



Lab Project Anti-Bacterial Drugs, Resistance, and Evolution

Antibacterial Drugs

Antibiotics: made by microbes (fungi, bacteria)

Synthetics: man-made, not true antibiotics!

KNOW target in cell, spectrum (broad, narrow)

History of Antibacterial Drugs

Ehrlich - Magic Bullet Concept (1909)

Hoped to develop something to treat syphilis

Compound that killed microbe, not host

Could only find TOXIC arsenic = salvarsan

Fleming - First Antibiotic (1928)

Penicillin made by soil fungus Penicillium

Mimics peptidoglycan; in wall, halts growth

Affects Gram (+) more - narrow spectrum

*Ampicillin = semi-synthetic version of penicillin with
"broadened" spectrum.*

Domagk - First Synthetic Drug (1935)

Sulfa mimics PABA - only bacteria convert to...

Folic acid; in enzyme, halts vital metabolic step
Affect Gram (-) and (+) - broad spectrum

Waksman - Second Antibiotic (1944)

Streptomycin - by soil Bacteria Streptomyces

Binds, halts bacterial small subunit ribosome

Affect Gram (-) and (+) - broad spectrum

Drug Resistance – ONE Change Only...

Change cell receptor - halt drug uptake

Change target or pathway - halt drug effect

Change pump - efflux, pump drug out of cell

Evolution - Mutation and Recombination

Mutations: DNA Pol errors = $\sim 1/10^6$, natural

Recombination: eukaryotic meiotic cross-over

OR prokaryotic conjugation (review/know!)...

OR prokaryotic transformation (review/know!)...

Transduction covered during virology lectures!

Mutation/Selection Application - Ames Test

Mutagenicity ~ Carcinogenicity, FDA testing

10^7 Salmonella on Nutrient – lawn

Same on Nutrient^{Str} – a few colonies (WHY?!?!)

Same with MUTAGEN on Nutrient^{Str} = MANY

ACTIVITIES – NOTE TWO FOLLOW-UP'S

Antibacterial Drug Testing - Work In Pairs, Incubate 37°

Label 2 nutrient agar plates (1 for each culture) and divide into as many drugs as will be tested (TBA)

Add 0.2 ml of each test culture and spread; aseptically place disks onto appropriate sections

Next time, measure zones of inhibition radii in mm – did data match lecture/text spectrum?

Notebook records - drawings, clearly labeled table of zone data with interpretation

Demonstrating Transformation, Set-Up - Work In Pairs, Incubate at 37°

Review PowerPoint: KNOW COLD – all Acinetobacter strains (A-R, A-S), all media (selective or not)

Divide nutrient plate into quarters: live A-R, live A-S, lysed A-R, lysed A-R + live A-S

Aseptically loop live controls into respective quadrants - spread into dime-sized circles

Add 0.5 ml SDS to A-R and incubate at 75°C 15 min - What does this do? Which molecules intact?

Loop lysed A-R into two appropriate quarters; when finished, loop live A-S exactly over lysed donor.

Notebook records – indicate if there were problems, contamination!

Demonstrating Transformation, Follow-Up - Work In Pairs, Incubate at 37°

Obtain (NS), divide into thirds (why?), label; using a loop, transfer all growth from (N) to (NS).

Notebook records – drawings, summary table of final data (3-4 sectors, 2 plates)

LAB MATERIALS TURN-IN

7 pts. Informal Notebook records: make sure all suggested guidelines above are followed.

4 pts. Each pair turns in fully-labeled (including selective, non-selective) transformation test plates.

4 pts. Each pair turns in fully-labeled antibacterial drug test plates after they have been analyzed