

# What are Beta-lactamases?

Proteins degrading Beta-lactam's

# The Beta-lactam antibiotics

- Isolated from *Penicillium chrysogenum*
- App. 50 % of the antibiotics used worldwide
- The Beta-lactam group is constantly expanding
- Is now being produced semi-synthetically
- Kills growing cells by interfering with the cell-wall synthesis
- One of the most important veterinary and human antibiotics.

# Narrow spectrum vs. Extended spectrum Beta-lactam's

## Narrow and moderate spectrum BL's

- Penicillin G and V (PEN)
- Methicillin (MET)
- amoxicillin (AMOX) and ampicillin (AMP)
- Cephalotin (CEP)

## Broad and Extended spectrum BL's

- Cefoxitin (FOX)
- Cefotaxime (CTX) and Ceftazidime (CAZ)
- Cefepime (FEB)
- Imipenem (IMI)

## What are ESBL's?

- Divided into: **ampC's**, “True **ESBL**” and **Metallo-BL's**.
- Able to **degrade** broad and extended spectrum beta-lactam's
- First identified 22 years ago (SHV-2)
- ESBL and plasmidic ampC's mainly in **Enterobacteriaceae**
- **Metallo-BL** mainly in *Pseudomonas*
- Now >200 different genes
- Approximately 20 different groups
- Big difference in homology
- Seen in all **environments** where extended spectrum beta lactam's are being used

## Beta-lactamase genes so far...

### Plasmidic AmpC's

CMY

ACC

DHA

FOX

BIL

MIR

ACT

KLU

### ESBL

TEM

SHV

OXA

CTX-M

VEB

PER

CME

SFO

FEC

GES

### MBL

IMP

VIM

SPM

GIM

Pitfalls:

- Reduced susceptibility can be caused by up-regulated efflux-pumps or defective influx pumps
- *E. coli* carries a down-regulated ampC beta-lactamase, which can be activated (up-regulated) by two mutations

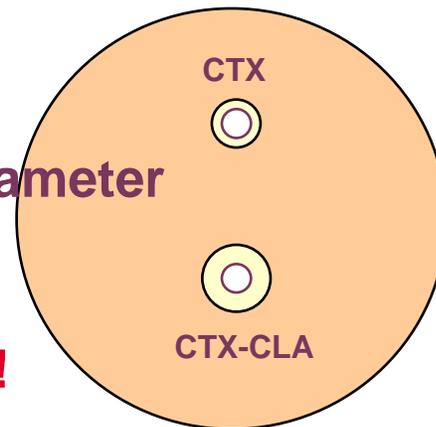
# The three different ESBL groups I

## The 'true' ESBL's

- Often located on transferable plasmids/elements
- Often found in bacteria with no chromosomal AmpC
- Rarely resistant to inhibitors (results in 'synergy effect')
- Resistant to both 3rd and 4th generation cephalosporins
- Inducible by beta-lactams

Difference in diameter  
> 5mm

**Synergy!**



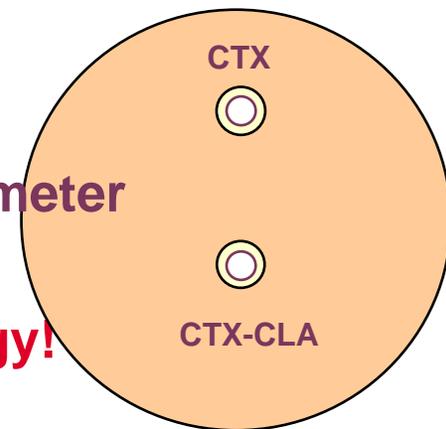
## The three different ESBL groups II

### The AmpC's

- Often located on chromosomes (*E. coli*, *Citrobacter*, *Enterobacter*)
- ...or on plasmids but originating from chromosomal versions
- Confers resistance to beta-lactam inhibitors (=> no 'synergy')
- Confers resistance to cefoxitin (FOX); a 2nd generation cephamycin
- Sensitive to 4th generation ceph's (like cefepime (FEB))
- Not inducible by beta-lactams

Difference in diameter  
< 5mm

No synergy!



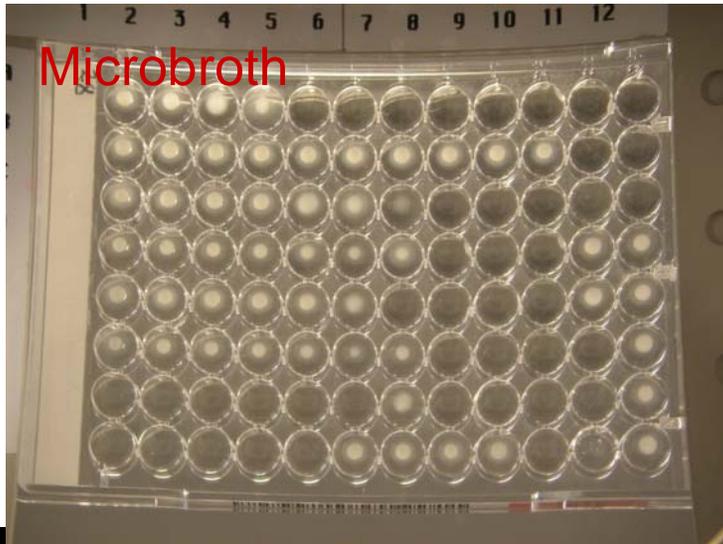
# The three different ESBL groups III

## The Metallo beta-lactamases

- Can be inhibited by metal chelators (like EDTA)
- Are mainly found in *Pseudomonas*
- Confers resistance to all generations of cephalosporins
- Confers resistance to carbapenems like imipenem
- Rarely found in Enterobacteriaceae

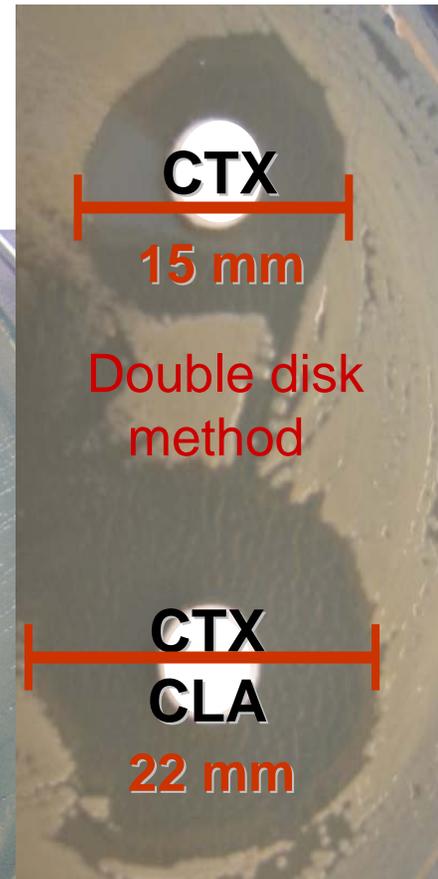
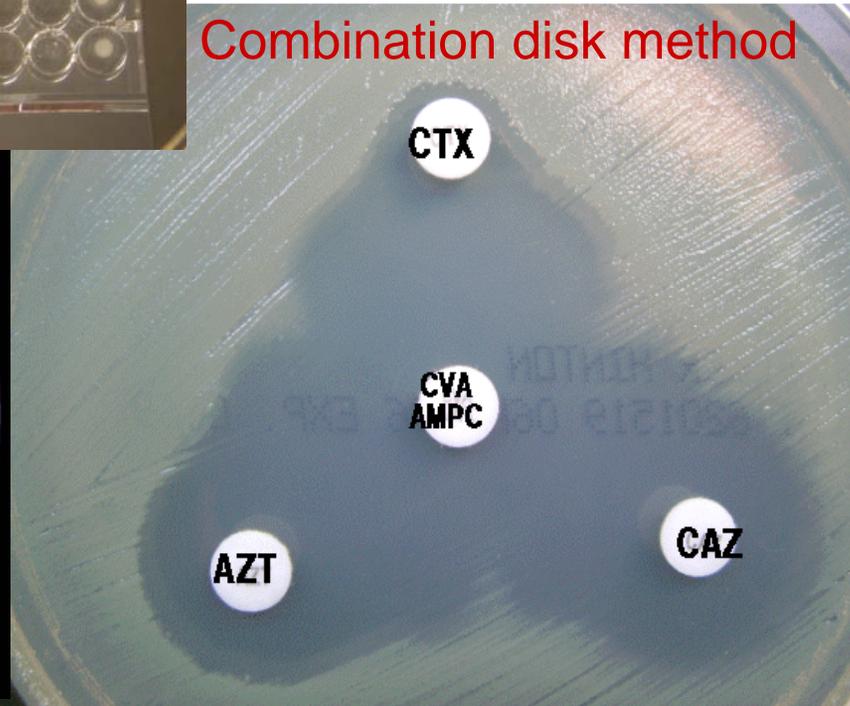
# How to detect ESBL? I

Phenotypically:



Microbroth

Combination disk method



CTX

15 mm

Double disk  
method

CTX

CLA

22 mm



E-test

## How to detect ESBL? II

### Initial screening test

Cefpodoxime  
Ceftazidime  
Aztreonam  
Cefotaxime  
Ceftriaxone

(The use of **more than one** antimicrobial agent for screening will improve the sensitivity of detection)

### Phenotypic confirmatory test

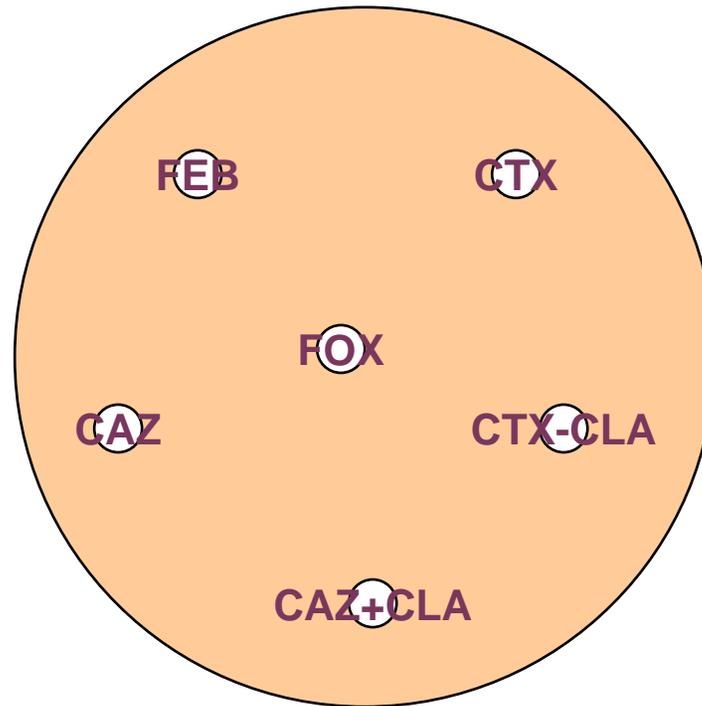
Ceftazidime (CAZ)  
CAZ + clavulanic acid  
and  
Cefotaxime (CTX)  
CTX + clavulanic acid

(Confirmatory testing requires use of **both cefotaxime and ceftazidime alone and in combination with clavulanic acid**)

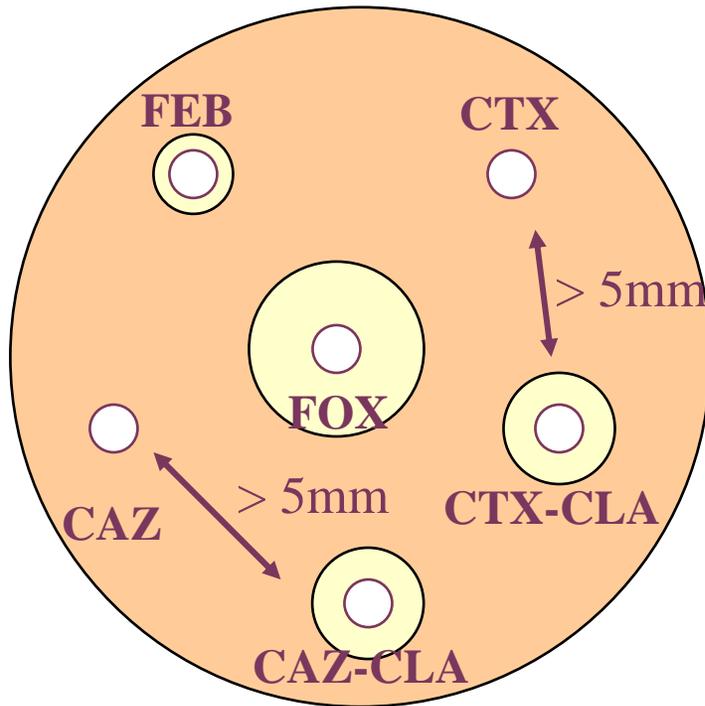
(CLSI M100-S18: App. A)

Synergy (CLSI):  $A \geq 3$  two-fold reduction in MIC (e.g. from 8 to 1  $\mu\text{g/ml}$ ) or a  $\geq 5\text{mm}$  increase in a zone diameter between CAZ and CAZ/Cl or CTX and CTX/Cl

# ESBL-tablet assay

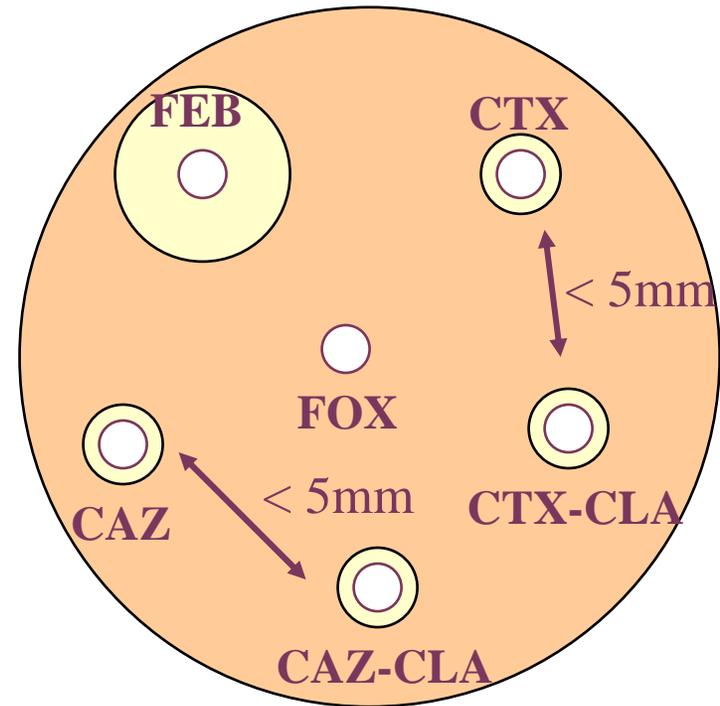


## ESBL



Synergy!

## ampC

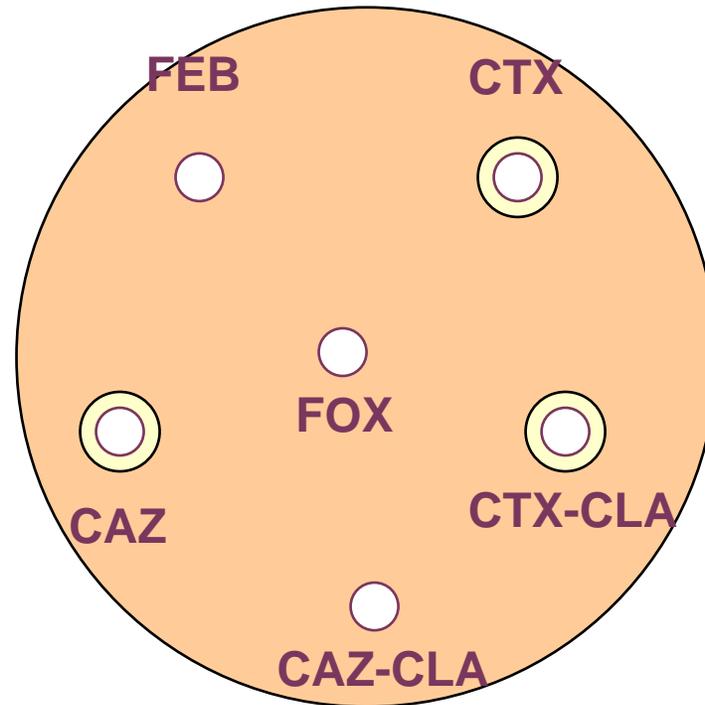


No synergy!

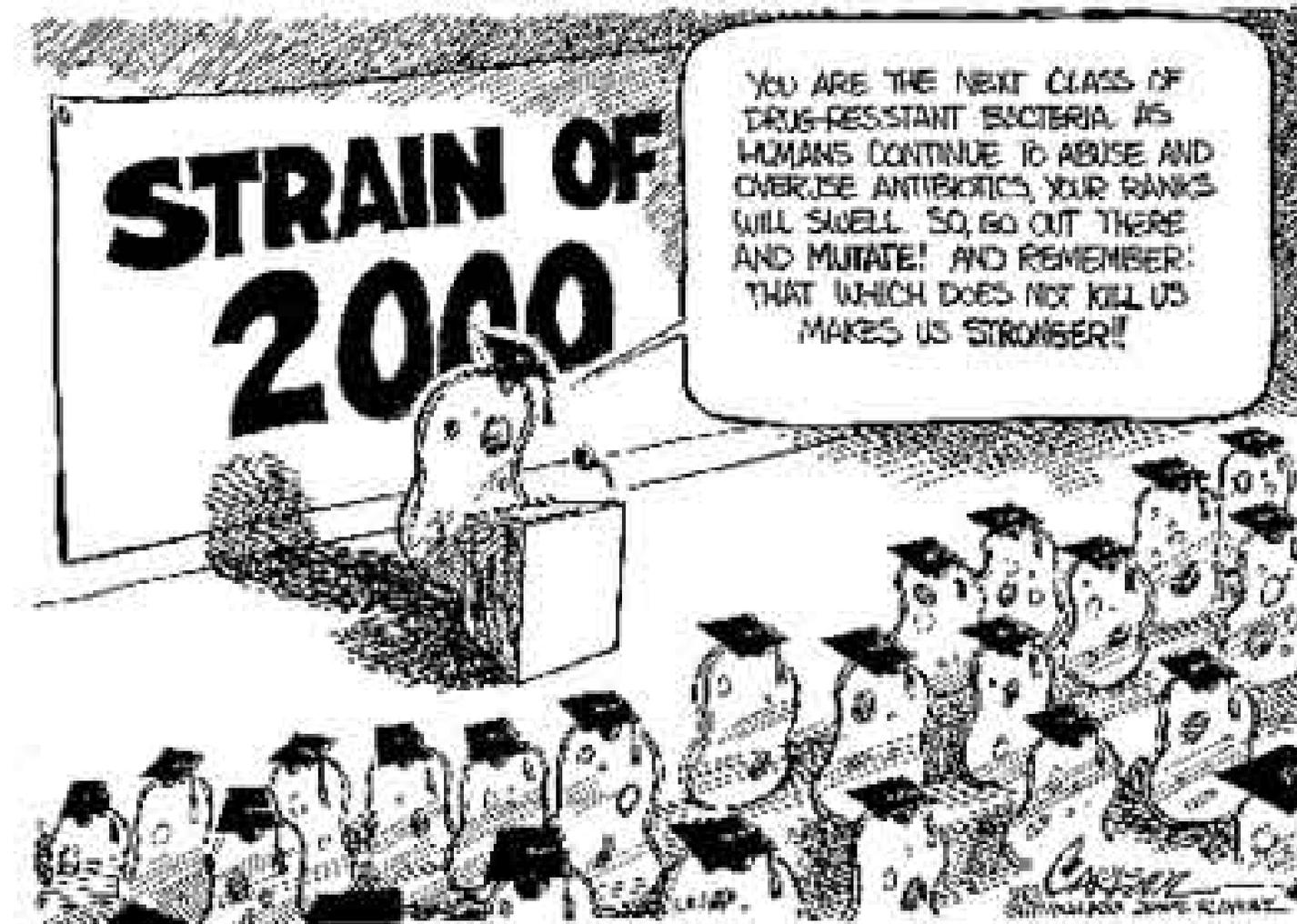
## But but but...

Different beta-lactamases can have different affinity towards different beta-lactams!

And one strain can easily have more than one beta-lactamase!



## ESBL's - an issue that requires our attention!



# Acknowledgement

Thanks to  
**Henrik Hasman and  
Susanne Karlsmose**  
for sharing slides



## Help Your Antibiotics Do Their Job

- Take as directed
- Finish the full prescription even if you are feeling better
- Help prevent antibiotic resistance

Thank you!

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