ALPHABET SOUP OF ANTIMICROBIAL RESISTANCE

LABORATORY
MEDICINE COURSE
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CLINICAL MICROBIOLOGY SERVICE

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ALPHABET SOUP OF ACRONYMS

- MRSA- METHICILLIN-RESISTANT *S. aureus*
  - ✓ 46% AT CUMC
- VISA- VANCOMYCIN (GLYCOPEPTIDE)-INTERMEDIATE *S. aureus*
- VRSA- VANCOMYCIN-RESISTANT *S. aureus*
- VRE- VANCOMYCIN R *Enterococcus faecium*
  - ✓ 80% AT CUMC
- ESBLs - Extended-spectrum β-lactamases
  - ✓ GRAM-NEGATIVE RODS
  - ✓ 18% AT CUMC

WHAT AFFECTS CHOICE OF ANTIMICROBIAL AGENTS ?

- ANTIMICROBIAL SUSCEPTIBILITY TEST RESULTS
- PHARMACODYNAMICS
  - ✓ AUC:MIC_{90} RATIO
  - ✓ HALF LIFE OF DRUG
  - ✓ TIME ABOVE THE MIC
  - ✓ CONCENTRATION DEPENDENT KILLING
    - Greater cidal activity with higher concen (e.g. aminoglycosides, β-lactams)

ANTIBIOTIC SUSCEPTIBILITY TESTS Role of Clinical Microbiology

- FOLLOW CURRENT NATIONAL COMMITTEE CLINICAL LAB STANDARDS (NCCLS)
- USE OPTIMAL SUSCEPTIBILITY METHODS & QUALITY CONTROL MEASURES
- PROVIDE MIC & INTERPRETATIONS
  - ✓ e.g. SUSCEPTIBLE, INTERMEDIATE, RESISTANT
- WHAT DRUGS SHOULD BE TESTED & REPORTED?
  - ✓ APPROPRIATE DRUG/BUG COMBINATIONS
  - ✓ ID, PHARMD & CLINICAL MICRO TEAM
- ANNUAL ANTIBIOGRAMS

NCCLS GUIDELINES

- SELECTIVELY TEST ONLY DRUG/BUG COMBINATIONS WITH IN VIVO/IN VITRO CORRELATION
  - ✓ *Campylobacter, Bacillus, Corynebacterium*
    - NO ESTABLISHED CRITERIA
  - ✓ *Enterococcus*
    - Do not report cephalosporins, aminoglycosides, clinda, T/S
  - ✓ *Salmonella, Shigella*
    - Stool: ONLY test ampicillin, quinolone, T/S
    - Extraintestinal: above + chloramphenicol, 3rd gen cephalosporin
  - ✓ *Enterobacter, Serratia*
    - Do not report ampicillin & 1st & 2nd gen cephalo
    - Routine resistance
  - ✓ *Stenotrophomonas*
    - Inherent resistance to nearly all antimicrobics
    - ONLY Test T/S, Timentin & fluoroquinolone

DEFINING CLASS DRUGS

OXACILLIN *Staphylococcus*	Cephalosporins & penicillins
TETRACYCLINE All except (*Staph* & *Acinetobacter*)	Doxycline, minocycline, chlortetracycline
ERYTHROMYCIN Gram + cocci	Clarithromycin, azithromycin
CEPHALOTHINS Enterobacteriaceae	Cefazolin, cefaclor, cephalexin

WHAT ARE MIC VALUES?

- MINIMUM INHIBITORY CONCENTRATION (MIC)
  - ✓ LOWEST CONCENTRATION OF ANTIMICROBIC WHICH WILL INHIBIT GROWTH
- METHODOLOGIES
  - ✓ MICROBROTH DILUTION BY SEMI-AUTOMATED INSTRUMENTS, e.g. MICROSCAN, VITEK
    - 2-FOLD ANTIMICROBIC DILUTIONS
  - ✓ E-TEST
    - PLASTIC STRIPS-GRADIATED ANTIBIOTIC CONCEN
- MIC BREAKPOINTS SEPARATE SUSCEPTIBLE, INTERMEDIATE & RESISTANT STRAINS
- REFLECTS ACHIEVABLE SERUM CONCENTRATIONS OF THE DRUG

SIR INTERPRETATIONS

- SUSCEPTIBLE (S)
  - ✓ INFECTION BY THE STRAIN MAY BE APPROPRIATELY TREATED WITH THE DOSE OF ANTIMICROBIC
- INTERMEDIATE (I)
  - ✓ RESPONSE RATES MAY BE LOWER THAN FOR SUSCEPTIBLE ISOLATES
- RESISTANT (R)
  - STRAINS NOT INHIBITED BY THE USUALLY ACHIEVABLE SERUM CONCEN OF THE AGENT WITH NORMAL DOSING

PREDICTABLE SUSCEPTIBILITIES

PATHOGEN	RESISTANT
K. pneumo	Ampicillin
Enterobacter	Ampicillin, 1st/2nd cephalo
Salmonella Shigella	1st/2nd cephalo, aminoglycosides
MRSA	All penems, cephems, pip/tazo, other ß-lactams
Lactobacillus	Vancomycin
Listeria	Cephalosporins
Enterococcus	Aminoglycosides, Carbapenems, T/S,often Vancomycin (*E. faecium*)

ANTIMICROBIC SUSCEPTIBILITY TESTS (AST)

MICRO BROTH	DESCRIPTION	DETECTION
MICRO SCAN		
VITEK		

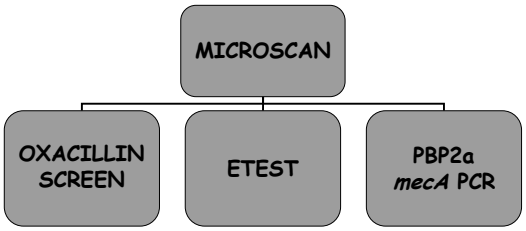
AST METHODS

AGAR	DESCRIPTION	DETECTION
E STRIP 		
KIRBY-BAUER		

MRSA PROFILE

- PENICILLIN INTRODUCED IN 1944
  - ✓ Plasmid-mediated resistance by β-lactamase that hydrolyzes β -lactam ring
  - ✓ Prevalent in hospitals in 1950s
- METHICILLIN INTRODUCED IN 1959
  - ✓ MRSA appeared in 1961 & prevalent in 1970s
  - ✓ Resistance from 4 Penicillin Binding Proteins (PBP) encoded by 4 *mec* genes (30-50 kb)
  - ✓ Chromosomal, not plasmid
  - ✓ MRSA acquired the *mec* A gene which codes for the production of unique PBP2a
- Oxacillin is the indicator drug for testing
  - ✓ *S.aureus* = MIC $\leq$ 2 ug/ml (S)
  - ✓ Coag Neg Staph = MIC $\leq$ 0.25 ug/ml (S)

MRSA DETECTION



STAPHYLOCOCCUS AUREUS WHAT'S UP DOC?

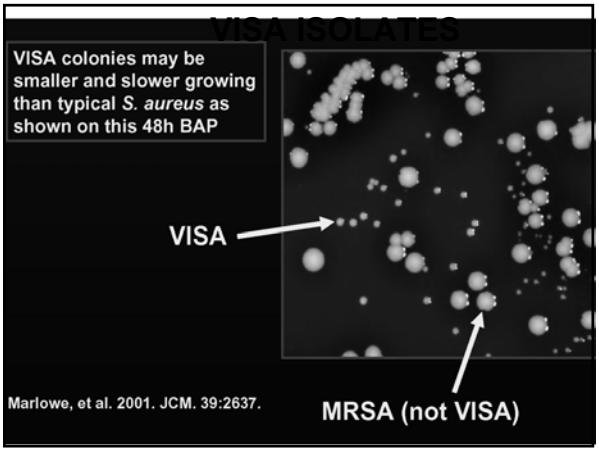
Clindamycin	S
Erythromycin	S
Oxacillin	R
Penicillin	R
Vancomycin	I/R

"Tu quoque, fili?" (You, my son, as well?)
Julius Caesar's outcry when he discovered Brutus, his adopted son, was ready to stab him.
Analogy: Vancomycin, now, as well?

VANCOMYCIN & STAPH

- Vanco is traditional MRSA treatment
  - ✓ 3-4% Hypersensitivity, no p.o.
- Vanco non-susceptible rare
  - ✓ VISA (11) and VRSA (3)
  - ✓ Linezolid (CAP, other infections), daptomycin (skin & soft tissue) are alternatives
- MIC Breakpoints to VANCOMYCIN
  - ✓ SUSCEPTIBLE ≤ 4 ug/mL
  - ✓ INTERMEDIATE 8-16 ug/mL
  - ✓ RESISTANT ≥ 32 ug/mL
- Retest *S. aureus* with MIC of ≥4 µg/ml & use alternate method
  - ✓ Vancomycin agar screen plates (test all MRSA), Etest, reference lab
- Disk test will NOT detect VISA

VISA ISOLATES



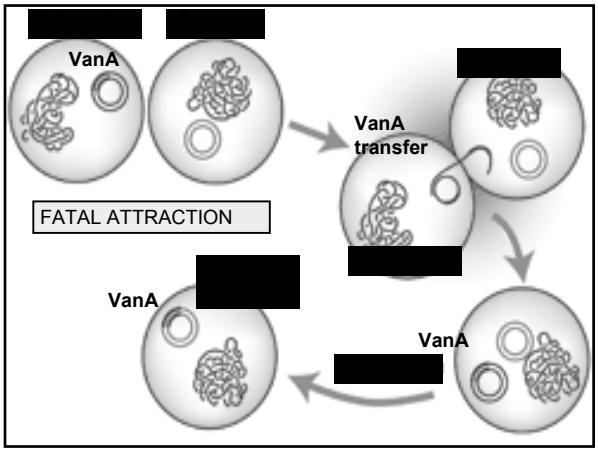
VISA

- VISA– INTERMEDIATE TO VANCO
  - ✓ 1ST ISOLATED IN 1996 IN JAPAN
  - ✓ 8 PTS TO DATE IN USA
  - ✓ MECHANISM OF RESISTANCE: THICKENED CELL WALL AND/OR AN EXTRACELLULAR MATRIX ??
  - ✓ PATIENTS HAD PRIOR EXPOSURE TO LONG TERM VANCOMYCIN THERAPY
- 2 VISA ISOLATES FOUND SUSCEPTIBLE TO OXACILLIN
  - ✓ ONE WAS *MECA* POS & ONE NEG
  - ✓ OXACILLIN RESISTANCE IS NOT NECESSARY FOR VISA PHENOTOYPE
- NO CLONAL SPREAD OF SINGLE STRAIN

VRSA JUNE 2002

- 1st case in 40 yr old diabetic woman from Michigan
- VRSA from dialysis cath tip
- Recurrent foot ulcer infected with VRE & MRSA

- THE USA VRSA ISOLATE
  - ✓ MRSA
  - ✓ VANCOMYCIN MIC 1,024 ug/mL
  - ✓ CONJUGATIVE TRANSFER
  - ✓ VRSA HAD *vanA* & *mecA*
  - ✓ *vanA TRANSPOSON JUMPED FROM VRE PLASMID TO MRSA→ VRSA*



VRSA NYC CASE

- March 17, 2003
- VRSA isolate from nursing home resident
- Initially called vanco susceptible by MicroScan MIC= 2 µg/mL
  - ✓ Vanco Screen plate showed resistance
  - ✓ ETest MIC = > 256 µg/mL
  - ✓ Strain had both mecA and vanA genes

  ALL SA HAVE VANCO SCREEN PLATE AS CONFIRMATORY TEST FOR VR

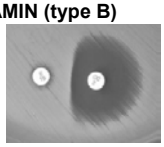
STAPHYLOCOCCUS AUREUS CLINDAMYCIN INDUCED RESISTANCE

MECHANISM	DETERMINANT (GENE)	ERY	CLINDA
EFFLUX			
RIBOSOMAL ALTERATION			

REQUIRES INDUCTION TO BE DEMONSTRABLE

MACROLIDE RESISTANCE

- MLSB
  - ✓ MACROLIDE LINCOSAMIDE (e.g. CLINDAMYCIN) STREPTOGRAMIN (type B)
  - ✓ R MEDIATED BY *erm* GENE
  - ✓ RIBOSOMAL METHYLATION
  - ✓ INDUCIBLE ($MLSB_i$)
  - ✓ CONSTITUTIVE ($MLSB_c$)



- ALSO APPLICABLE FOR GROUP B STREP

ENTEROCOCCI

- COMMENSAL ORGANISM
  - ✓ INFECTION OR COLONIZATION
- RESISTANCE
  - ✓ INTRINSIC R (aminoglycosides & β-lactams)
  - ✓ ACQUIRED R (chloramphenicol, tetracycline, macrolides, quinolones)
  - ✓ SOURCE OF R GENES
- INFECTIONS
  - ✓ CLINICAL
  - ✓ NOSOCOMIAL
    - INFECTION CONTROL
      - VRE SCREENING (PERI-RECTAL/ANAL SWABS)
      - MOLECULAR TYPING TO DETERMINE CLONAL SPREAD

ENTEROCOCCI: LAB TESTING

- ANTIBIOTICS
  - ✓ AMPICILLIN MIC, β–LACTAMASE, VANCO SCREEN, OTHERS (e.g. Linezolid)
- SYNERGY SCREEN
  - ✓ BLOOD ISOLATES TEST
    - COMBINATION OF β -LACTAM (e.g. PENICILLIN OR VANC WITH AN AMINOGLYCOSIDE (GENT OR STREP) BACTERICIDAL
    - HLG (Gentamicin 500 ug/mL);Strep (2000 ug/mL)

VANCOMYCIN-RESISTANT ENTEROCOCCI (VRE)

- SPECIATION NECESSARY
  - ✓ Intrinsic resistance (*E. gallinarum* & *E. casseliflavus*)
  - ✓ Acquired resistance (*E.faecium* & *E.faecalis*; also in *E.raffinosus, E.avium, E.durans*)
- Higher Vanco R in *E. faecium* vs. *E. faecalis*
  - ✓ 8% (*E.faecalis*) & 80% (*E.faecium*) CUMC 2003

GENE	VANCO (ug/mL)
Van A	>128
Van B	16-64
Van C (Intrinsic)	2-16
Van D	64-128

EXTENDED SPECTRUM ß-LACTAMASES

- FIRST DESCRIBED IN 1983
- ESBLS ARE ß-LACTAMASES THAT MEDIATE R TO
  - ✓ 3rd generation cephalosporins, (e.g cefotaxime, ceftriaxone, ceftazidime) but these can appear susceptible when tested in lab
  - ✓ Monobactams (e.g. aztreonam)
  - ✓ Extended spectrum penicillins (e.g. piperacillin)
- STRUCTURAL GENES
  - ✓ PLASMID- MEDIATED
    - Altered configuration of TEM-1 & 2, SHV-1 near active sites to increase hydrolytic ability for cephalosporins
    - Susceptible to cefoxitin (cephamycin), ß-lactamase inhibitors (but enzyme hyperproduction might overwhelm inhibitors)
    - Susceptible to carbapenems
  - ✓ CHROMOSOME-MEDIATED
    - AmpC in SPICE (*Serratia, Pseudo, Proteus, Citro, Enterobacter*) Also have plasmid-mediated AmpC
    - K1 in *K. oxytoca*
    - Resistant to cefoxitin (cephamycin) & ß-lactamase inhibitors

CARBAPENEM R

- Carbapenems (imipenem, meropenem)
  - ✓ Used as antibiotics of last resort for multidrug-resistant GNR
  - ✓ Drug of choice for ESBL producers
- Mechanisms include
  - ✓ Altered porins, metallo-ß-lactamases or other carbapenemases
- Etest strips
- More likely found in *Pseudomonas* or *Acinetobacter*
  - ✓ Polymyxin is drug of last resort

AMINOGLYCOSIDE R

- Aminoglycosides (e.g. gentamicin, tobramicin, amikacin)
  - ✓ Used as antibiotics usually in combination with β-lactams
  - ✓ Drug of choice for *Enterobacteriaceae* or *P. aeruginosa*
- Mechanisms include
  - ✓ Inactivation of drug by aminoglycoside-modifying enzymes (AME's), ribosomal alterations, efflux, permeability loss
  - ✓ AME's most common. Can be passed via plasmids & transposons

TOUGH BUGS ON THE BLOCK

- Resistant Staph: MRSA, VISA, VRSA
  - ✓ Cost to treat MRSA 3X MSSA
  - ✓ 44% MRSA CUMC
- 18% ESBLs CUMC
- 81% VRE (*E. faecium*)
- Metallo-ß-lactamases
  - ✓ *Acinetobacter baumannii*
  - ✓ *Pseudomonas aeruginosa*
  - ✓ *Stenotrophomonas maltophilia*
- Penicillin R *S. pneumoniae*
  - ✓ 48% Susceptible
  - ✓ 24% Low Level Resistance
  - ✓ 28% High Level Resistance

FUTURE NIGHTMARES

- Widespread Linezolid resistance in VRE and Staph
- *Van* A gene transfer to all *S. aureus* to result in increase in VRSA
- Spread of metallo-ß-lactamases in nosocomial GNR carbapenem resistant GNR
- Depletion of antimicrobial agents
  - ✓ Few new classes, e.g. ketolides (telithromycin) for RTIs