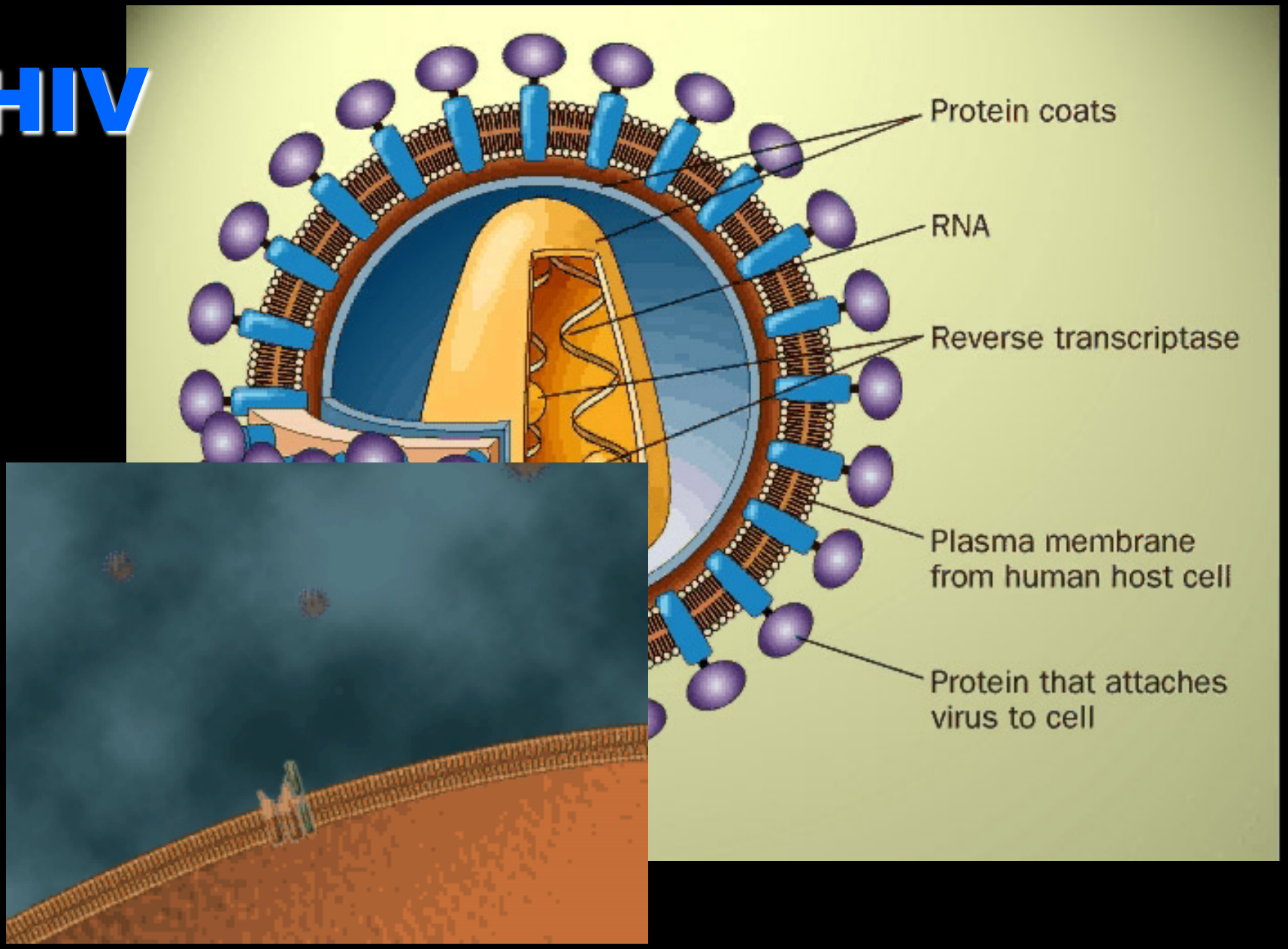


Bacteriophage Cycle




DNA Injection

HIV


Protein coats
RNA
Reverse transcriptase
Plasma membrane from human host cell
Protein that attaches virus to cell

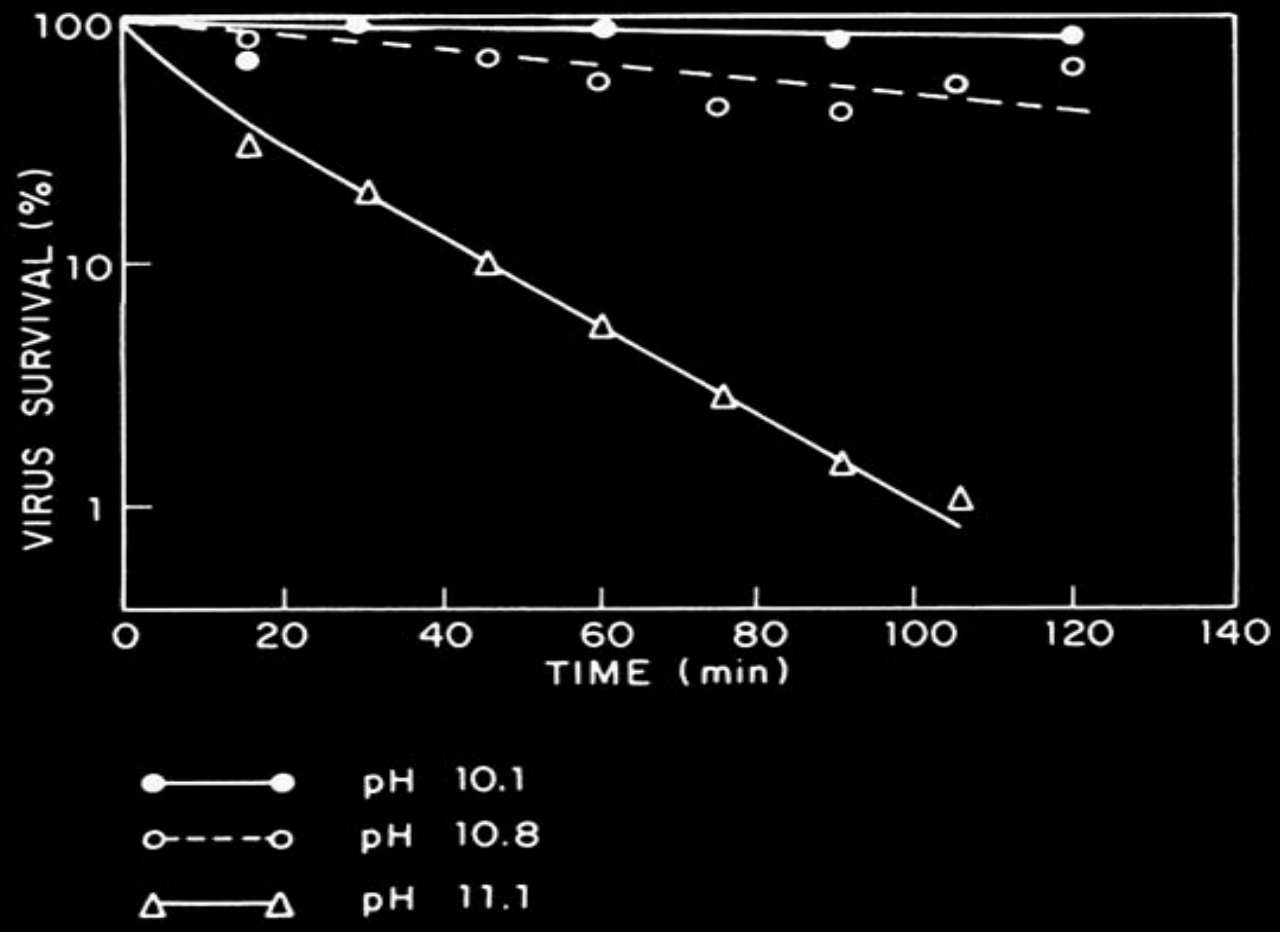
Ailments Associated with Viruses

Polio (Poliovirus)
Meningitis (Coxsackievirus)
Conjunctivitis (Andenovirus)
Meningitis, epidemic exanthem, infantile diarrhea (ECHO Virus)
Hepatitis (Hepatitis A and B virus)
Gastroenteritis (Reovirus, Rotavirus, Norwalk agent)

Also: colds, flu, fever, rash, eye infection, respiratory illness, smallpox, measles, mumps, herpes, AIDS, cold sores, warts

H_2O'C

Effect of pH on Viruses



Effect of high pH on poliovirus 1 (LSc). From G. Berg et al. (1968), *J. Am. Water Works Assoc.* **60:**193.

Inactivation of Viruses

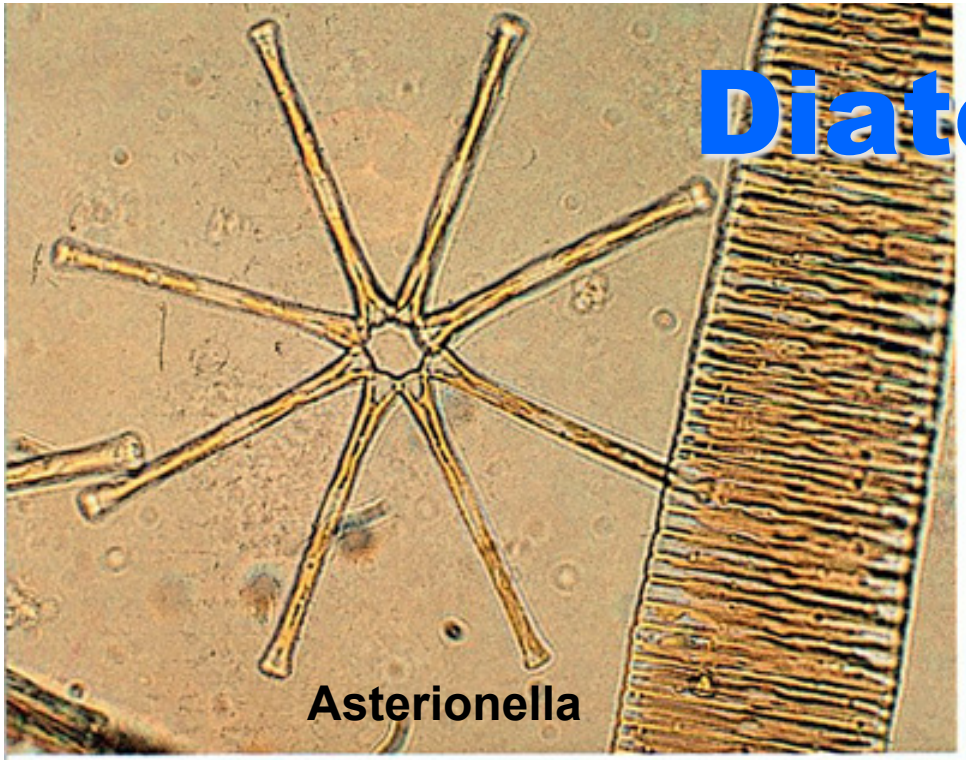
- pH > 11
- Chlorine
  - 0.5 ppm, pH 7.8, 2°C, 3 to 60 min. contact time ⊥99.99% kill
  - free chlorine 50 times more effective than chloramines
- Ultraviolet Light
- Ozone (very effective)
- Heat (60°C)

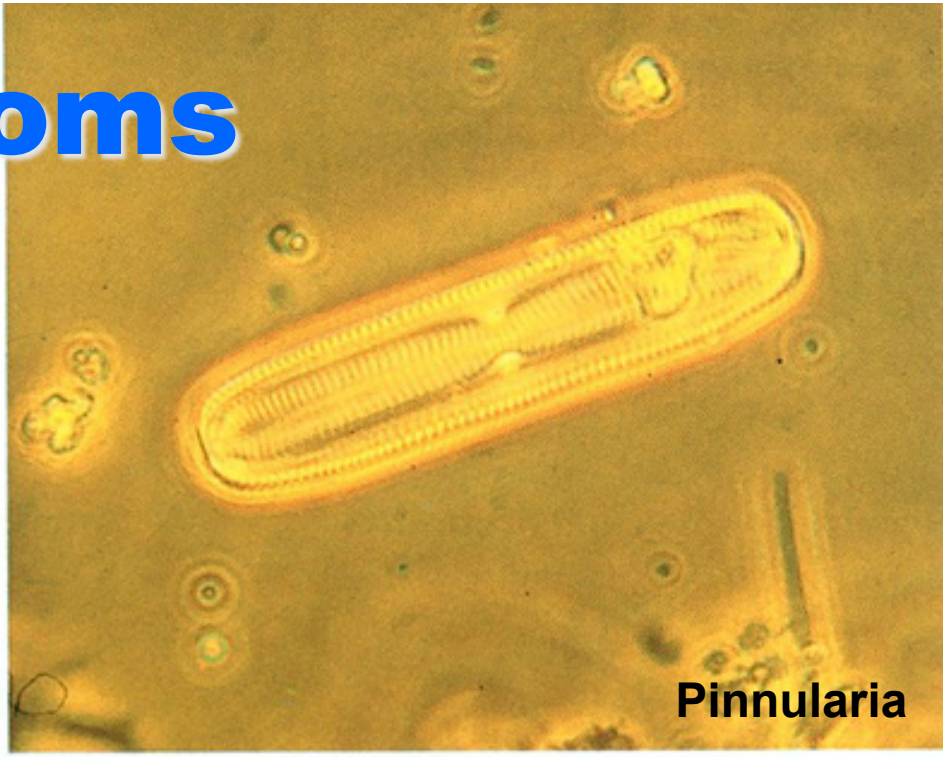
Algae

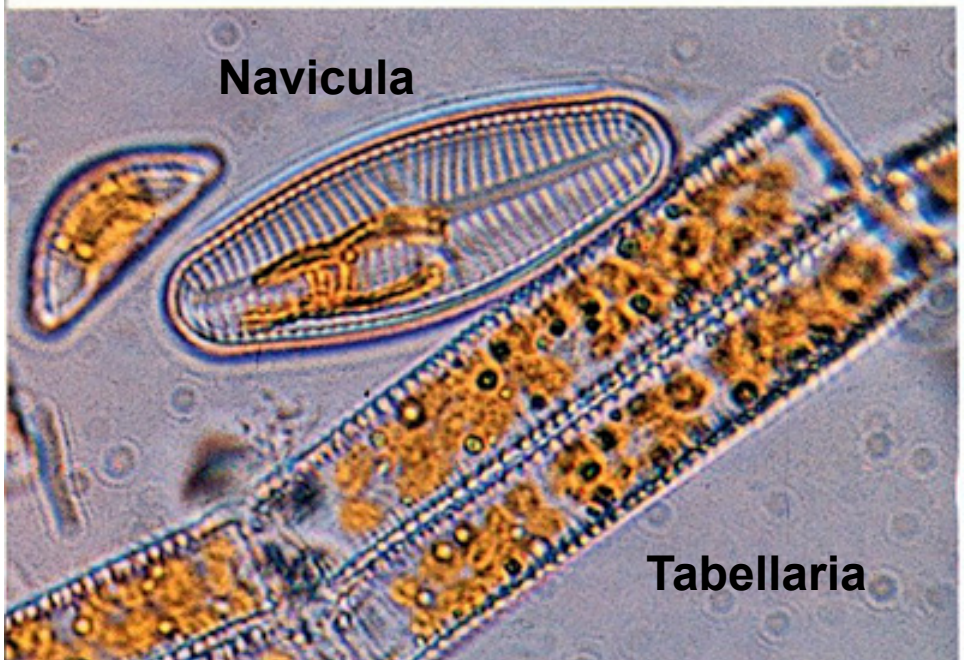
Algae Color Plate

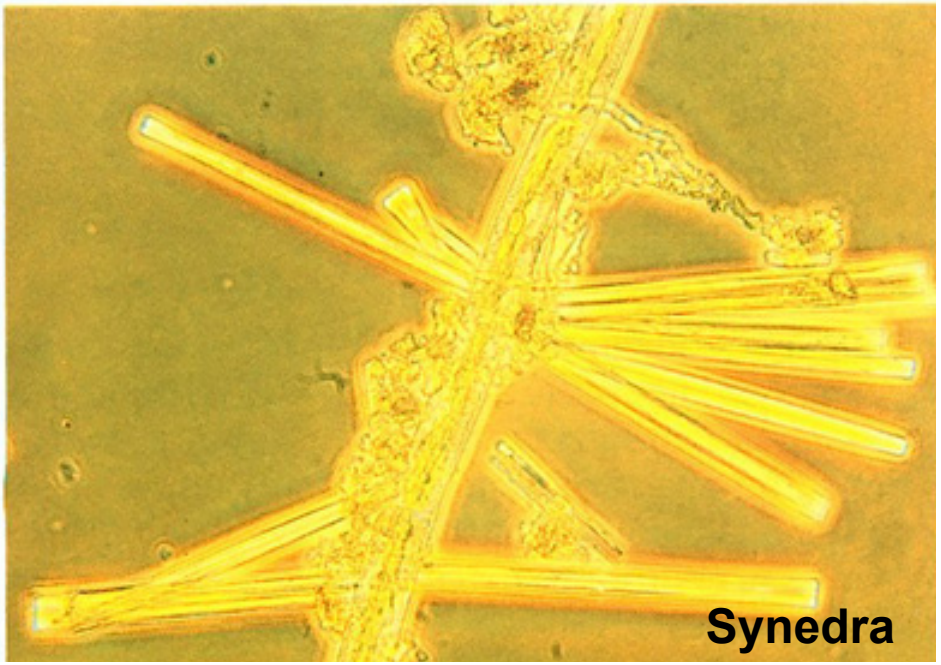
Standard Methods

Diatoms

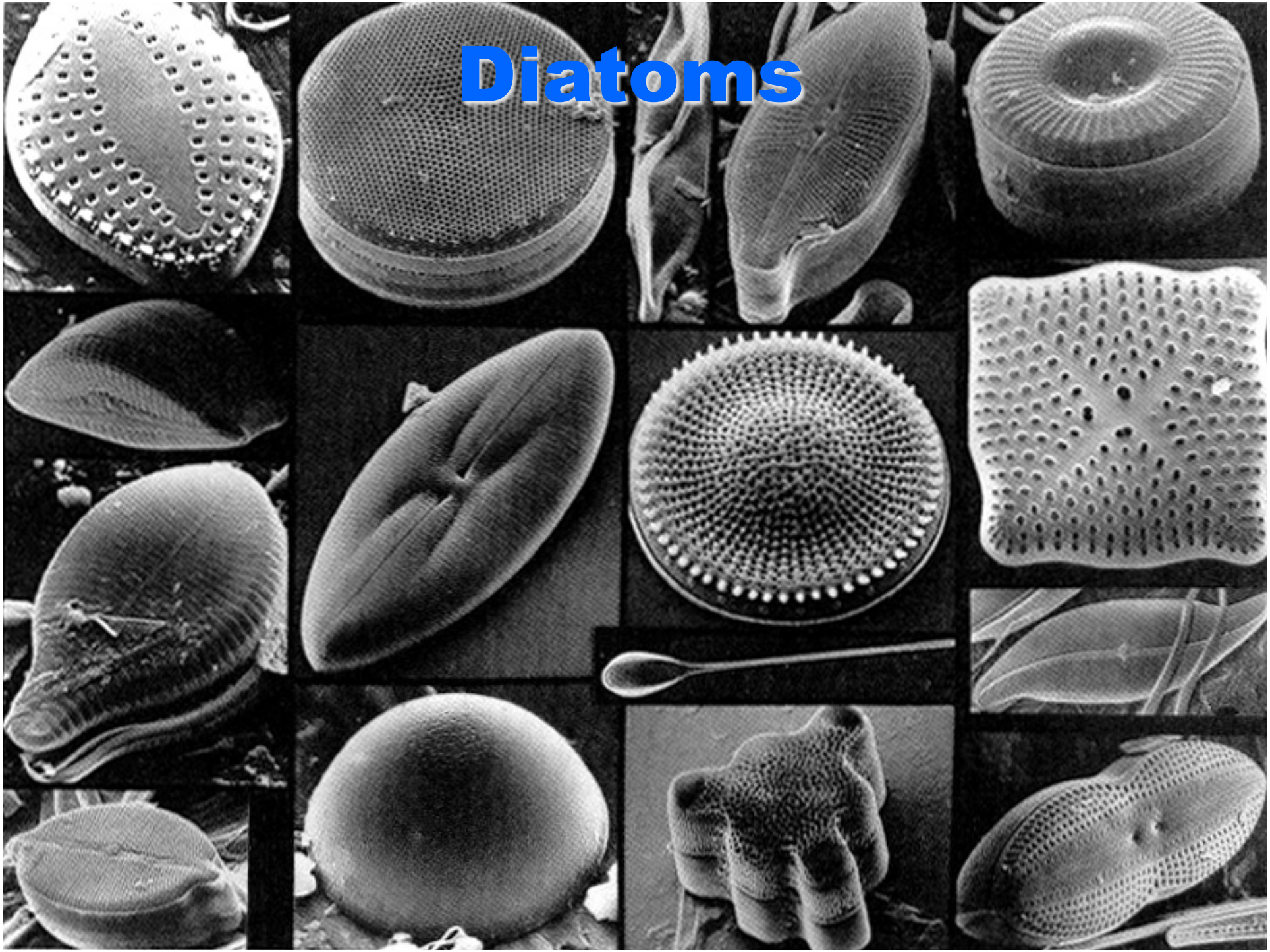




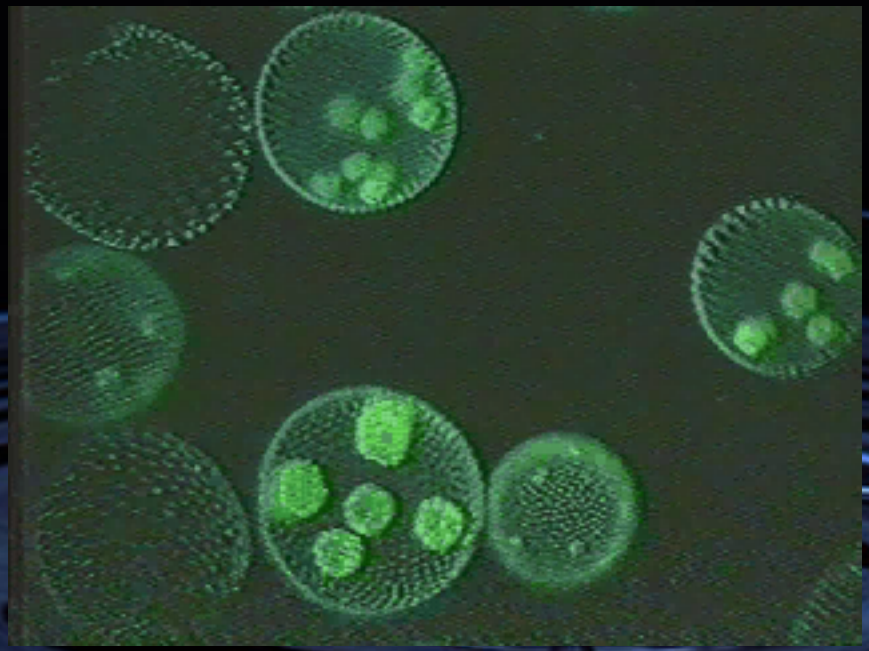




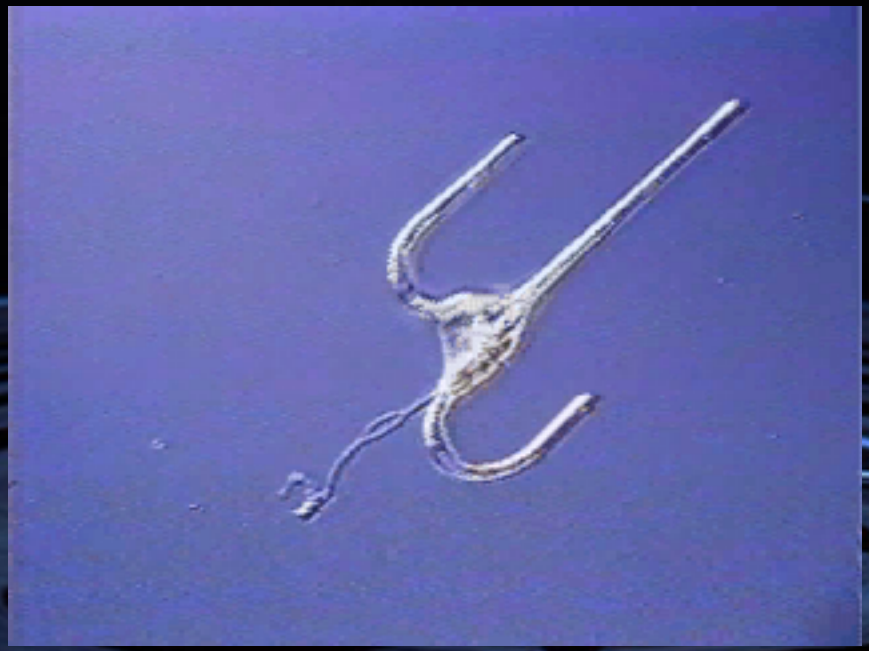
Diatoms



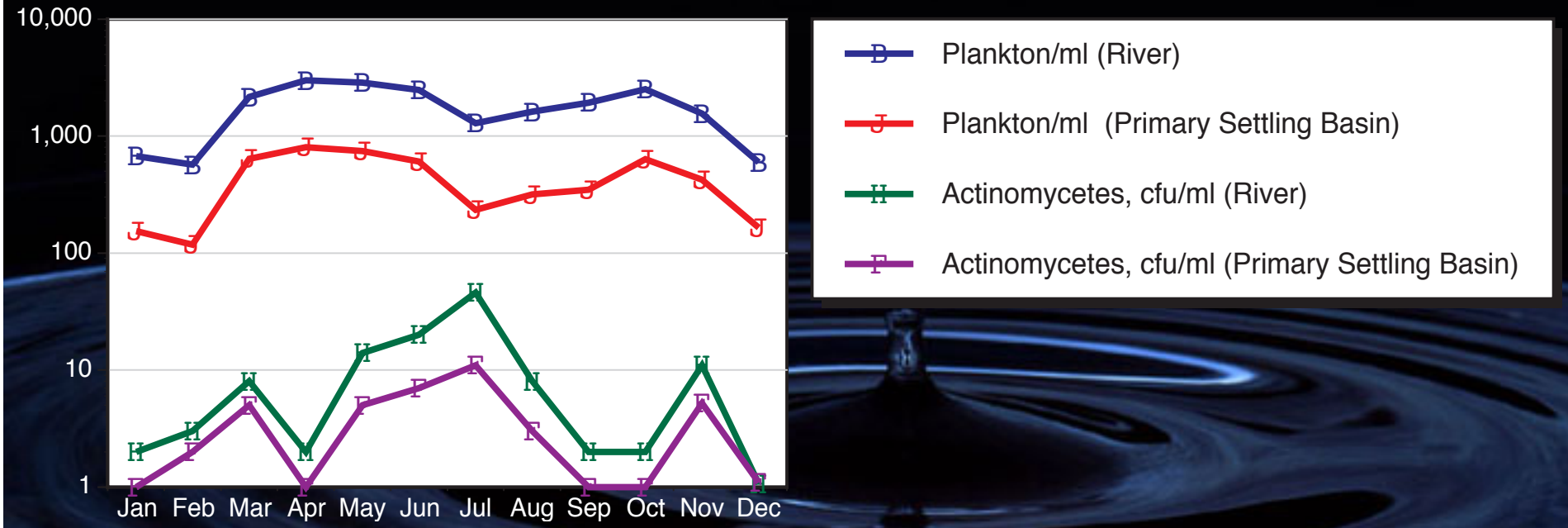
Volvox and Synura



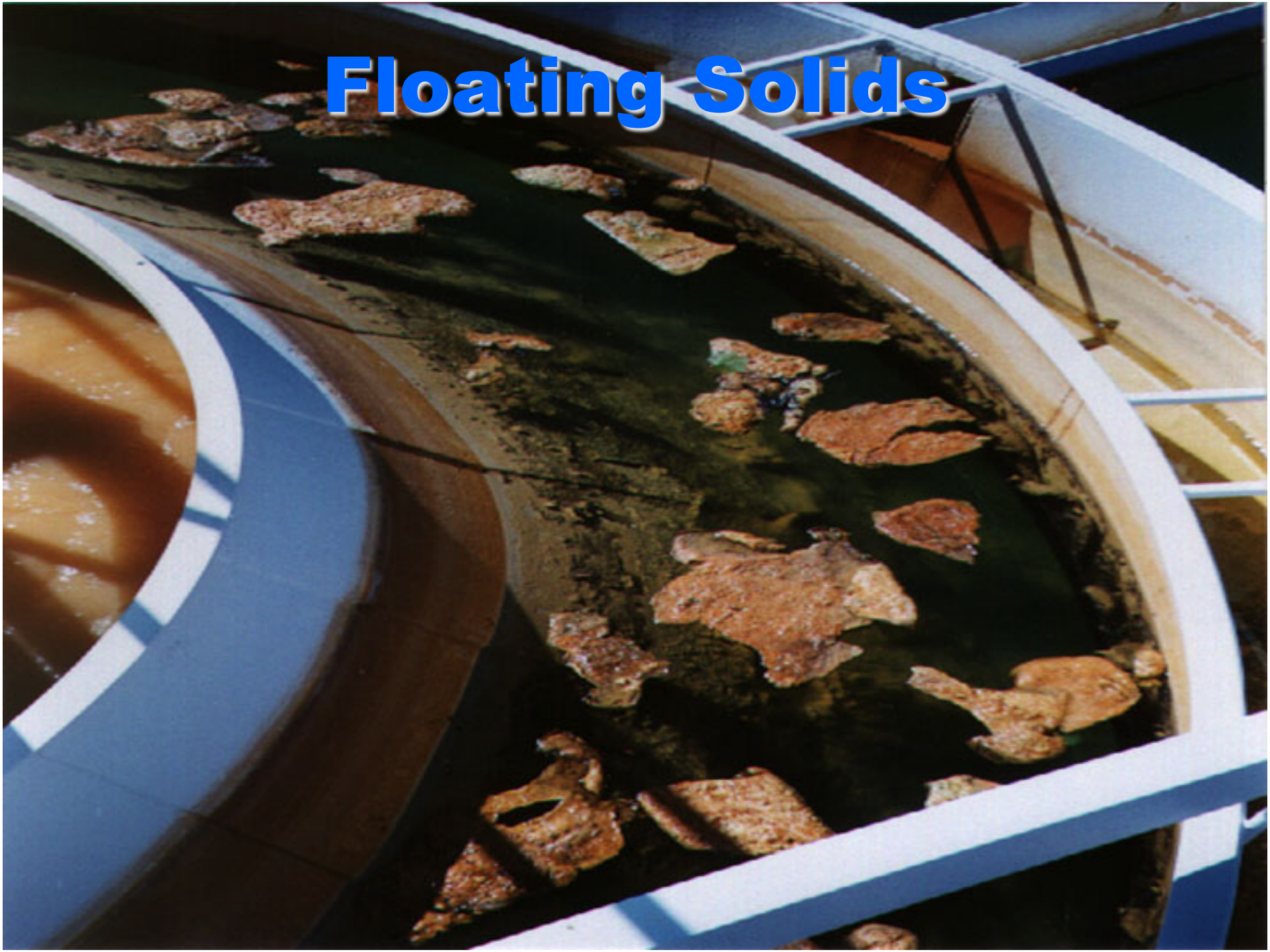
Dinoflagellate

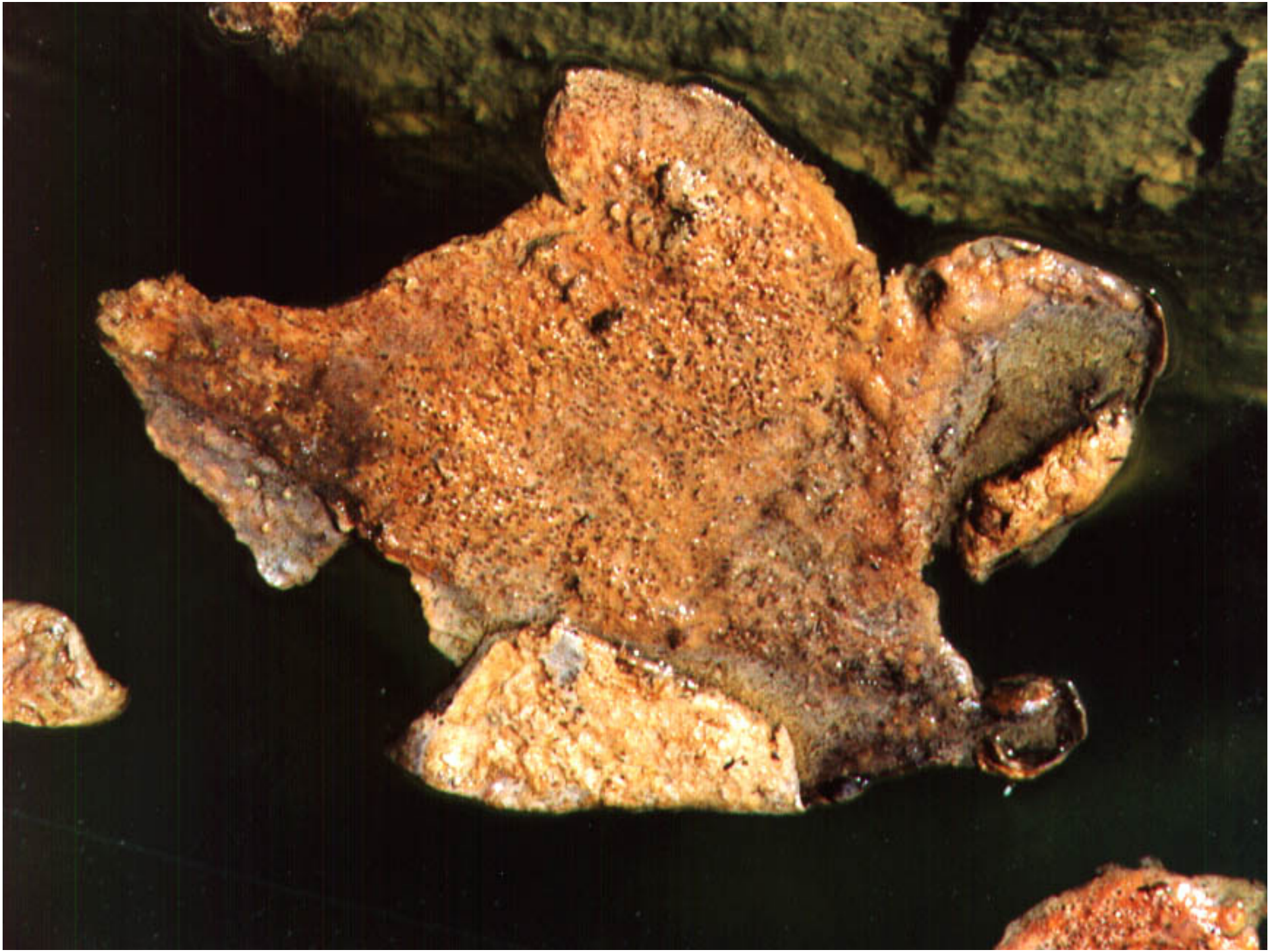


Plankton & Actinomycetes

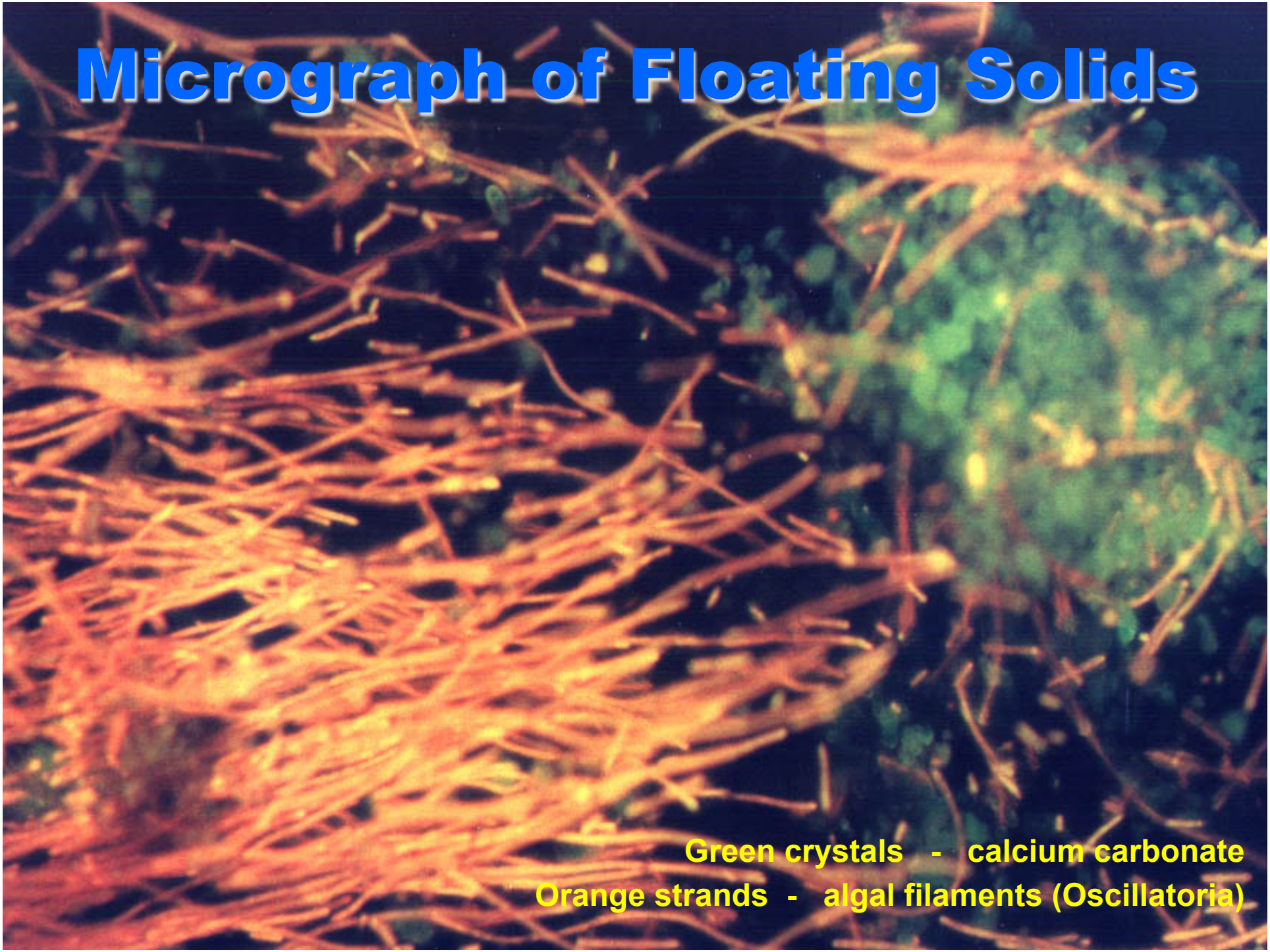


Floating Solids





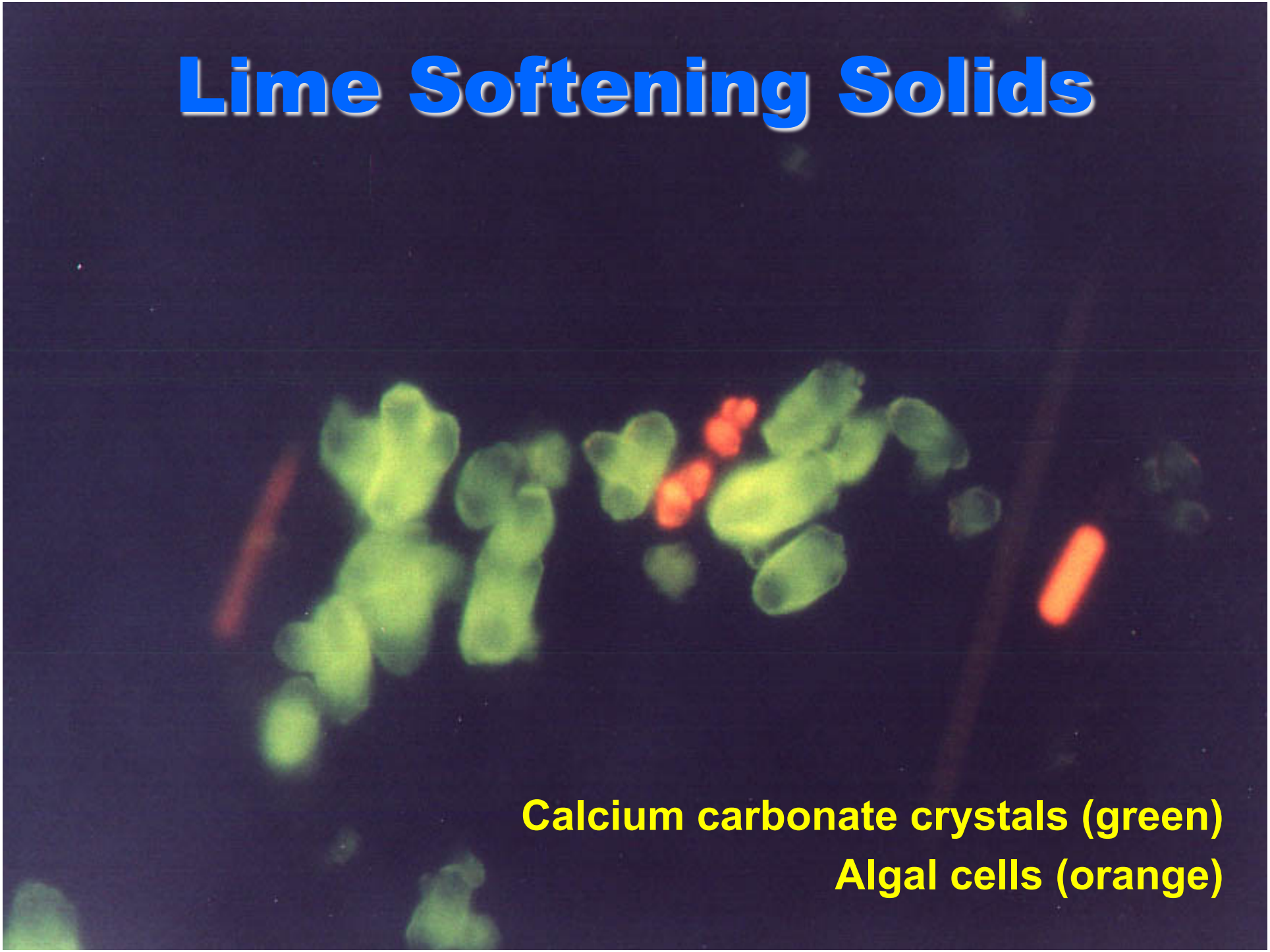
Micrograph of Floating Solids



Green crystals - calcium carbonate

Orange strands - algal filaments (Oscillatoria)

Lime Softening Solids

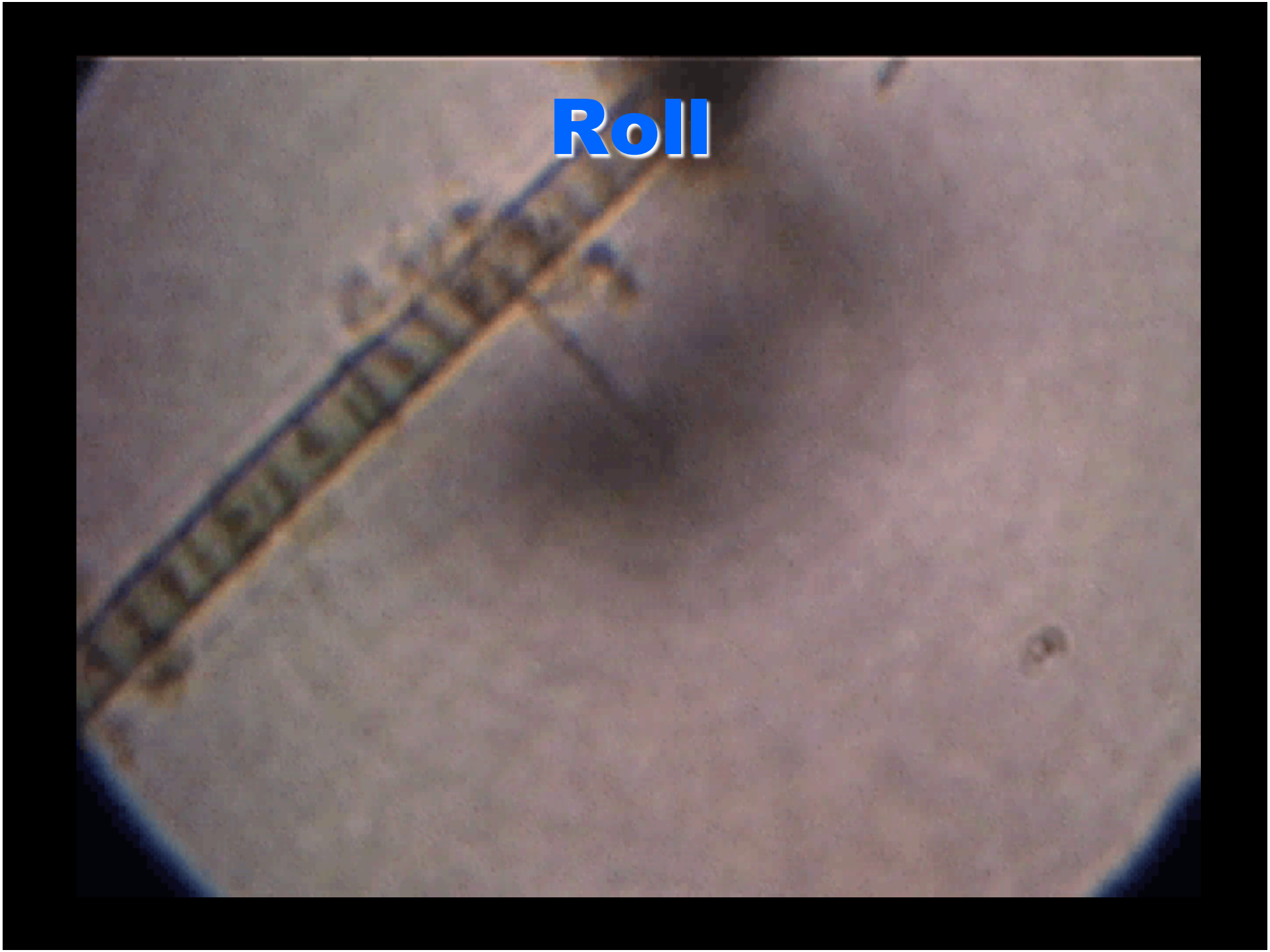


Calcium carbonate crystals (green)

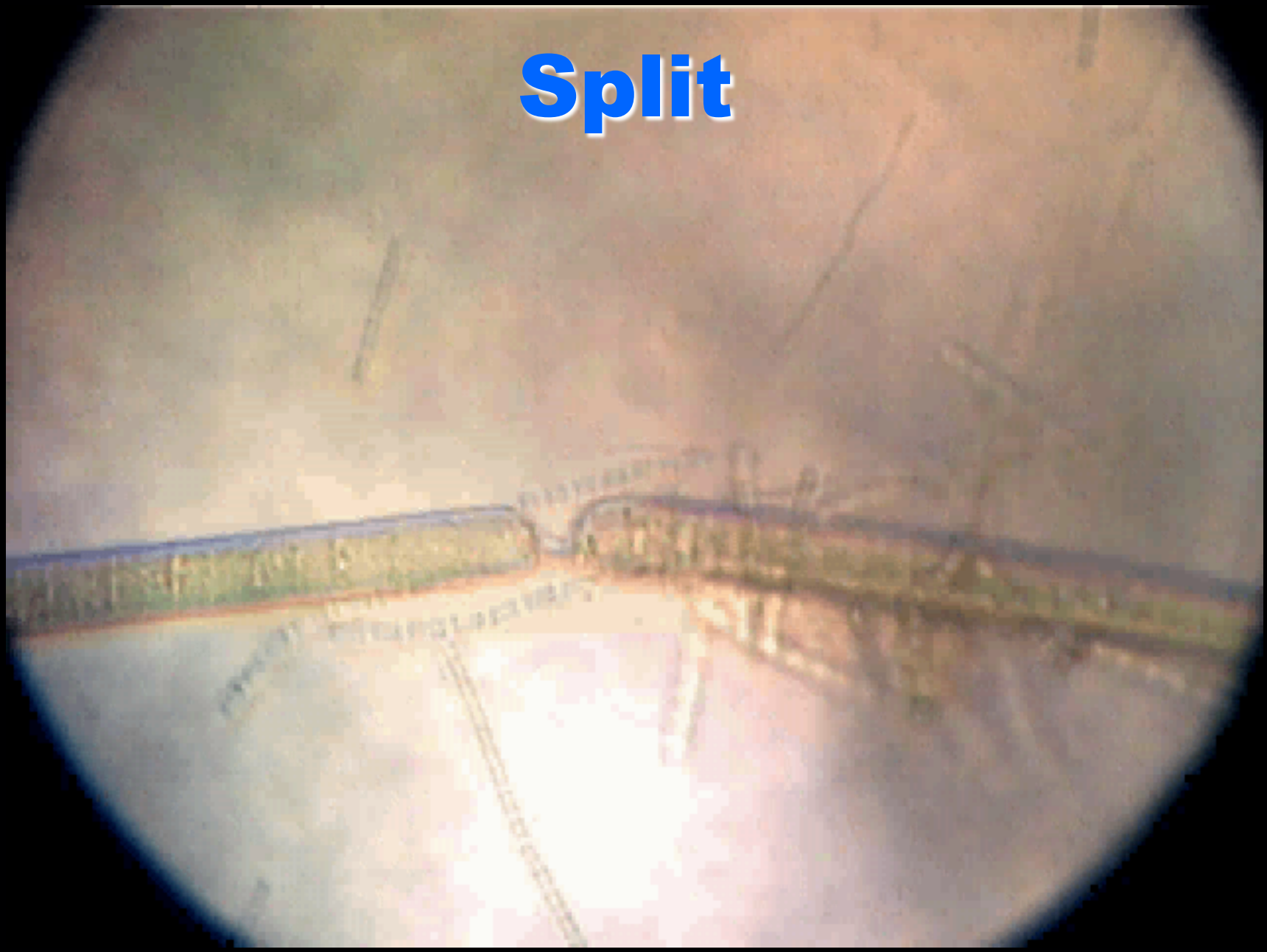
Algal cells (orange)

Algae, Bacteria, $CaCO_3$

Rotation


Roll

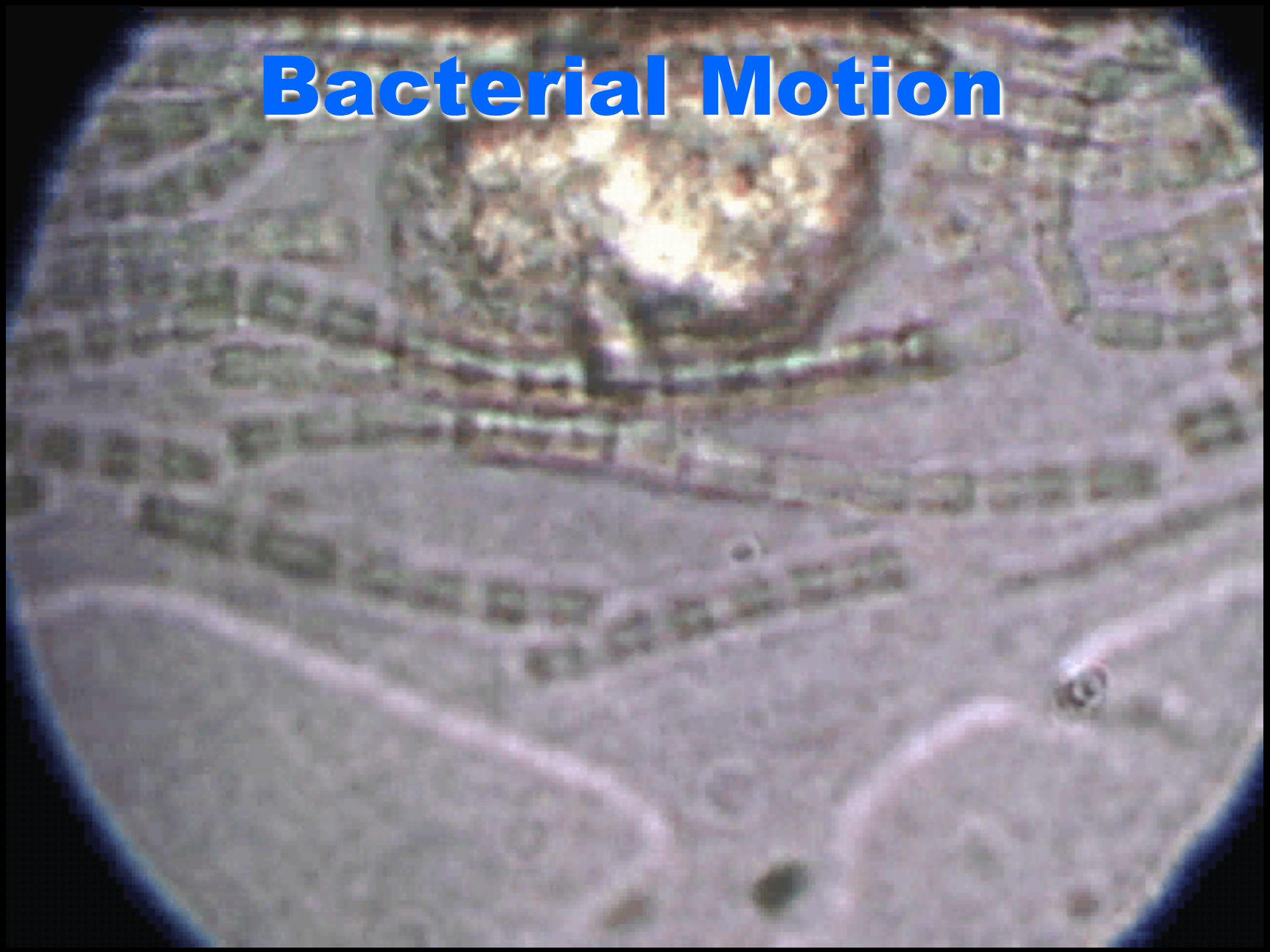
Split



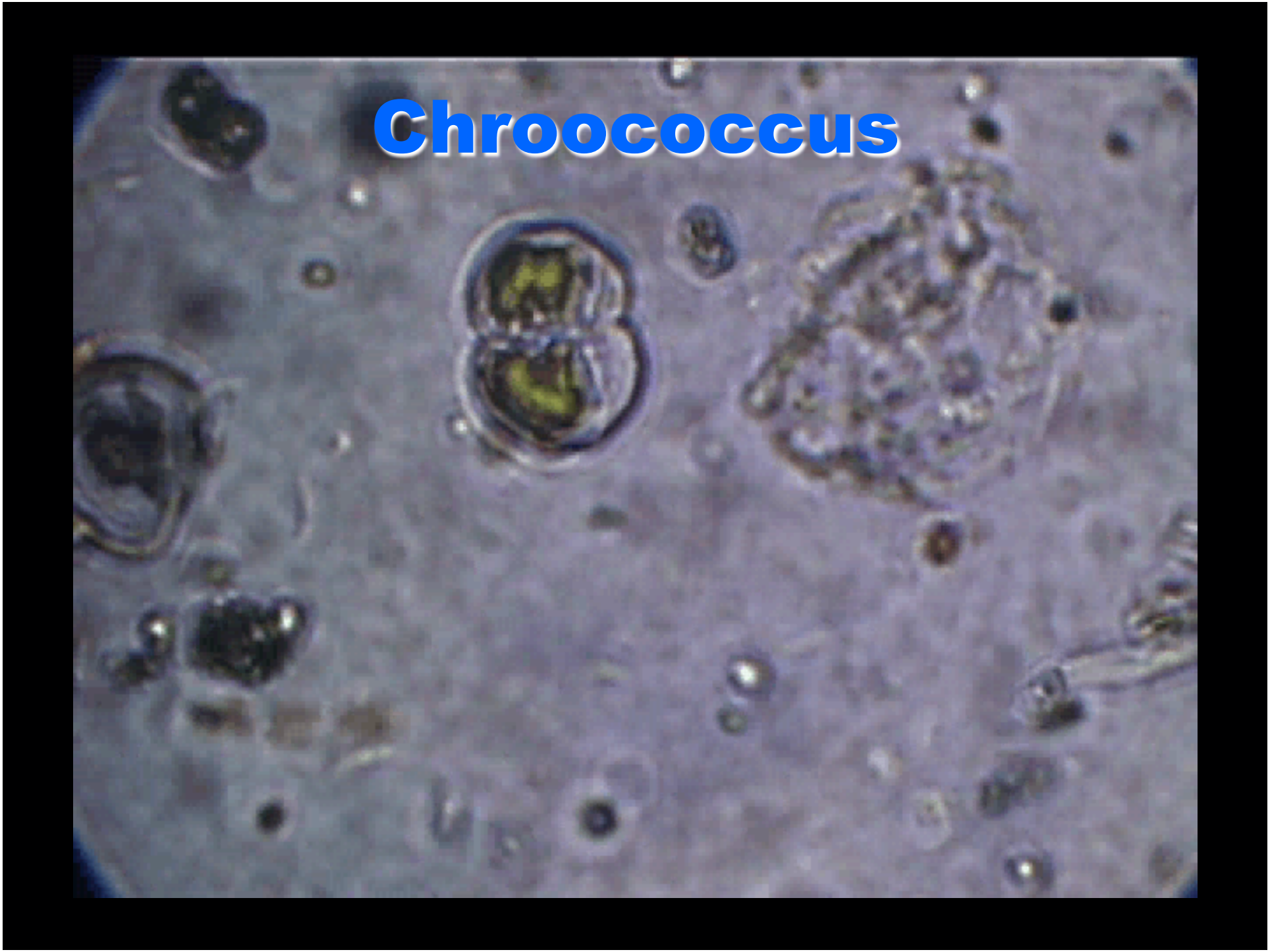
Protozoan, Algae, Bacteria

Bacterial Chains & Filaments

Bacterial Motion

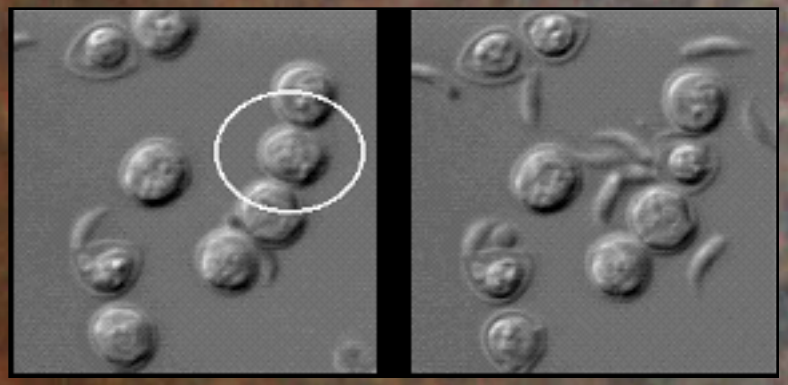


More Chains and Filaments


Chroococcus

Nematodes in the Mat

Protozoans



Cryptosporidium

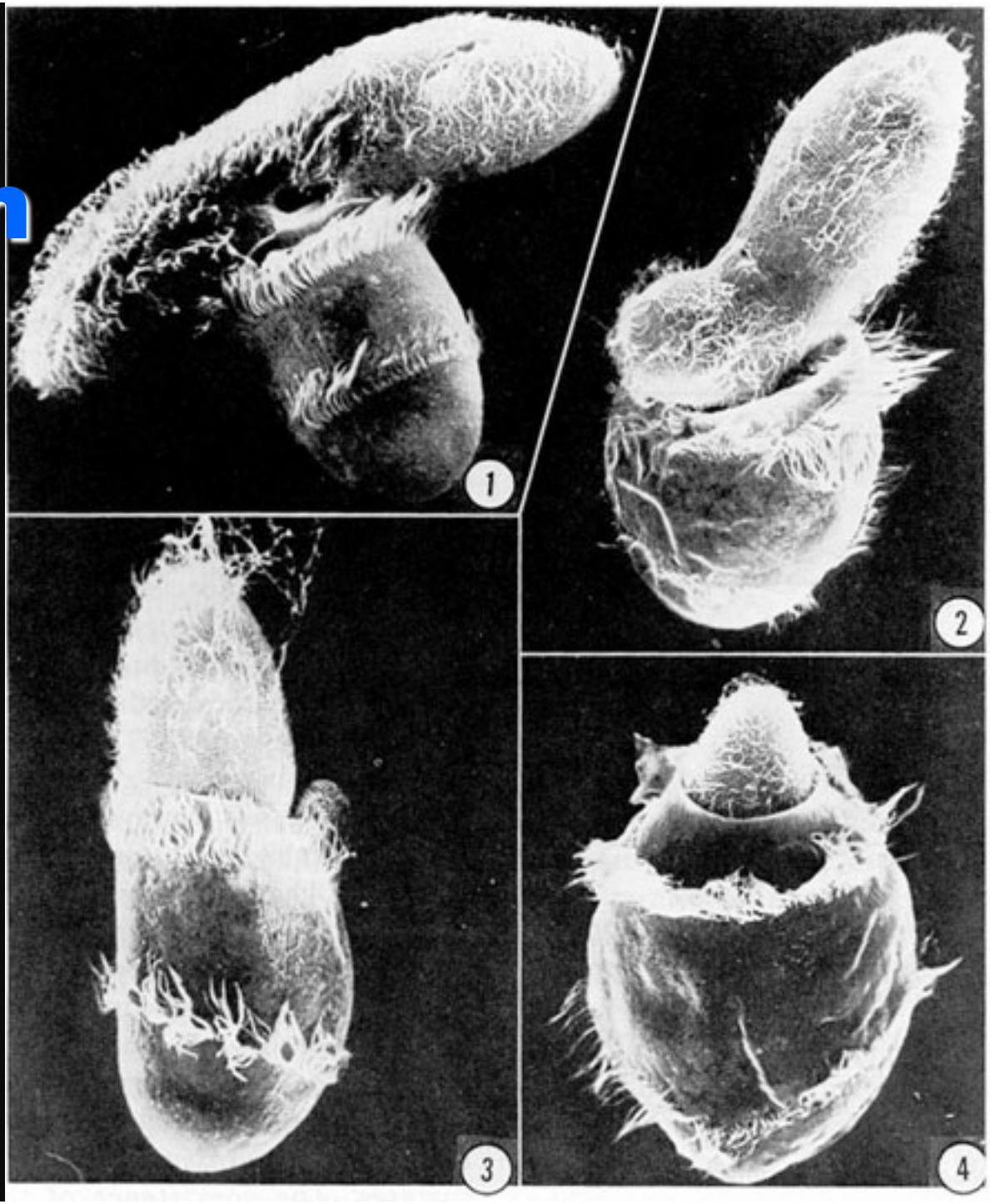
- 40,000 species
- single-celled
- up to several mm long

Paramecium

Paramecium cilia

Life in the Food Chain

Didinium catches, eats a Paramecium



Ameba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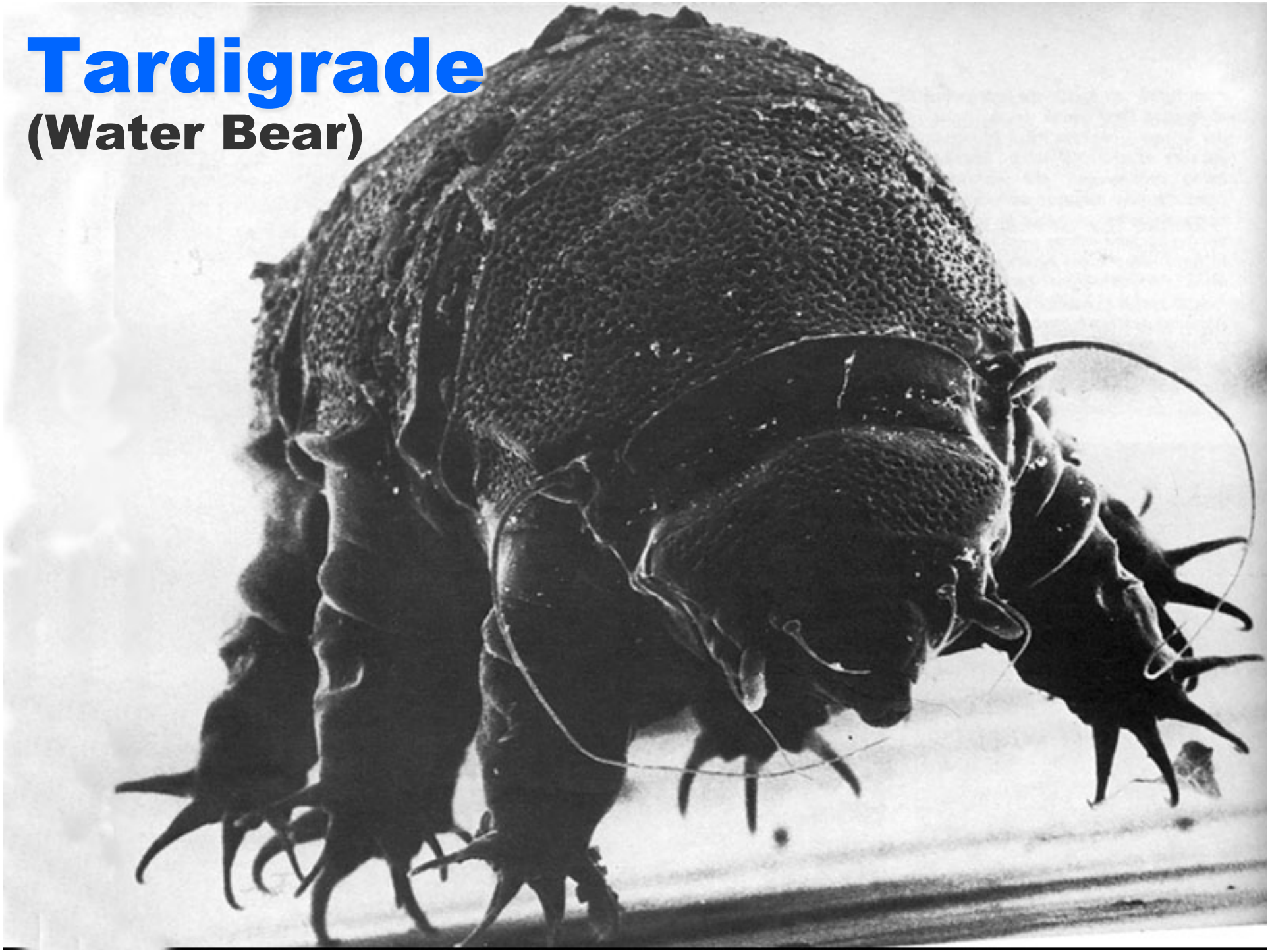
Tardigrade
(Water Bear)



Ailments
Attributed to Protozoans

amoebic dysentary (Entamoeba histolytica)
giardiasis (Giardia lamblia)
giant roundworm (Ascaris lumbricoides)
cryptosporidiosis (Cryptosporidium)

Lines of Defense

- Source Water Protection
- Physical Removal
- Kill / Inactivation
- Disinfectant Residual
- Maintain Integrity of Distribution and Storage