

# ECOFFs

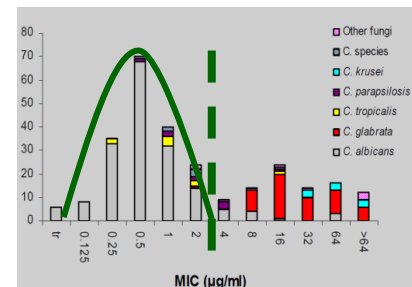
## Use in antifungal susceptibility testing

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**Unit of Mycology**  
**Statens Serum Institut**  
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# In the ideal world we have....

## ■ MIC distributions

- Per species
- Several data sets
- Epidemiological Cut Off Value (ECOFF)



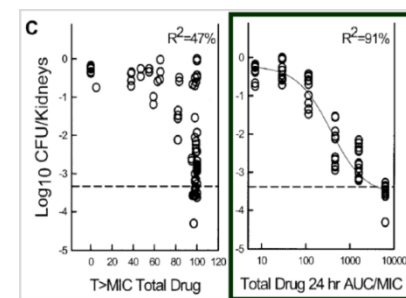
## ■ MIC-clinical outcome relationships

- Per species
- For wild type and non-wild type isolates

MIC in mg/L	Candidaemia		OPC ≥ 100 mg/d		All response
	No. cure/Total	% response	No. cure/Total	% response	
< 0.5	98/107	92	26/26	100	93
1	6/6	100	4/4	100	100
2	1/1	100	1/1	100	100
4	3/3	100	5/9	56	67
8	2/5	40	7/32	22	24
≥16	3/4	75	0/60	0	5

## ■ PK/PD

- defining the important parameter and ratio (AUC/MIC, Peak/MIC, Time/MIC)
- are non wild type covered during standard dosing?



# In real life...

- ... we often don't..
- still, there is a need to translate the MIC into clinical guidance
- incomplete data/evidence is better than nothing!
  
- AFST has established clinical Breakpoints on existing data
  - minimum: ECOFFs and clinical evidence the wild type organism respond to treatment

# EUCAST *Candida* Breakpoints 2013

Breakpoints (BPs): S:  $\leq X$  / R:  $> Y$

Antifungal agent	MIC breakpoint (mg/l)						
	<i>C. albicans</i>	<i>C. glabrata</i>	<i>C. krusei</i>	<i>C. parapsilosis</i>	<i>C. tropicalis</i>	<i>C. guilliermondii</i>	Non-species related breakpoints
Ampho. B	1/1	1/1	1/1	1/1	1/1	IE	IE
Anidulafungin	0.03/0.03	0.06/0.06	0.06/0.06	<u>I/R cat*</u>	0.06/0.06	IE <sup>2</sup>	IE
Fluconazole	2/4	<u>I/R cat*</u>	-	2/4	2/4	IE	2/4
Micafungin	0.016/0.016	0.03/0.03	IE <sup>2</sup>	0.002/2	IE <sup>2</sup>	IE <sup>2</sup>	IE
Posaconazole	0.06/0.06	IE <sup>2</sup>	IE <sup>2</sup>	0.06/0.06	0.06/0.06	IE <sup>2</sup>	IE
Voriconazole	0.125/0.125 <sup>4</sup>	IE	IE	0.125/0.125 <sup>4</sup>	0.125/0.125 <sup>4</sup>	IE <sup>2</sup>	IE

<sup>2</sup> denotes the MICs for this species is generally higher than those for *C. albicans*

<sup>4</sup> denotes that voriconazole should be reserved for situations where fluconazole is not appropriate

- denotes the organism is regarded a poor target for the antimicrobial agent

## Revisions on consultation:

- \* For *C. glabrata* and fluconazole: S:  $\leq 0.002$  mg/l and R:  $>32$  mg/l
- \* For *C. parapsilosis* and anidulafungin: S:  $\leq 0.002$  mg/l and R:  $>4$  mg/l

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	<i>C. albicans</i>	<i>C. glabrata</i>	<i>C. krusei</i>	<i>C. parapsilosis</i>	<i>C. tropicalis</i>	<i>C. guilliermondii</i>	Non-species related breakpoints
Ampho. B	1/1	1/1	1/1	1/1	1/1	IE	IE
Anidulafungin	0.03/0.03	0.06/0.06	0.06/0.06	<u>I/R cat*</u>	0.06/0.06	IE <sup>2</sup>	IE
Fluconazole	2/4	<u>I/R cat*</u>	-	2/4	2/4	IE	2/4
Micafungin	0.016/0.016	0.03/0.03	IE <sup>2</sup>	0.002/2	IE <sup>2</sup>	IE <sup>2</sup>	IE
Posaconazole	0.06/0.06	IE <sup>2</sup>	IE <sup>2</sup>	0.06/0.06	0.06/0.06	IE <sup>2</sup>	IE
Voriconazole	0.125/0.125 <sup>4</sup>	IE	IE	0.125/0.125 <sup>4</sup>	0.125/0.125 <sup>4</sup>	IE <sup>2</sup>	IE

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Different from  
the ECOFF  
(one step higher)

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# Micafungin breakpoint setting

## ■ Step one – ECOFFs! (7 dataset available)

Species (no.)	0.004	0.008	0.016	0.031	0.063	0.125	0.25	0.5	1	2	4	ECOFF	No. (%) above the ECOFF
<i>C. albicans</i> (1569)	290	273	241	4	5							0.016	49 (3.1%)
	ND	ND	ND	107									
	ND	ND	520	35	2	1			2				
	ND	87	2										
<i>C. glabrata</i> (692)	90	182	100	33	3	5 (1/1)				2		0.03	12 (1.7%)
	ND	ND	ND	100									
	ND	ND	173	2				1	1				
<i>C. krusei</i> (483)	1		4	26	185	215	37	9	2 (2/2)	4		0.25	15 (3.1%)
<i>C. parapsilosis</i> complex(743)			3	1		1	35	113	332	244	14	2	14 (1.9%)
<i>C. tropicalis</i> (732)	48	51	247	200	59	14	2 (2/2)	1 (1/1)	6 (4/4)	4 (4/4)		0.06	29 (4.0%)
	ND	ND	ND	98		1				1 (1/1)			
<i>C. guilliermondii</i> (22)						1	5	7	8	1		ND	

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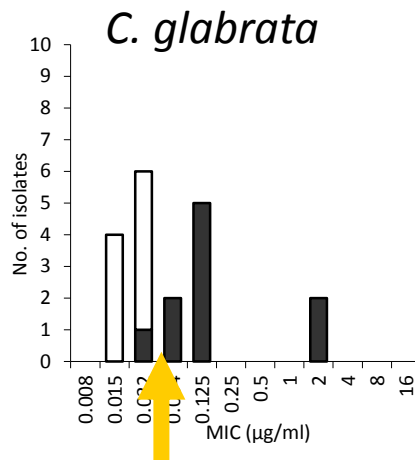
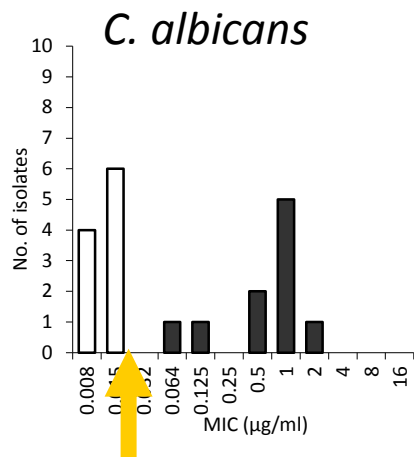
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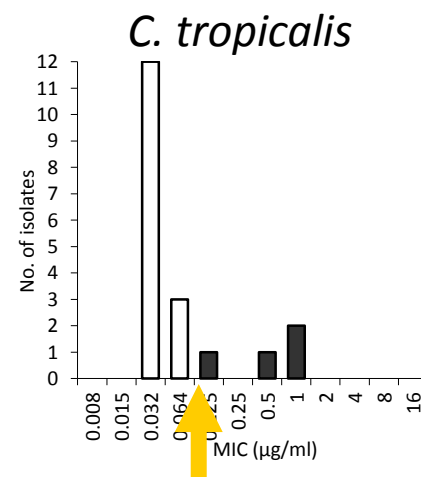
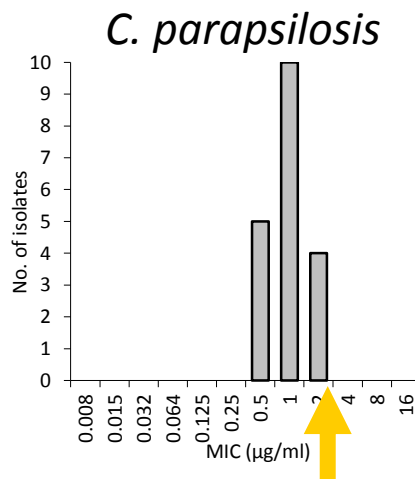
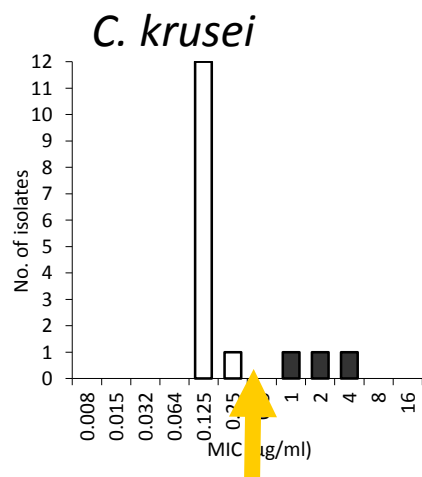
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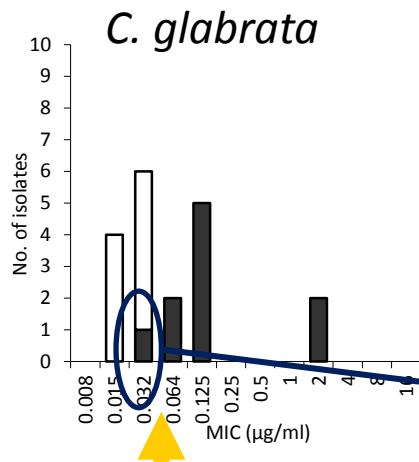
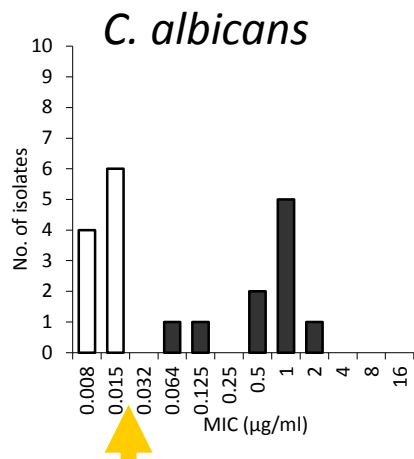
# Experience with *FKS* mutants



↑ Indicates the ECOFF

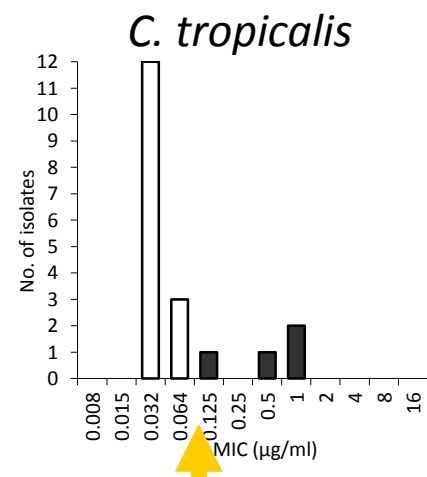
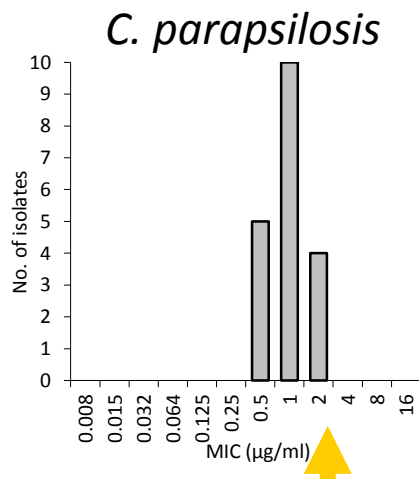
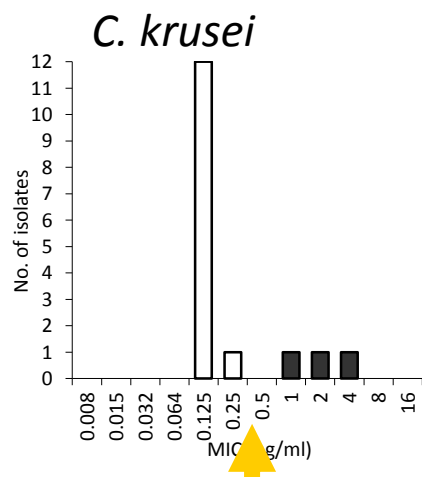


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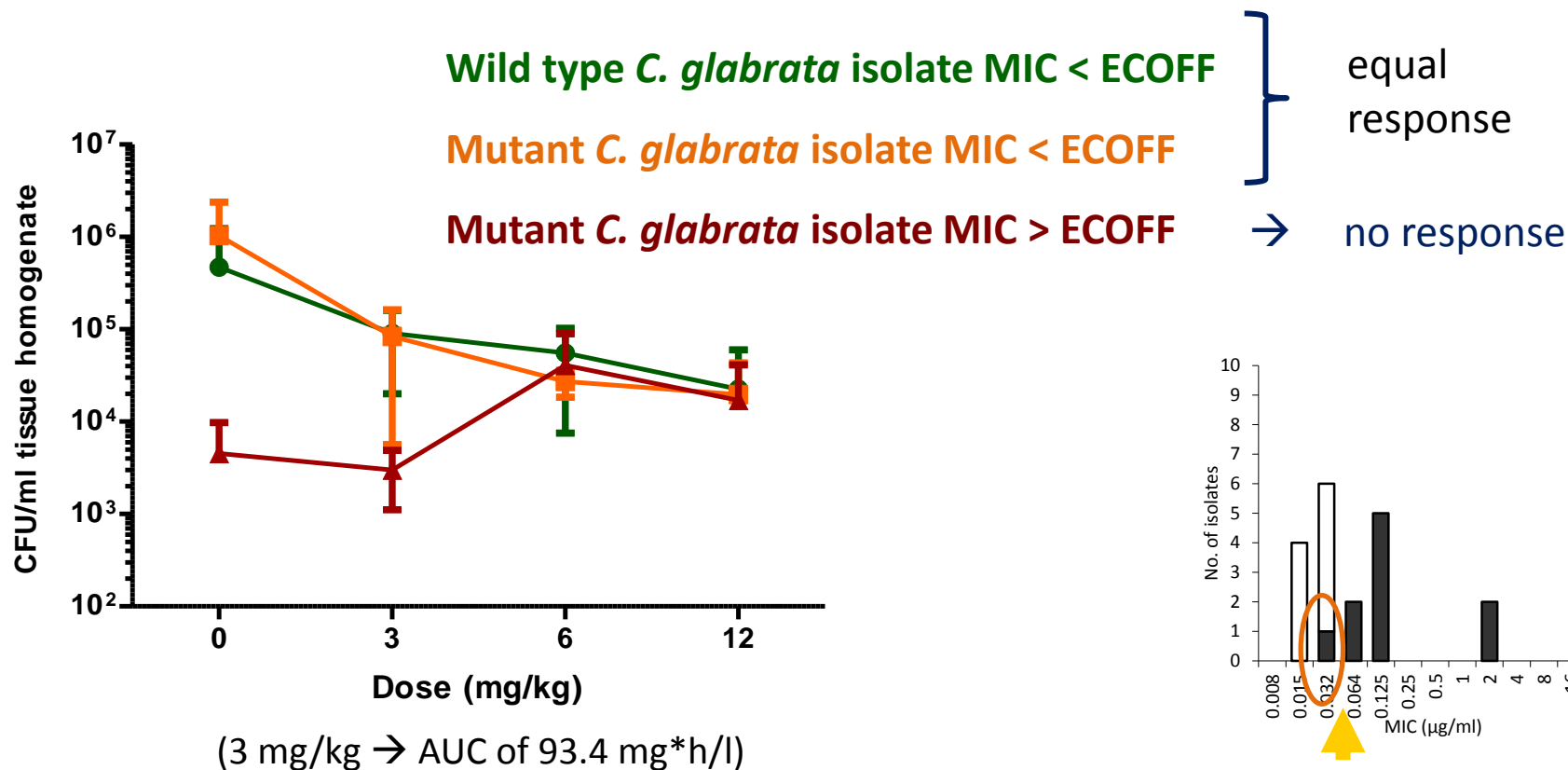


▲ Indicates the ECOFF

*fks* mutant  
classified as S!!!



# Animal model



## Step 2: MIC - outcome data

- No such clinical data using EUCAST MICs
  - Micafungin equal to Amphotericin
    - no difference between the species
  - Micafungin equal to Caspofungin
    - no difference between the species
  - Breakthrough cases
    - *C. parapsilosis* over-represented
- All good targets
- however
- C. parapsilosis* associated w breakthroughs

# Step 3: PK/PD & Monte Carlo

## ■ Animal studies and EUCAST MICs

MIC (mg/l)	Population above the target	
	<i>C. albicans</i> (AUC/MIC 2420)	<i>C. glabrata</i> (AUC/MIC 1283)
0.016	89.2 %	96.3%
0.03	51.0%	88.0%
0.06	2.5%	45.0%
0.125	0%	1.1%

← ECOFF

← ECOFF

## ■ Clinical studies and CLSI MICs

- non-*C. parapsilosis* cohort (AUC/MIC ratio > 5000),
  - Target attained for MIC values ≤ 0.016 mg/l
- *C. parapsilosis* (AUC/MIC ratio > 285)
  - Target attained for MIC values ≤ 0.25 mg/l (below the wild type)

# Micafungin EUCAST breakpoints

## 8. Clinical breakpoints

Non-species-related breakpoints	There is insufficient evidence to set non-species-related breakpoints.
Species-related breakpoints	<p>Breakpoints are based on PK data, laboratory animal PK/PD data, microbiology</p> <p><i>C. albicans</i>, S <math>\leq</math> 0.016 mg/L, R &gt; 0.016 mg/L  <i>C. glabrata</i>, S <math>\leq</math> 0.03 mg/L, R &gt; 0.03 mg/L  <i>C. parapsilosis</i>, S <math>\leq</math> 0.002 mg/L, R &gt; 2 mg/L *(see comment below)</p> <p>*<i>C. parapsilosis</i> harbours an intrinsic alteration in the target gene and the MICs species. Clinical trials suggest that the echinocandins can be used to treat invasive <i>parapsilosis</i> breakthrough cases have been significantly linked to micafungin (a classify the wild-type population as "I" and indicate that the organism is intrinsic if micafungin is used.</p>
Species without breakpoints	<p>MICs for <i>C. tropicalis</i> are 1-2 dilution steps higher than for <i>C. albicans</i> and <i>C. g</i> slightly lower for <i>C. tropicalis</i> than for <i>C. albicans</i> at both dosages (100 and 150 whether it translates into a relevant clinical difference is unknown. Hence there</p> <p>MICs for <i>C. krusei</i> are approximately three dilution steps higher than those for ( approximately eight twofold dilutions higher. In addition, only a small number of</p>

# EUCAST *Aspergillus* BPs 2012

BPs indicated as S  $\leq$  x / R > y

AF compound	<i>Aspergillus</i>				
	<i>flavus</i>	<i>fumigatus</i>	<i>nidulans</i>	<i>niger</i>	<i>terreus</i>
Amphotericin	IE*	1/2	Note	1/2	Poor Target
Itraconazole	1/2	1/2	1/2	IE*	1/2
Posaconazole	IE*	0.125/0.25**	IE*	IE*	Note
Voriconazole	IE*	1/2	Note	IE*	IE*

\* MICs are higher than for *A. fumigatus*

\*\* provided sufficient levels can be achieved

Note: the MICs are similar to *A. fumigatus* but insufficient clinical data for BP setting

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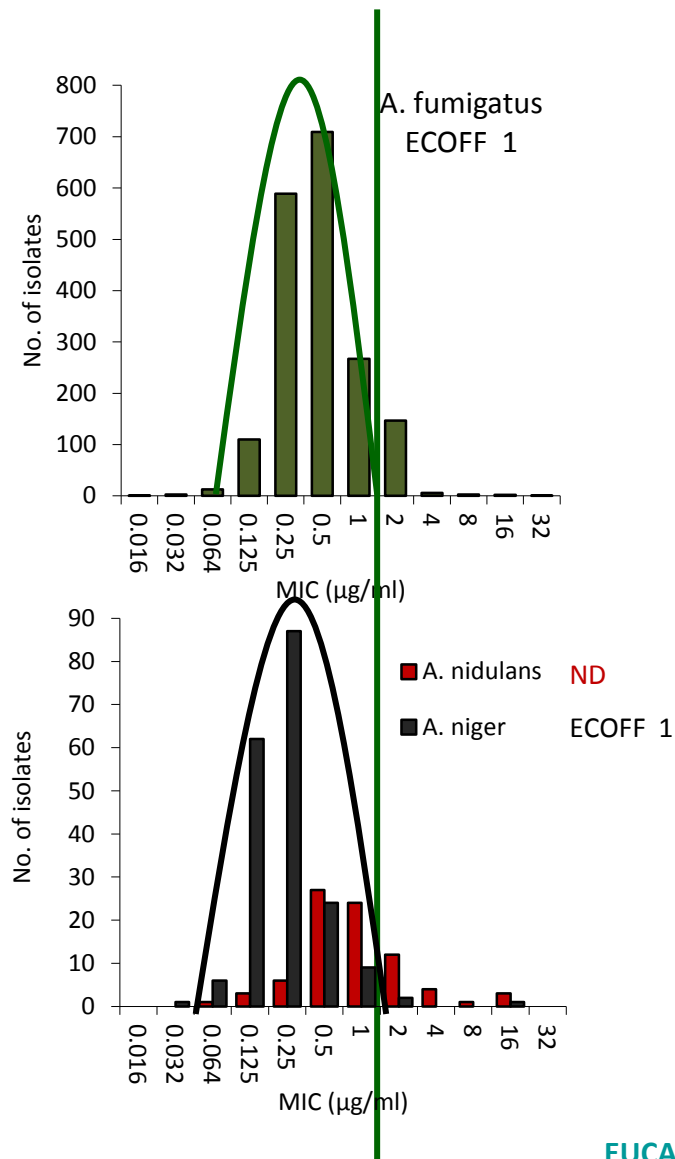
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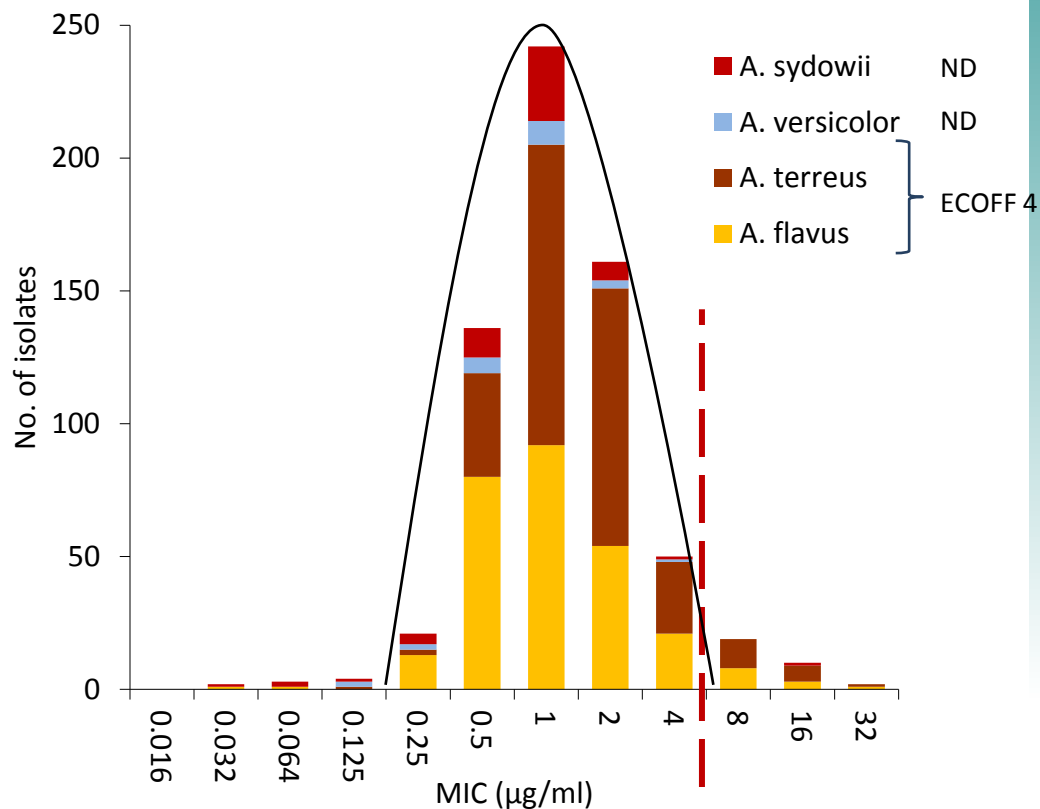
Different from  
the ECOFF



# Aspergillus and Amphotericin B



- Aspergillus good target (except *A. terreus*), but most experience is with *A. fumigatus*.
- No MIC - clinical outcome or animal PK/PD data



# EUCAST *Aspergillus* BPs 2012

BPs indicated as S ≤x / R >y

AF compound	<i>Aspergillus</i>				
	<i>flavus</i>	<i>fumigatus</i>	<i>nidulans</i>	<i>niger</i>	<i>terreus</i>
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Posaconazole	IE*	0.125/0.25**	IE*	IE*	Note
Voriconazole	Note	1/2	Note	Note	Note

\* MICs are higher than for *A. fumigatus*

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Note: the MICs are similar to *A. fumigatus* but insufficient clinical data for BP setting

# Posaconazole Overlap WT & non-WT

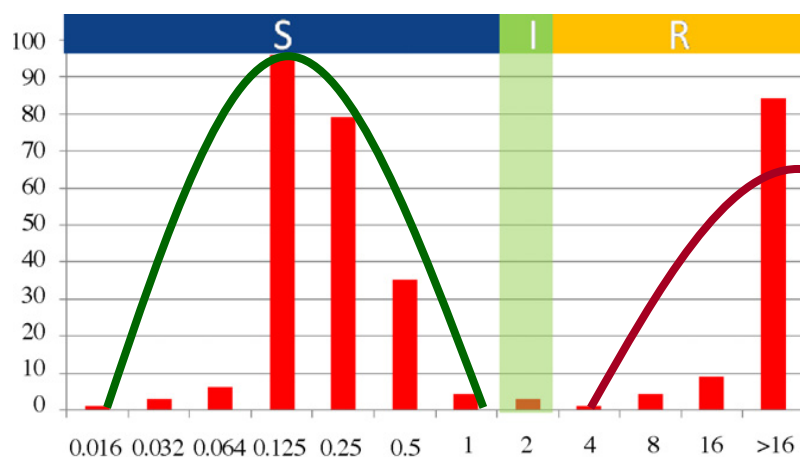
8 Individual dataset for *A. fumigatus*

		MIC (mg/L)													
		0.008	0.015	0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	
EUCAST		7	70	384	205	55	12	3	3	3	4				2095 MICs
EUCAST			5	28	50	11	4		1		1				
EUCAST	3	1	7	5	6	8	3	1			3				
EUCAST			113	116	128	100	46	16	2		5				
EUCAST			6	49	163	57	7	6	2		1	14			
EUCAST		5	12	98	77	48	44	69	18	1	2	1	6		
EUCAST 24h						1	3	15	1						
CLSI				21	1	73	87	1	1						

# Overlap between WT and non-WT

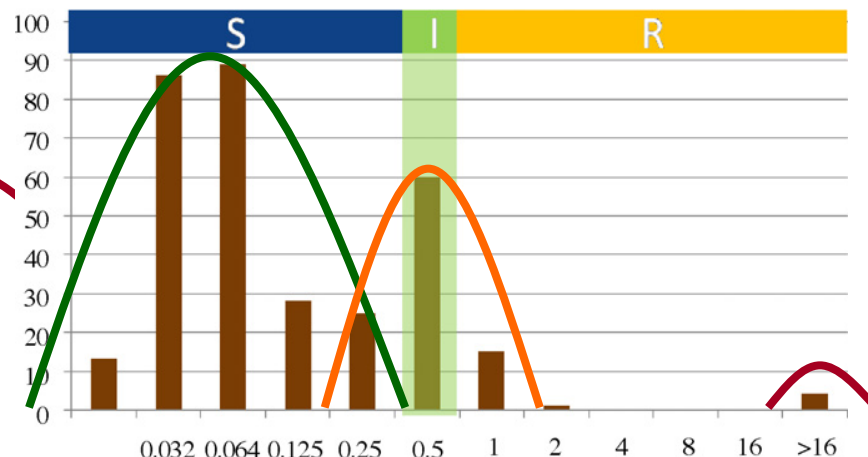
## *A. fumigatus*

### Itraconazole



Clear separation **WT** and **non-WT**

### Posaconazole



Overlap **WT** and **non-WT** → BP need to be < ECOFF to capture all non-WT

# Clinical data & animal PK/PD

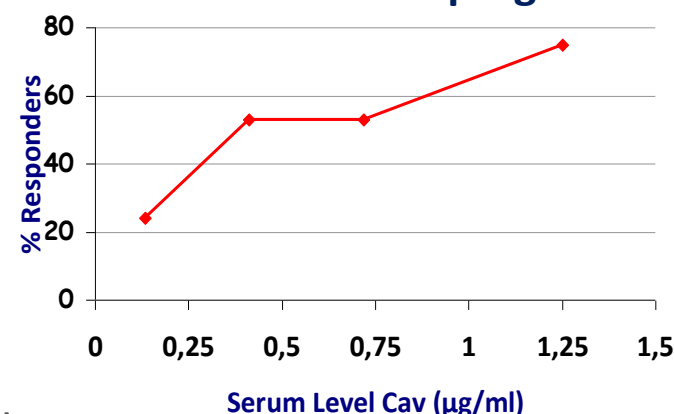
## ■ Borderline exposure with the current formulation

- $C_{av}$  of 1.25 mg/L  $\rightarrow$  better outcome
- $C_{av}$  of 1.25 mg/L  $\sim$  AUC of  $\sim 30$  mg.h/L

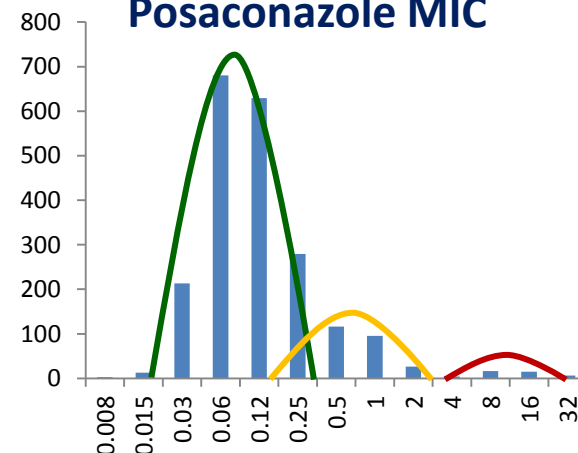
## ■ Mouse models

- AUC:MIC of 167  $\rightarrow$  ED<sub>50</sub> (include 0.125)
  - GM as endpoint, neutropenic model
- AUC:MIC of 498  $\rightarrow$  ED<sub>50</sub> (include 0.06)
  - Survival as endpoint, non-neutropenic model
- Poorer outcome for *cyp51A* mutants
  - some of which may have an MIC of 0.25 mg/L

Posaconazole & aspergillosis



Posaconazole MIC

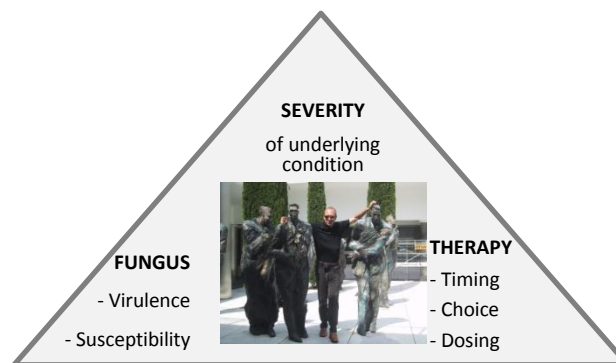


# Conservative Posaconazole BP

- *Aspergillus fumigatus*
  - $S \leq 0.125$ ,  $R > 0.25$  mg/L
    - Provided adequate drug exposure has been confirmed using therapeutic drug monitoring
  - TDM targets:
    - $> 1$  mg/L at steady state for salvage treatment
    - $> 0.7$  mg/L for prophylaxis
- App. 15% WT may be classified as non-S
- Itraconazole “S” → probably also posaconazole “S”

# ECOFFs

- Allow the clinician to differentiate between
- Normal
  - I can rely on my clinical experience for this infection
- Different
  - I should be aware this case may respond different



# May thanks for your attention

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# EUCAST Fluconazole MIC & outcome

Fluconazole 258 cases

(128 candidaemia 58% *C. albicans*, 133 OPC all *C. albicans*)

MIC in mg/l	Candidaemia		OPC $\geq 100$ mg/d		All
	No. cure/Total	% response	No. cure/Total	% response	% response
< 0.5	98/107	92	26/26	100	93
1	6/6	100	4/4	100	100
2	1/1	100	1/1	100	100
4	3/3	100	5/9	56	67
8	2/5	40	7/32	22	24
$\geq 16$	3/4	75	0/60	0	5

S

I/SDD

R

# Step 1 MIC distributions & ECOFFs

## ■ Anidulafungin (■ indicated Mode + 2 dilutions)

2. MIC wild type distributions (based on MIC-values determined with EUCAST and Etest methodology*)																		
Species	≤0.002	0.004	0.008	0.016	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	Tot	ECOFF≤ (mg/L)**
<b>Candida albicans</b> Full distribution	284	<u>360</u>	208	77	16	6	4	1	1							1	958	0.016-0.032
Truncated at 0.032					618	3	0	0	1	1	0	0	0	0	0	0	623	
Truncated at 0.016				233	5	1	0	1	0	0	0	0	0	0	0	0	240	
<b>Candida glabrata</b> Full distribution	55	38	60	<u>149</u>	62	8	6	8	2	2	0	0	2	0	0	0	392	0.032-0.064
Truncated at 0.032					233	1	1	0	2	0	0	0	0	0	0	0	237	
Truncated at 0.016				0	13	49	16	1	0	1	1	0	0	0	0	0	81	
<b>Candida krusei</b> Full distribution	2	1	12	<u>21</u>	7	12	2	3	0	0	0	0	0	0	0	0	60	0.064
Truncated at 0.032					58	7	0	0	0	0	0	0	0	0	0	0	65	
Truncated at 0.016																	0	
<b>Candida parapsilosis</b>	0	3	1	0	4	6	2	36	78	<u>171</u>	96	13	7	0	0	2	419	4
<b>Candida tropicalis</b> Full distribution	18	19	12	<u>30</u>	22	4	4	1	0	0	0	0	0	0	0	0	110	0.032-0.064
Truncated at 0.032					120	2	0	0	0	0	0	0	0	0	0	0	122	
Truncated at 0.016				8	22	11	0	0	0	0	0	0	0	0	0	0	41	
<b>Candida guilliermondii</b>	0	0	0	0	0	1	0	2	5	17	6	1	0	0	1	0	33	ND

## Step 2: MIC – clinical outcome data

### ■ No such data using EUCAST!

### ■ Anidulafungin superior to fluconazole

- *C. albicans* (135 patients) (81% > 62%)  $P < 0.05$
- *C. tropicalis* (22 patients) (93% > 50%)  $P < 0.05$
- *C. glabrata* (38 patients) (56% > 50%)
- *Candida* spp. (7 patients) (75% > 67%)

Good targets  
for  
anidulafungin

### ■ Anidulafungin inferior to fluconazole

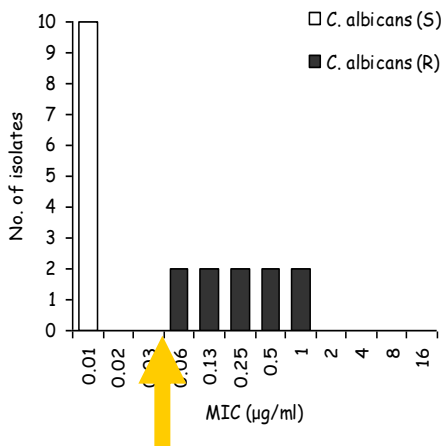
- *C. parapsilosis* (23 patients) (64% > 83%)

Poorer target  
for  
anidulafungin

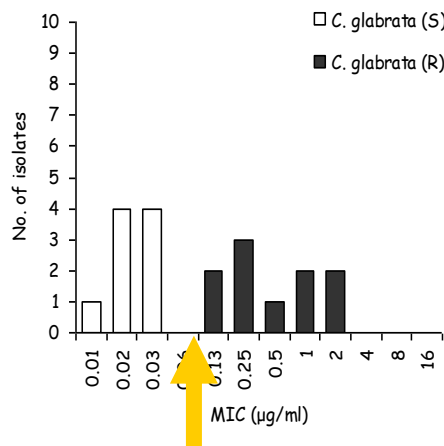
# Step 2: MIC distribution *fks* mutants

ANZI

*C. albicans*



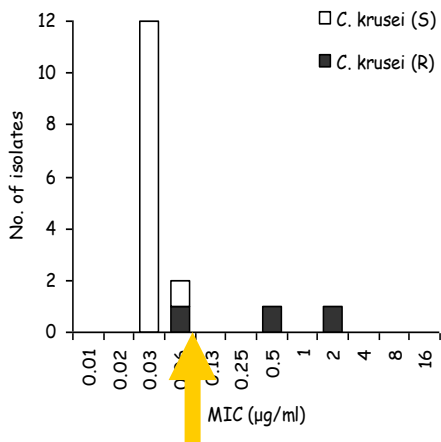
*C. glabrata*



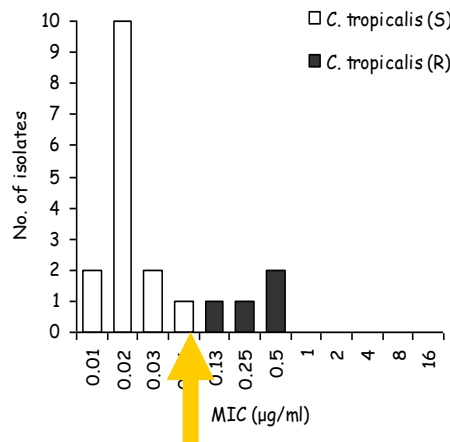
## MICs for *fks* mutants

<i>C. albicans</i>	≥0.06
<i>C. glabrata</i>	≥0.125
<i>C. krusei</i>	≥0.06/ ≥0.5
<i>C. tropicalis</i>	≥ 0.125

*C. krusei*



*C. tropicalis*



# Step 3: No PK/PD → ECOFFs as BP

## Clinical breakpoints based upon ECOFFs

*C. albicans* :  $S \leq 0.03$ ,  $R > 0.03$  mg/l

*C. glabrata* :  $S \leq 0.06$ ,  $R > 0.06$  mg/l

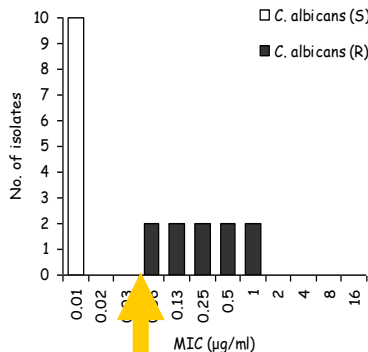
*C. tropicalis* :  $S \leq 0.06$ ,  $R > 0.06$  mg/l

*C. krusei* :  $S \leq 0.06$ ,  $R > 0.06$  mg/l

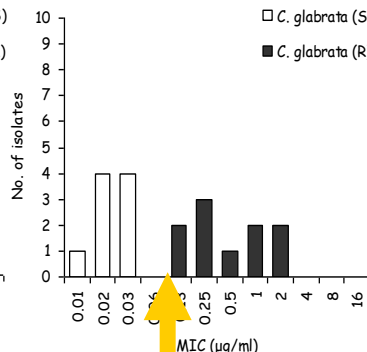
*C. parapsilosis*: Proposed revision:  $S \leq 0.002$ ,  $R > 4$  mg/l

*C. guilliermondii* : No Breakpoints (Higher MICs and insufficient data)

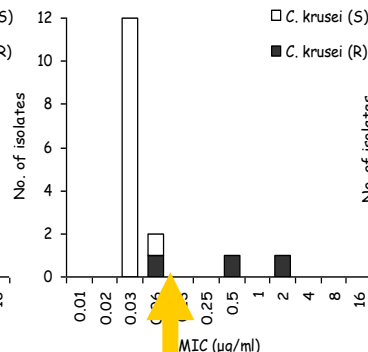
*C. albicans*



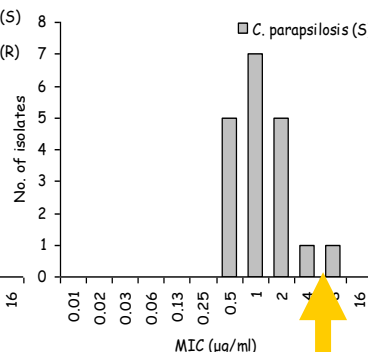
*C. glabrata*



*C. krusei*



*C. parapsilosis*



*C. tropicalis*

