

Antifungal susceptibility test by Etest

Azian Harun

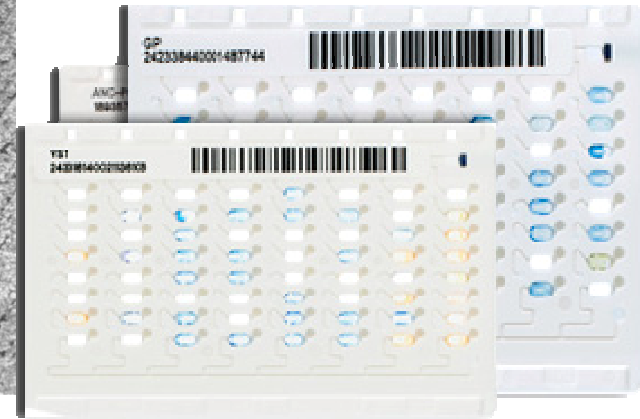
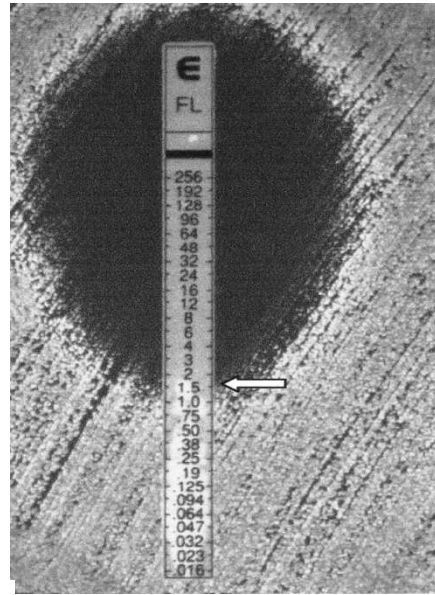
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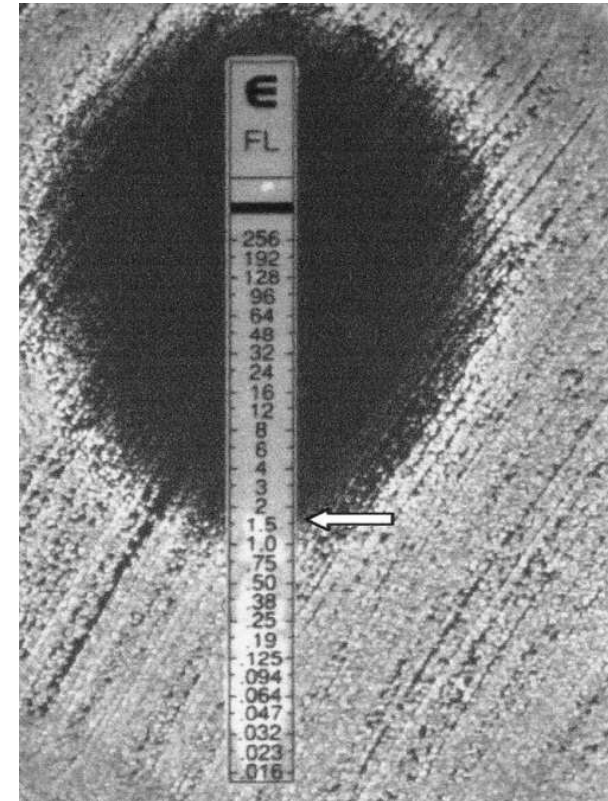


Antifungal susceptibility test



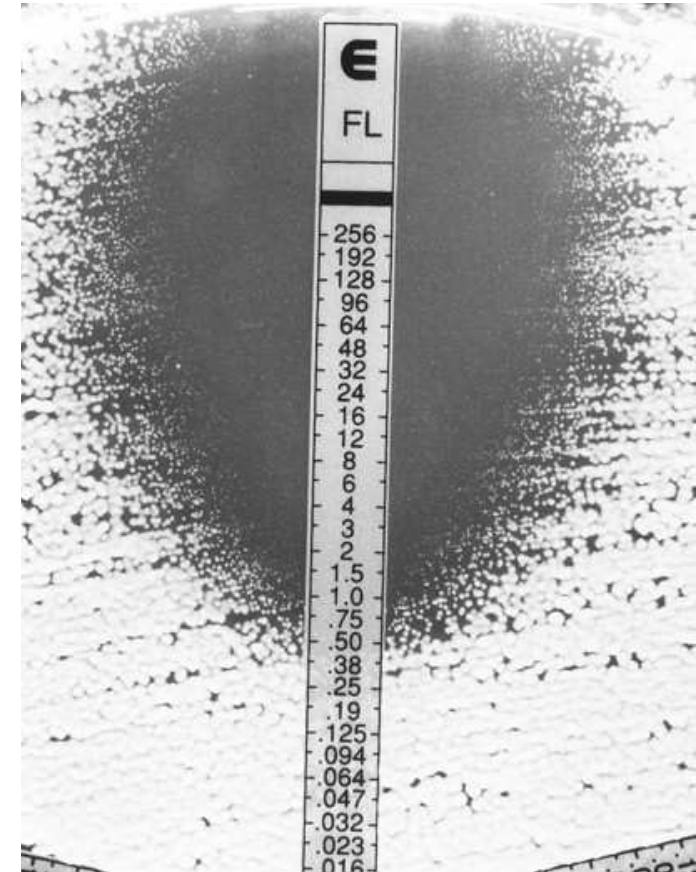
Etest

- Etest is based on a combination of the concepts of dilution and diffusion tests.
- Directly quantifies antifungal susceptibility in terms of discrete MIC values.



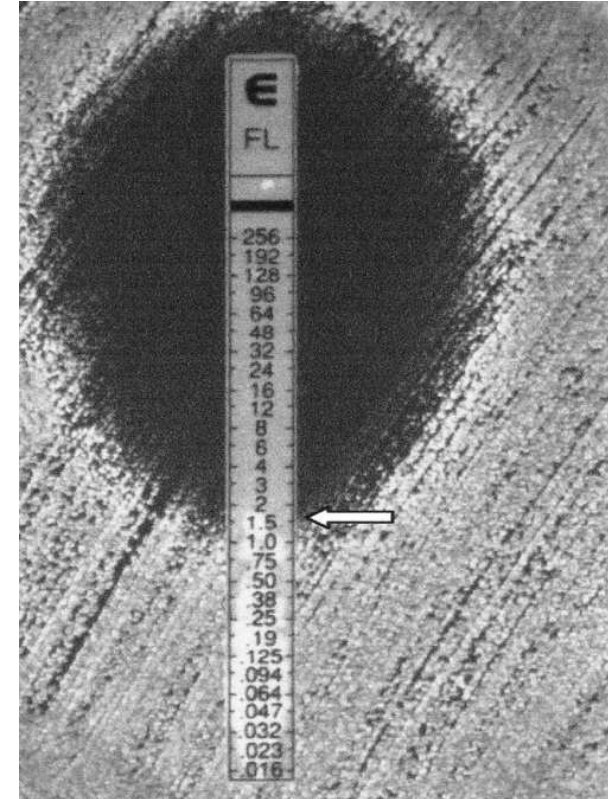
Etest

- Etest consists of a thin, inert and non-porous plastic strip (5 x 60 mm).
- One side of the strip is calibrated with MIC values in $\mu\text{g/ml}$
- Two-letter code designates the identity of the drug
- Predefined and exponential gradient of the dried and stabilised antifungal agent - immobilised on the other surface of the strip



Etest

- When applied onto an inoculated agar plate, there is an immediate release of the agent from the plastic surface into the agar matrix.
- After incubation, whereby growth becomes visible, a symmetrical inhibition ellipse centred along the strip is seen.
- The zone edge intersects the strip at the MIC value.



Etest

Medium

- RPMI 1640 broth + MOPS
+ 2% Glucose + 1.5% agar



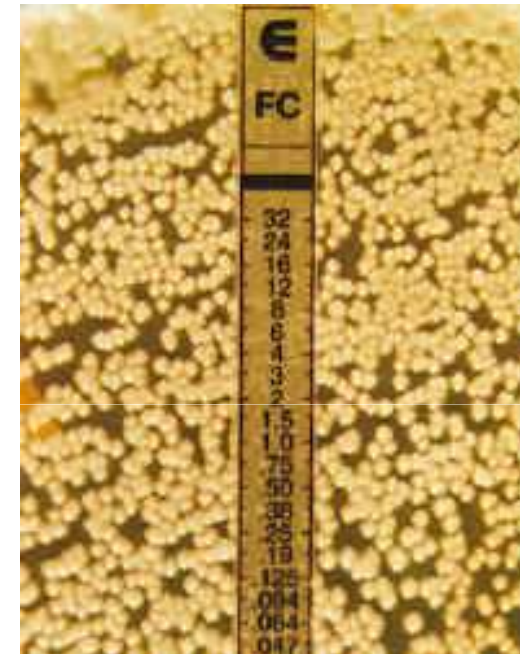
Etest - reading

Flucytosine (read at almost complete inhibition, ignoring small micro-colonies)



MIC 1 mg/ml

MIC >32 mg/ml
(macrocolonies in ellipse)

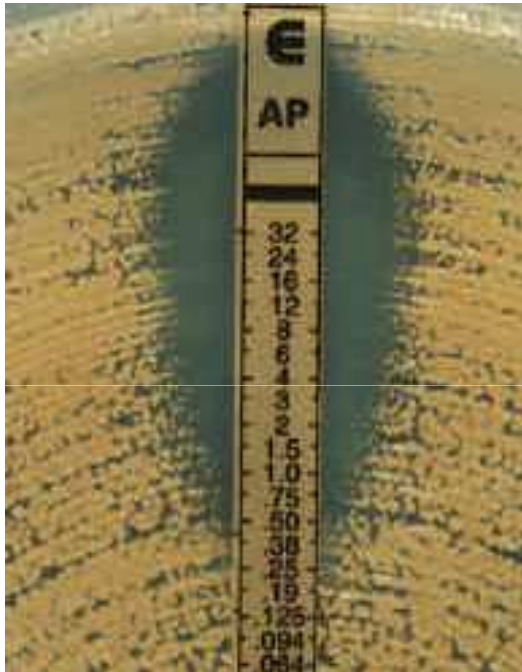


MIC >32 mg/ml
(homogenously resistant)



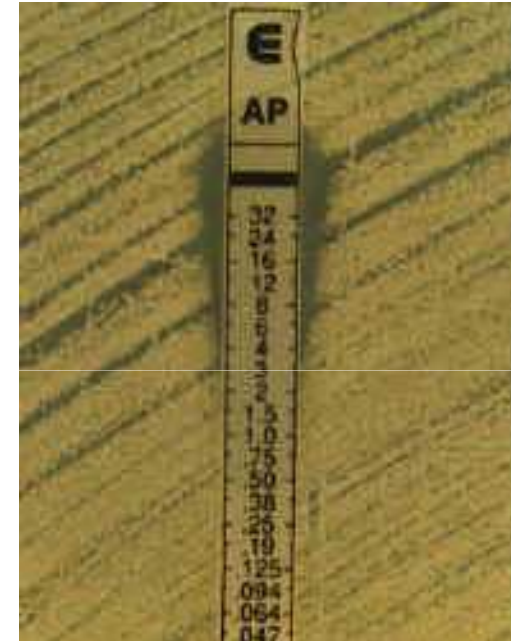
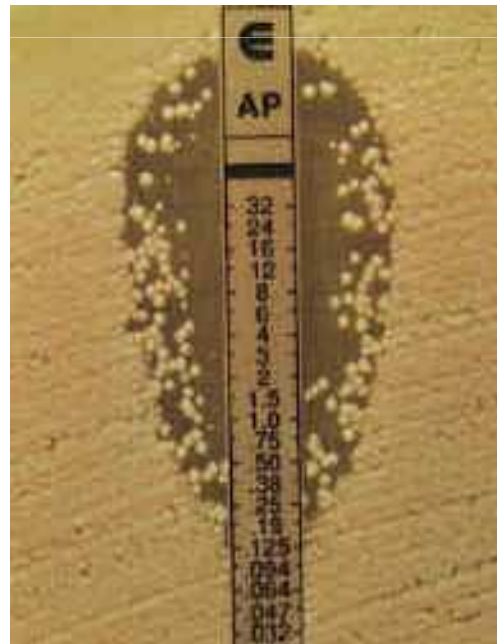
Etest - reading

Amphotericin (read at complete inhibition of all growth)



MIC 1 µg/ml (include all microcolonies)

MIC 32 µg/ml (macrocolonies in ellipse)

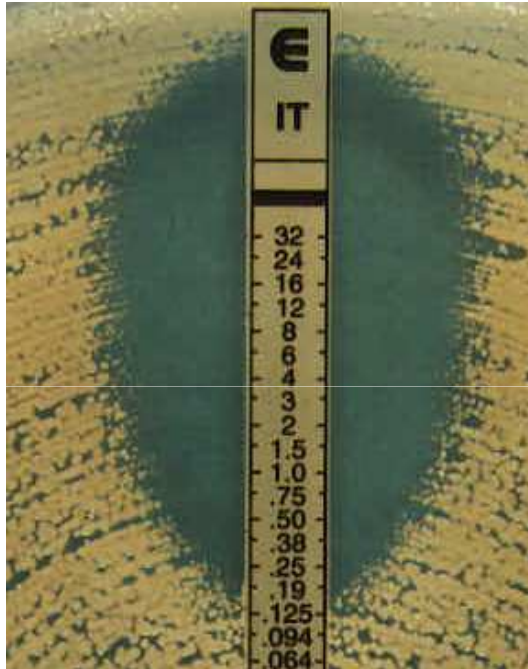


MIC 4 µg/ml (small and slim ellipse)



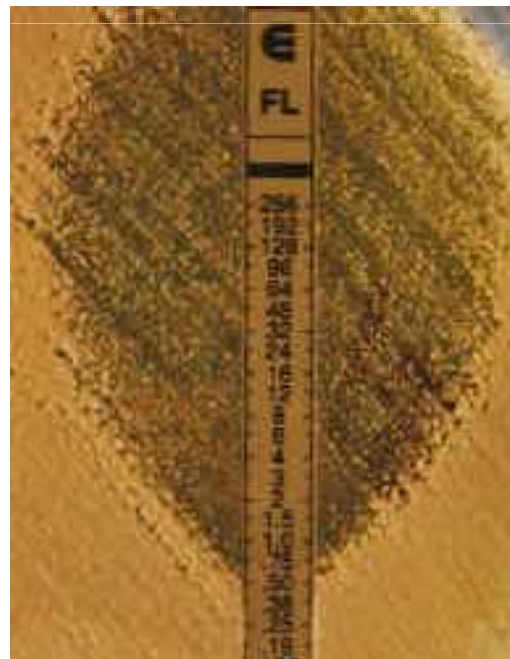
Etest - reading

Azoles (read at first point of significant inhibition i.e. so-called 80% inhibition)



MIC 0.5 $\mu\text{g/ml}$
(lawn of microcolonies within a discernable ellipse)

MIC 0.125 $\mu\text{g/ml}$
(sharp end point)



MIC >256 $\mu\text{g/ml}$
(macrocolonies in ellipse)



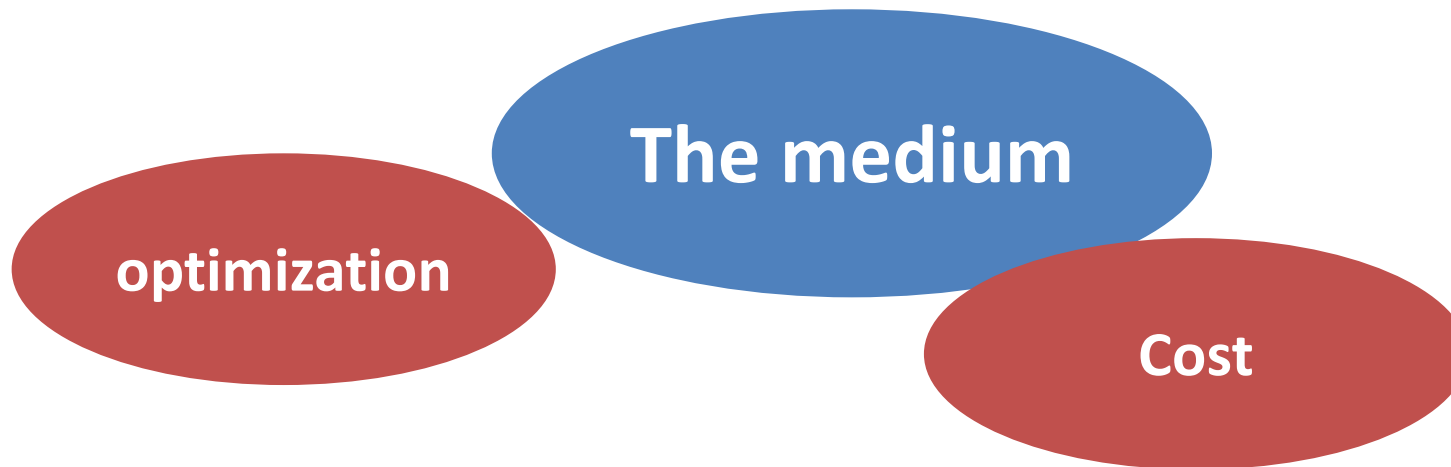
Etest - problems

QC

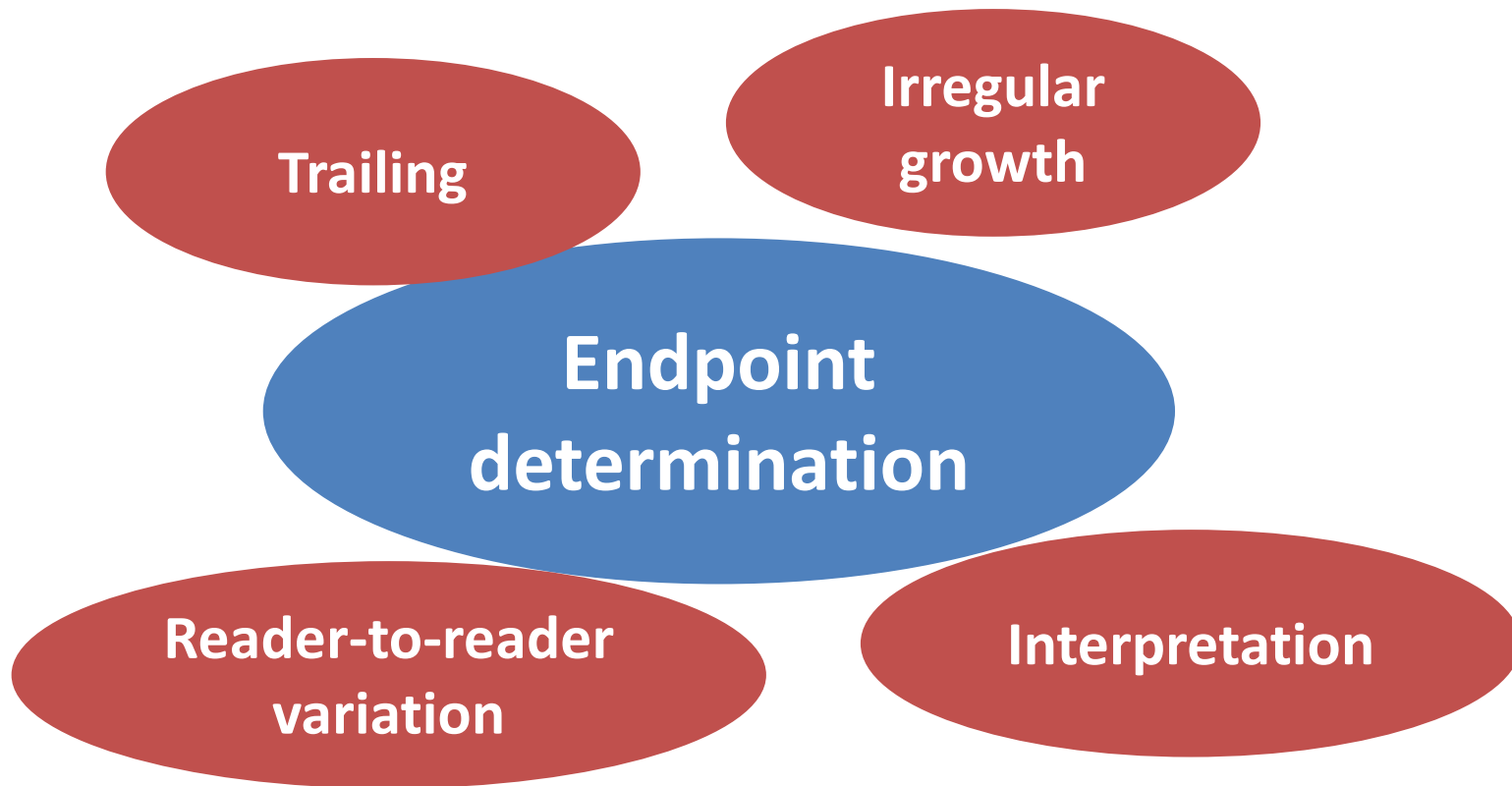
C. krusei ATCC 6258

C. parapsilosis ATCC 22019

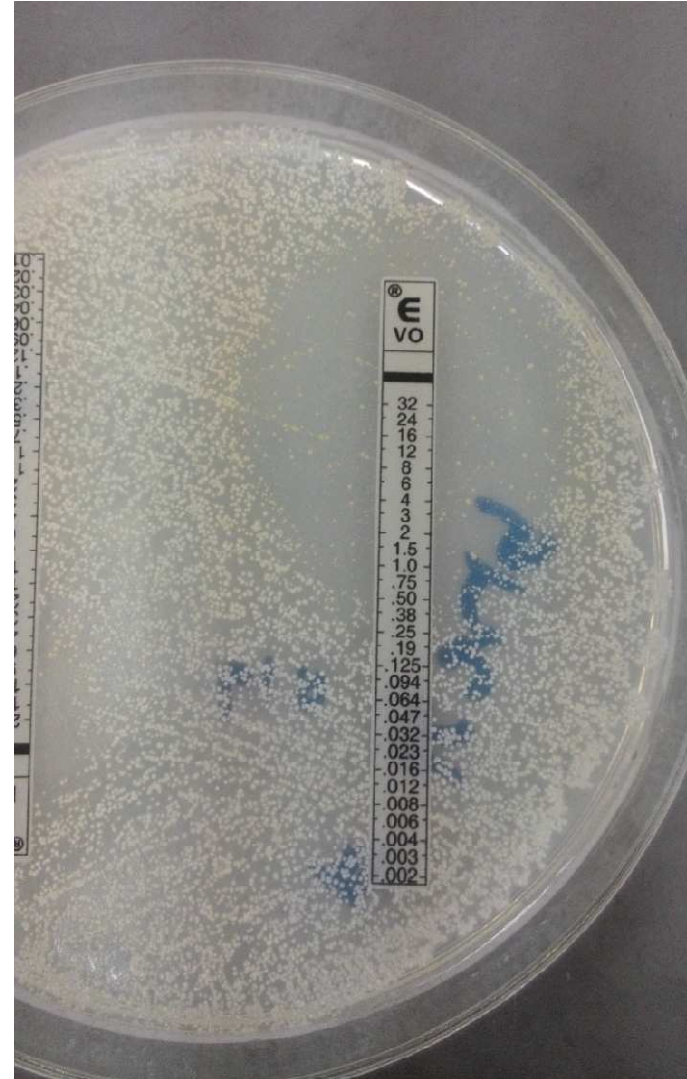
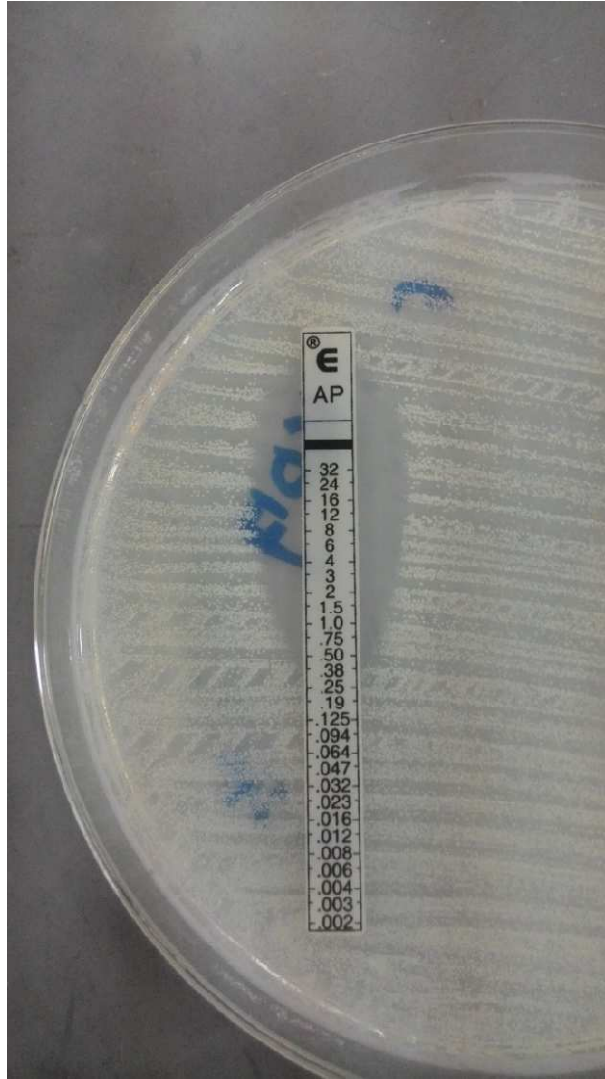
C. albicans ATCC 90028



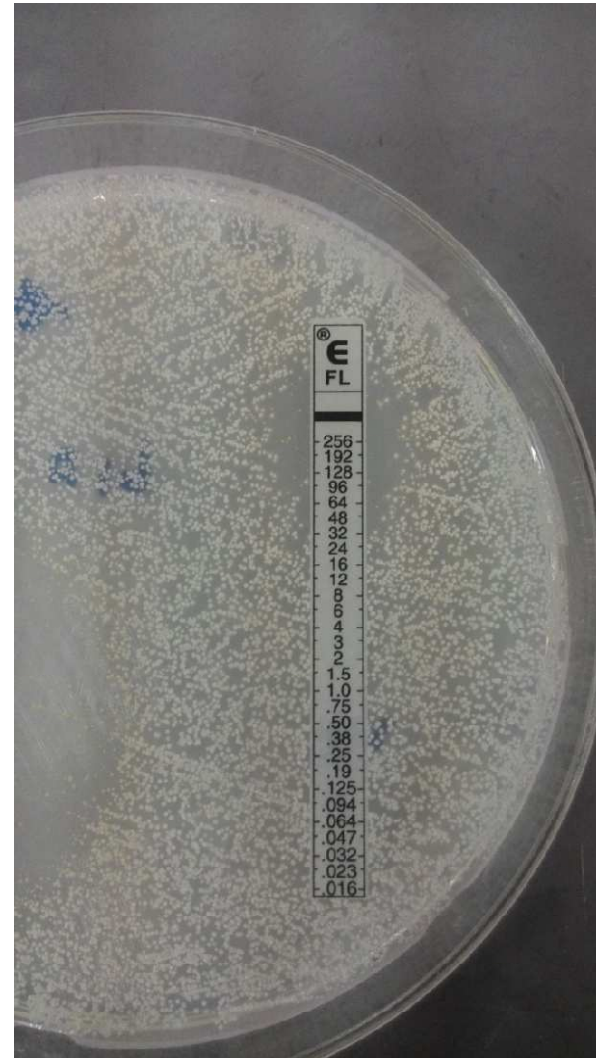
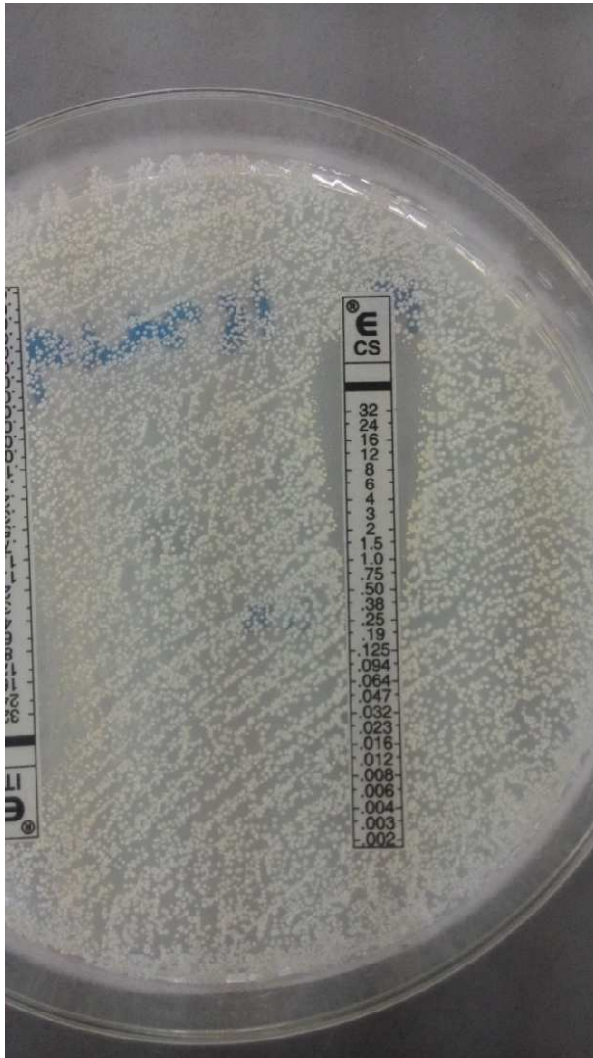
Etest - problems



Etest – skills & experience



Etest – skills & experience



Etest – interpretation!!

- Headache... headache...
- 95% agreement with broth microdilutions for FLU, VOR
- MIC by Etest – generally a bit lower than broth dilutions

Chen et al. 1996, Maxwell et al. 2003



Conclusion

Antifungal
susceptibility by
Etest - ok

Needs
training

Needs SOP &
guidelines

- Media prep.
- Reading &
interpretation

Epidemiological
cut off
Local data
analysis

