

Synergistic anti-biofilm activity of exebacase and rifampin, vancomycin or daptomycin against *Staphylococcus epidermidis* strains responsible for bone and joint infections

Aubin Souche, Camille Kolenda, Raymond Schuch, Tristan Ferry,
Frédéric Laurent, Jérôme Josse

CIRI – Centre International de Recherche en Infectiologie

Equipe Pathogénie des Staphylocoques

INSERM U1111 – CNRS UMR5308 – ENS Lyon – Université de Lyon

Conflict of interest

1

- This study was founded by ContraFect Corporation
- ContraFect Corporation have no impact on study design, results and interpretation

BJI epidemiology

2

Bone and Joint Infections

M & M

Bactericidal synergies ?

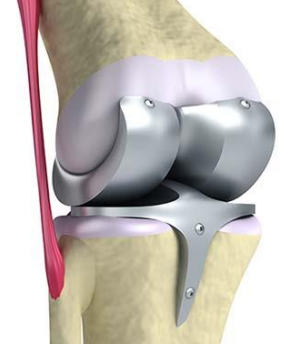
Anti-biomass synergies ?

Conclusion



BJI

- Prevalence 70/100 000 inhbt
- Morbidity 40% - Mortality 5%
- Increasing number



BJI epidemiology

2

Bone and Joint Infections

M & M

Bactericidal synergies ?

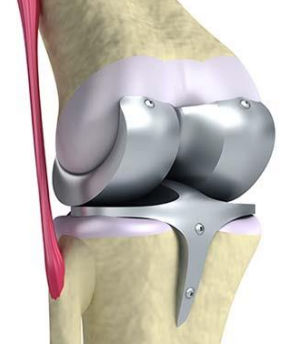
Anti-biomass synergies ?

Conclusion



BJI

- Prevalence 70/100 000 inhbt
- Morbidity 40% - Mortality 5%
- Increasing number



S. aureus
30-70%



S. epidermidis
17-48%

BJI epidemiology

2

Bone and Joint Infections

M & M

Bactericidal synergies ?

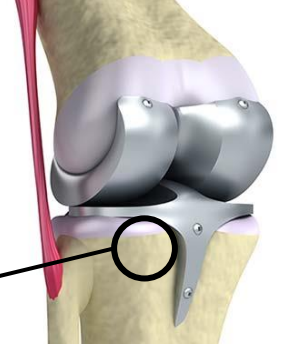
Anti-biomass synergies ?

Conclusion

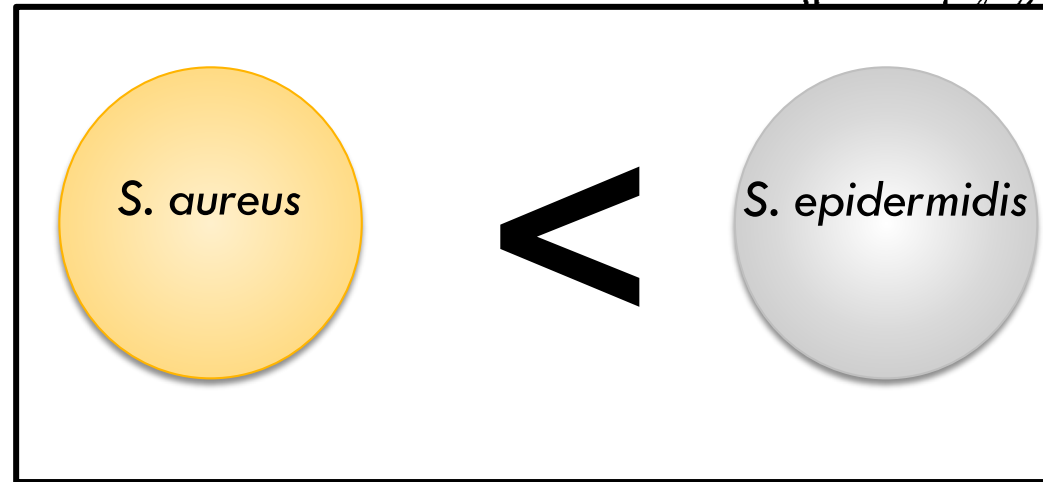


BJI

- Prevalence 70/100 000 inhbt
- Morbidity 40% - Mortality 5%
- Increasing number



30% = PJI



BJI epidemiology

2

Bone and Joint Infections

M & M

Bactericidal synergies ?

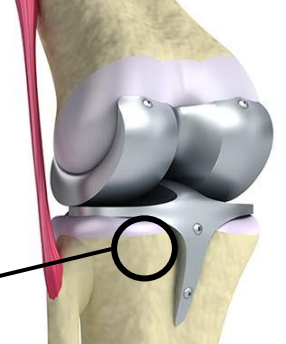
Anti-biomass synergies ?

Conclusion

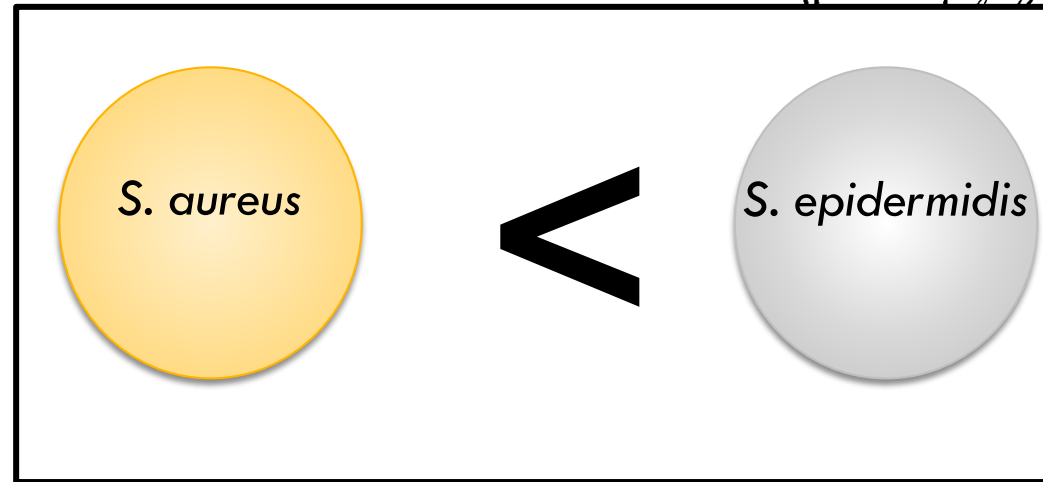


BJI

- Prevalence 70/100 000 inhbt
- Morbidity 40% - Mortality 5%
- Increasing number



30% = PJI



Chronic infection

Chronic BJI/PJI

3

Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion



Over 1 month evolution

Chronic BJI/PJI

3

Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

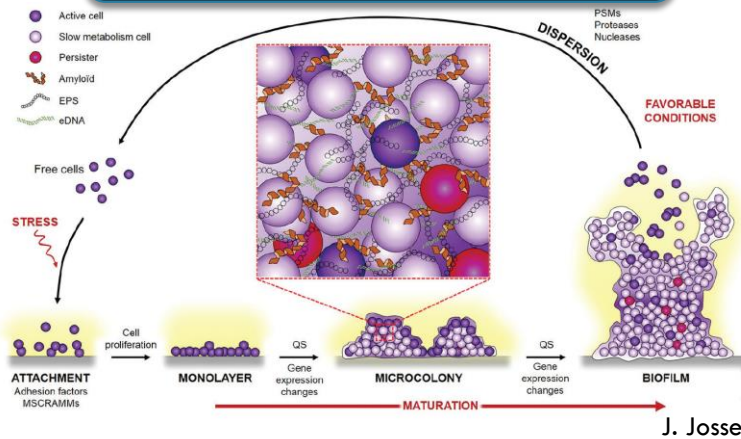


Over 1 month evolution

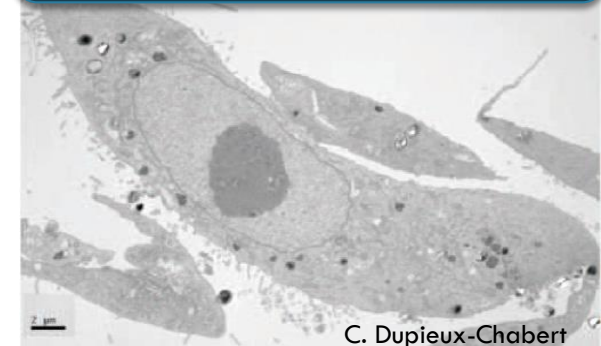
Resistances



Biofilm



Internalization



Chronic *S. epidermidis* PJI

3

Bone and Joint Infections

M & M

Bactericidal synergies ?

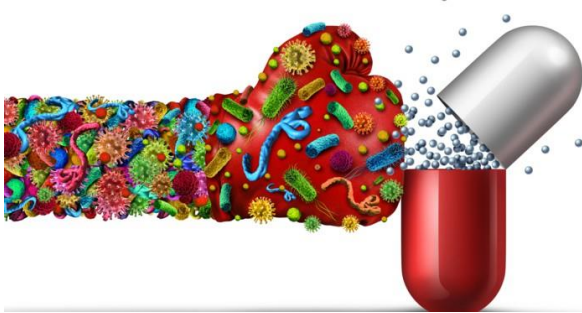
Anti-biomass synergies ?

Conclusion

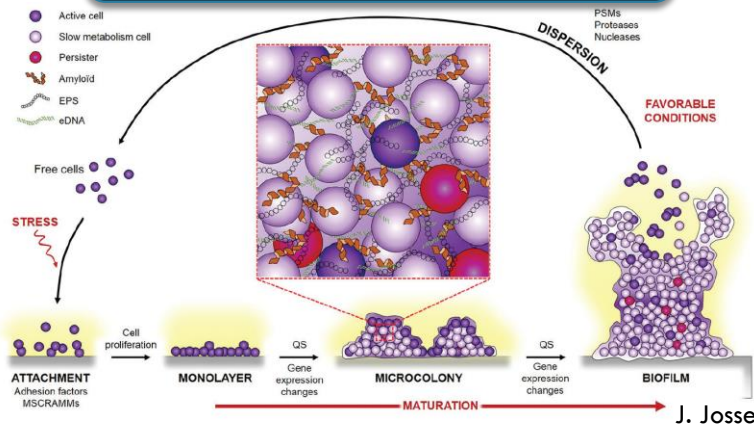


Over 1 month evolution

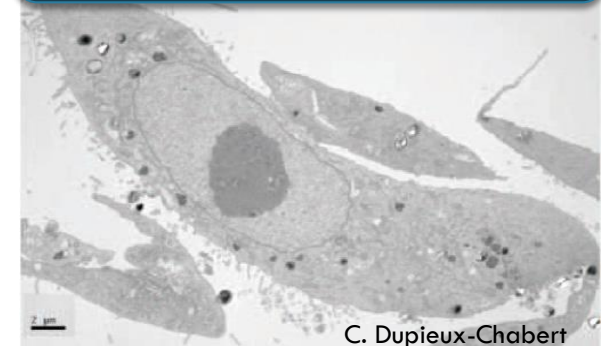
Resistances



Biofilm



Internalization



Difficult to treat infections

Chronic *S. epidermidis* PJI

3

Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

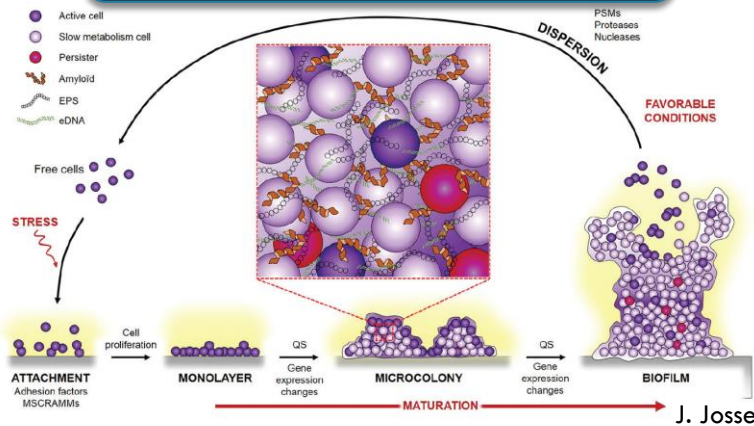


Over 1 month evolution

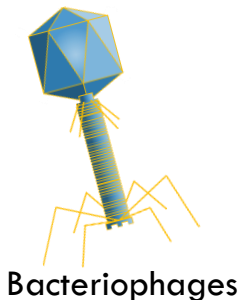
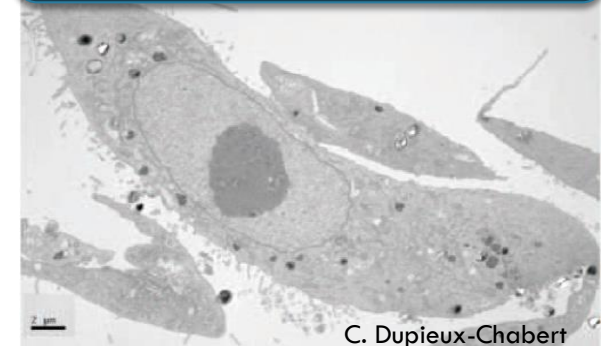
Resistances



Biofilm



Internalization



Bacteriophages

Difficult to treat infections New therapeutics needed



(Endo)lysin

Objectives

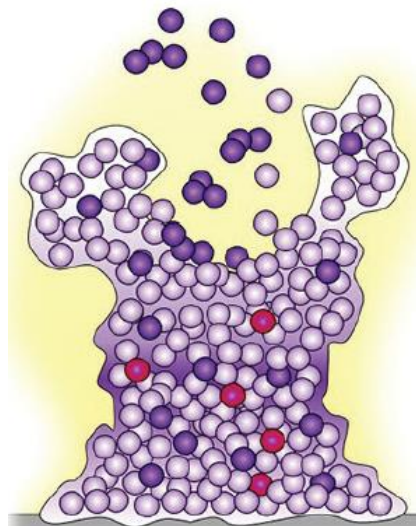
4



Exebacase VS antibiotics
Compared anti-biofilm activity ?



Exebacase + antibiotics Synergistic
anti-biofilm activity ?



Against biofilm

S. epidermidis isolates

5

Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

Antibiotic susceptibility

Strains	Biofilm production	Location	Oxacillin MICs (mg/L)	Rifampin MICs (mg/L)	Vancomycin MICs (mg/L)	Daptomycin MICs (mg/L)	Exebacase MICs (mg/L)
12	None	Knee	S (<0,25)	S (<0,03)	S (1)	S (0,38)	0,125
22	None	Knee	S (<0,25)	S (<0,03)	S (1)	S (0,38)	1
4	Low	Knee	R (>2)	S (<0,03)	S (1)	S (0,5)	1
10	Low	Hip	R (>2)	S (<0,03)	S (1,5)	S (0,25)	0,25
19	Low	Hip	S (<0,25)	S (<0,03)	S (2)	S (0,5)	1
24	Low	Knee	R (>2)	S (<0,03)	S (1)	S (0,5)	1
33	Low	Shoulder	R (>2)	S (<0,03)	S (1)	S (0,5)	0,125
34	Low	Knee	R (>2)	R (>2)	S (1)	S (0,5)	0,25
3	Moderate	Knee	R (>2)	S (<0,03)	S (1)	S (0,75)	2
7	Moderate	Knee	R (>2)	S (<0,03)	S (2)	S (0,19)	1
11	Moderate	Knee	S (<0,25)	R (>2)	S (2)	S (0,5)	0,125
13	Moderate	Knee	R (>2)	R (>2)	S (1)	S (0,5)	0,125
20	Moderate	Knee	R (>2)	R (>2)	S (1)	S (0,5)	0,25
25	Moderate	Hip	R (>2)	S (<0,03)	S (2)	S (0,5)	1
32	Strong	Knee	S (<0,25)	S (<0,03)	S (1)	S (0,38)	0,5
39	Strong	Shoulder	S (<0,25)	S (<0,03)	S (2)	S (0,38)	4
41	Strong	Knee	S (<0,25)	S (<0,03)	S (2)	S (0,5)	0,125
51	Strong	Hip	S (<0,25)	S (<0,03)	S (2)	S (0,125)	1
52	Strong	Knee	R (>2)	R (>2)	S (1)	S (0,064)	2

Exebacase MICs

- 0,125 - 4 mg/L
- Susceptible

S Susceptible, R Resistant. Biofilm production evaluated according to Stepanović *et al.* 2000.

Exebacase and biofilm: synergies with antibiotics ?

6

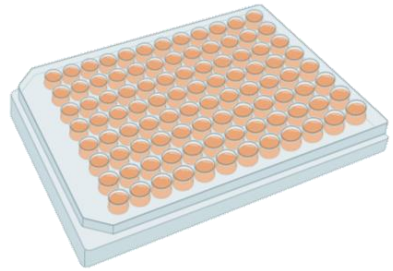
Bone and Joint Infections

M & M

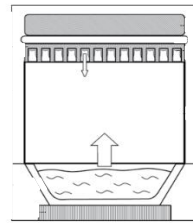
Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion



Biofilm formation
37°C – 24h



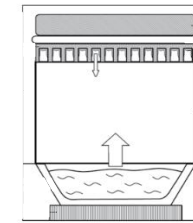
Steam wash
BiofilmCare®
(Tasse et al. 2018)
40 minutes

Treatment

Exebacase → 24h

Antibiotics → 24h

Exebacase + Antibiotics → 24h



Steam wash
BiofilmCare®
(Tasse et al. 2018)
40 minutes



Exebacase

150, 50 or 5 mg/L

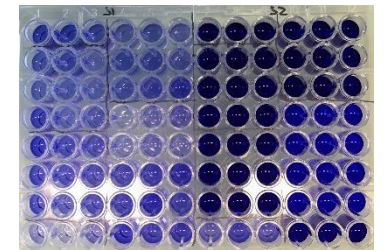


Antibiotics

Rifampin : 1 mg/L / Vancomycin : 10 mg/L / Daptomycin : 10 mg/L



Enumeration
Serial dilutions-plating
Bactericidal evaluation



Crystal violet
Biomass evaluation

Exebacase and biofilm: synergies with antibiotics ?

6

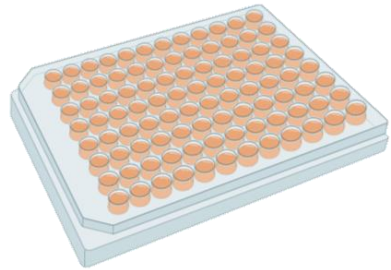
Bone and Joint Infections

M & M

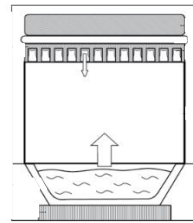
Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion



Biofilm formation
37°C – 24h



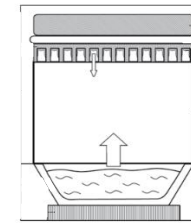
Steam wash
BiofilmCare®
(Tasse et al. 2018)
40 minutes

Treatment

Exebacase → 24h

Antibiotics → 24h

Exebacase + Antibiotics → 24h



Steam wash
BiofilmCare®
(Tasse et al. 2018)
40 minutes



Exebacase

150, 50 or 5 mg/L

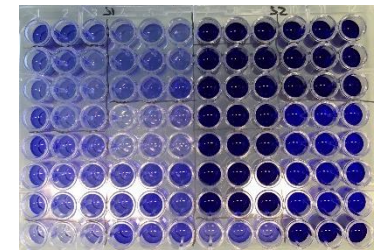


Antibiotics

Rifampin : 1 mg/L / Vancomycin : 10 mg/L / Daptomycin : 10 mg/L



Enumeration
Serial dilutions-plating
Bactericidal evaluation



Crystal violet
Biomass evaluation



Synergy: $\text{Effect AB} > \text{Effect A} + \text{Effect B}$

Bactericidal synergies on pre-formed biofilm ?

7

Bone and Joint Infections

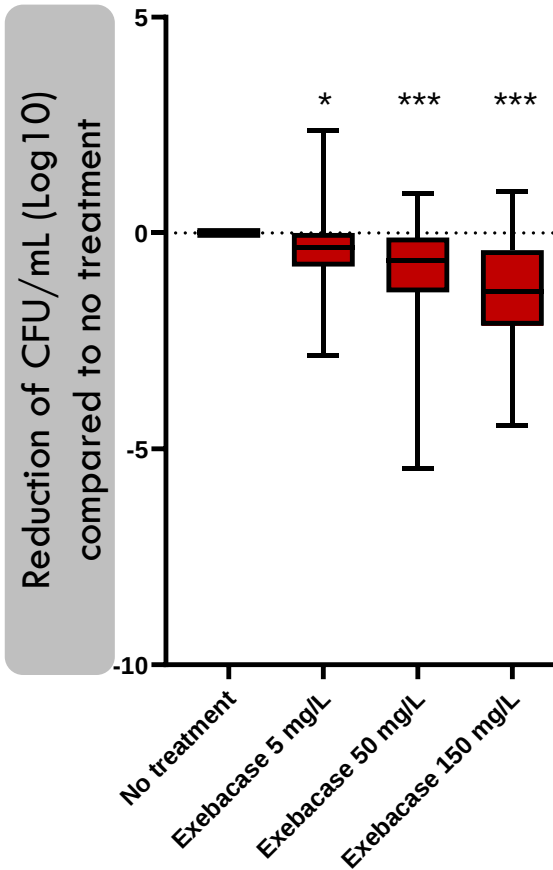
M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

Isolates : n=19
except for rifampin: n=14



Exebacase

- Dose dependant
- Max effect: -1,7 log CFU

Bactericidal synergies on pre-formed biofilm ?

7

Bone and Joint Infections

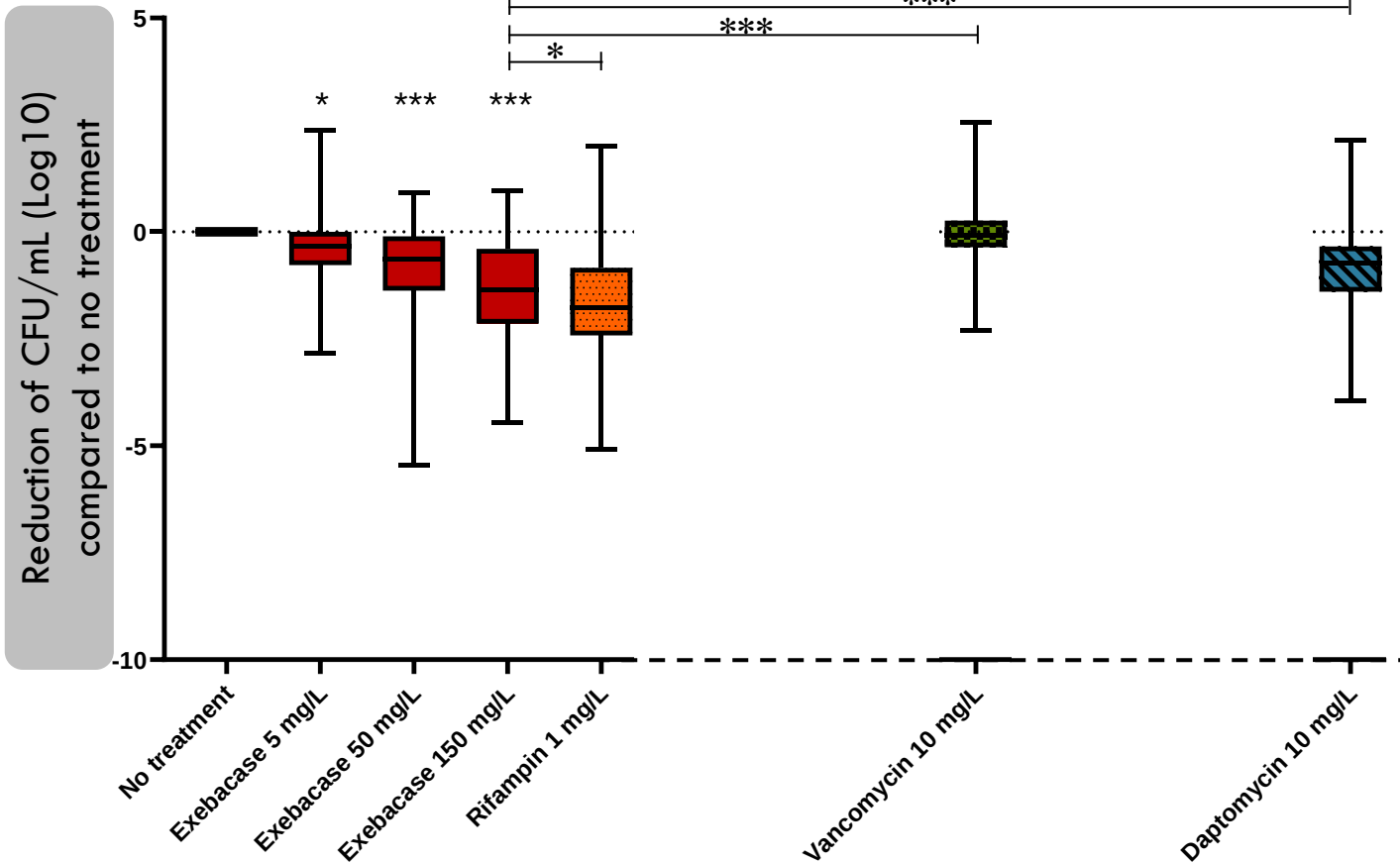
M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

Isolates : n=19
except for rifampin: n=14



Exebacase

- Dose dependant
- Max effect: -1,7 log CFU
- < Rifampin (p=0,021)
- > Vancomycin (p<0,001)
- > Daptomycin (p<0,001)

Bactericidal synergies on pre-formed biofilm ?

7

Bone and Joint Infections

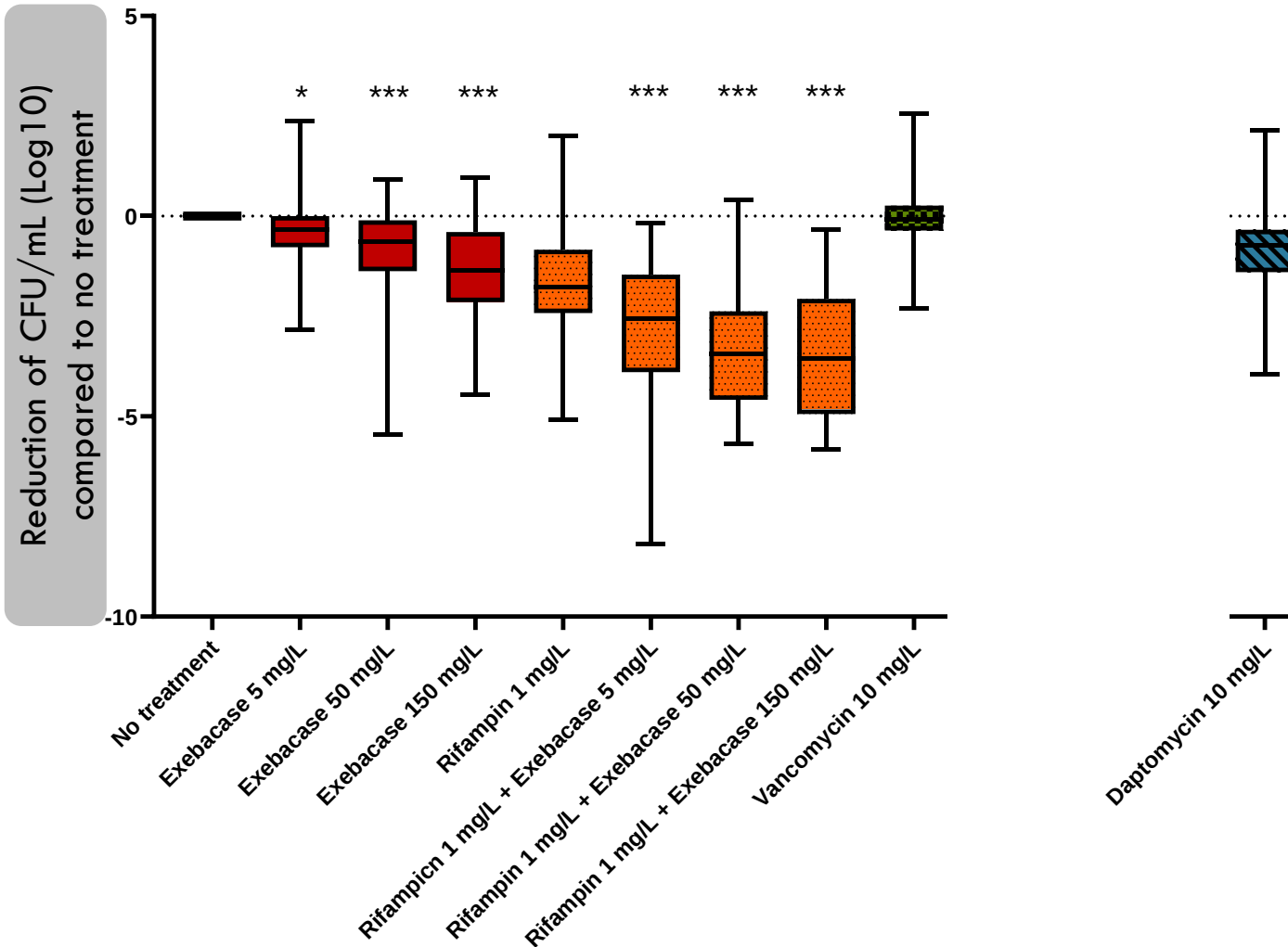
M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

Synergy



Isolates : n=19
except for rifampin: n=14

Synergies with rifampin

■ Max effect: -3,5 log CFU

Bactericidal synergies on pre-formed biofilm ?

7

Bone and Joint Infections

M & M

Bactericidal synergies ?

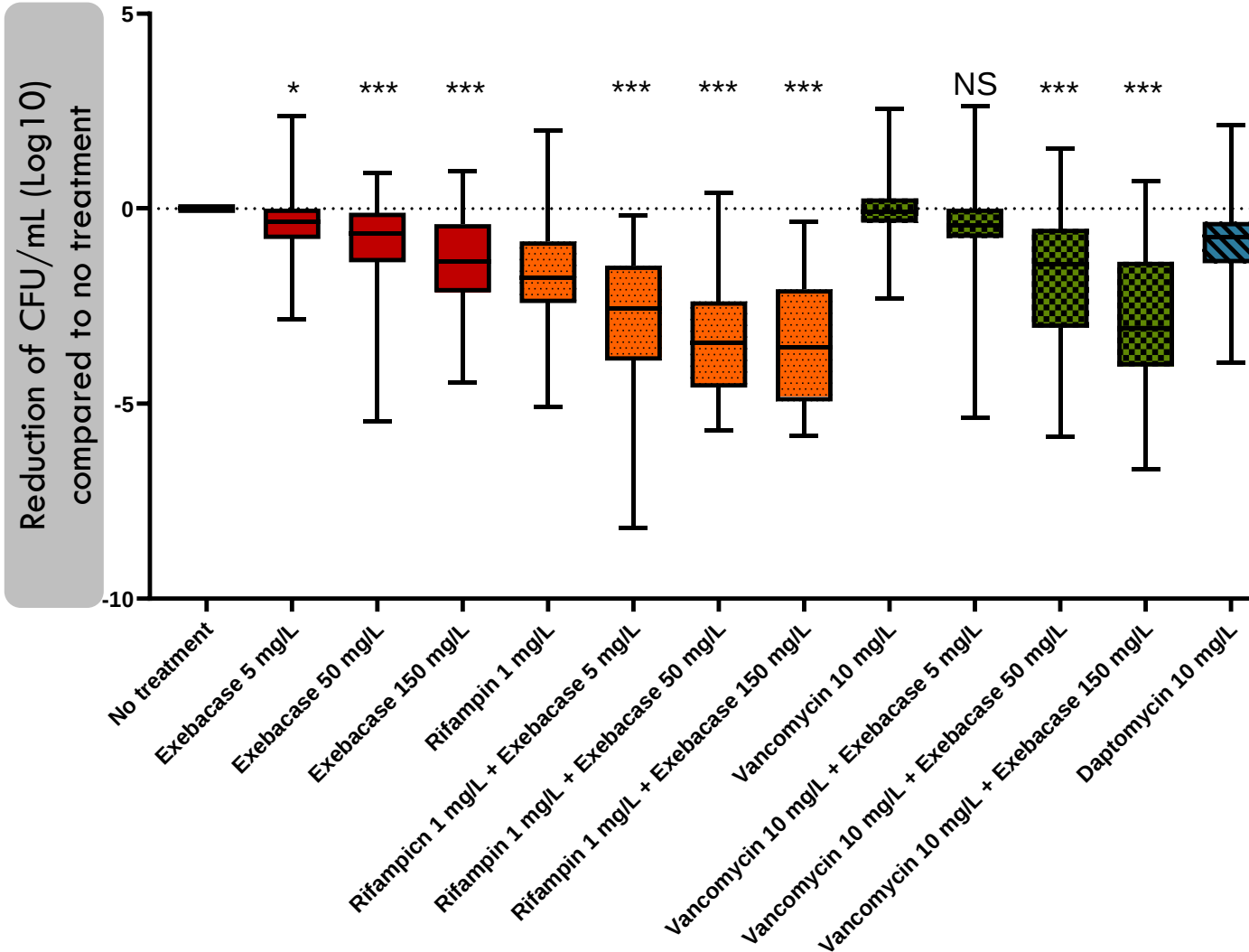
Anti-biomass synergies ?

Conclusion

Synergy

Synergy

Isolates : n=19
except for rifampin: n=14



Synergies with
vancomycin

Max effect: -2,82 log CFU

Bactericidal synergies on pre-formed biofilm ?

7

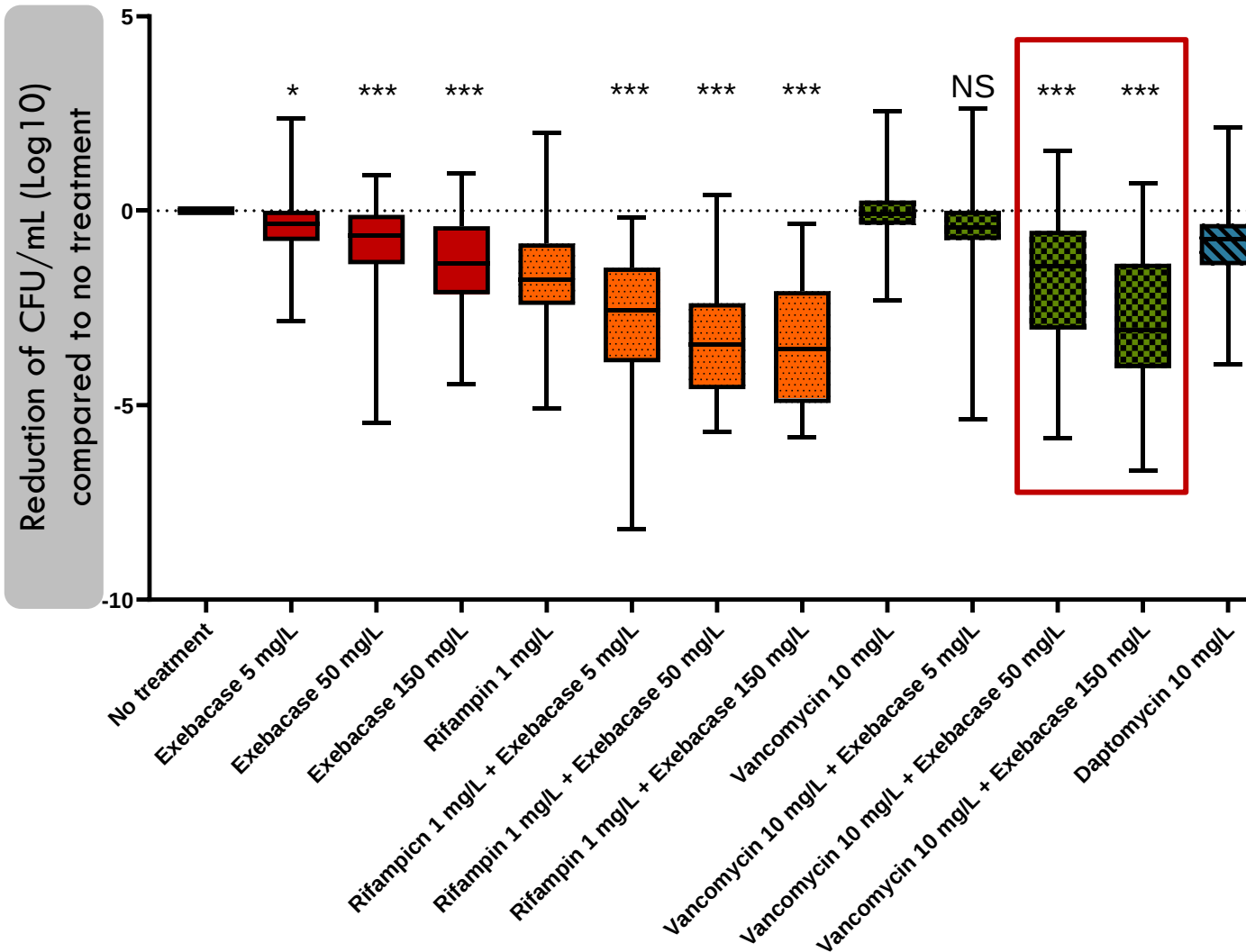
Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion



Isolates : n=19
except for rifampin: n=14

Synergies with
vancomycin

Max effect: -2,82 log CFU

Bactericidal synergies on pre-formed biofilm ?

7

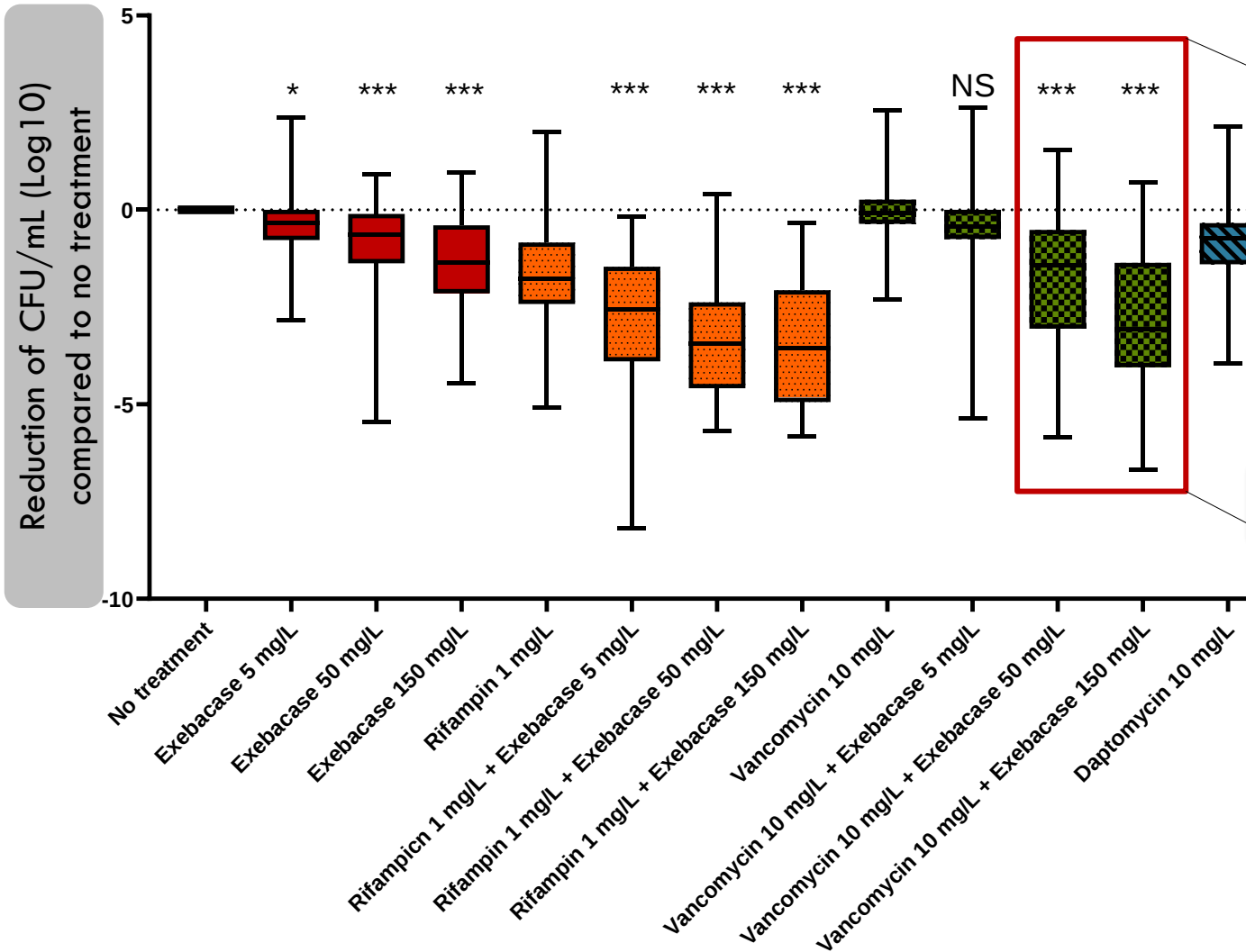
Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

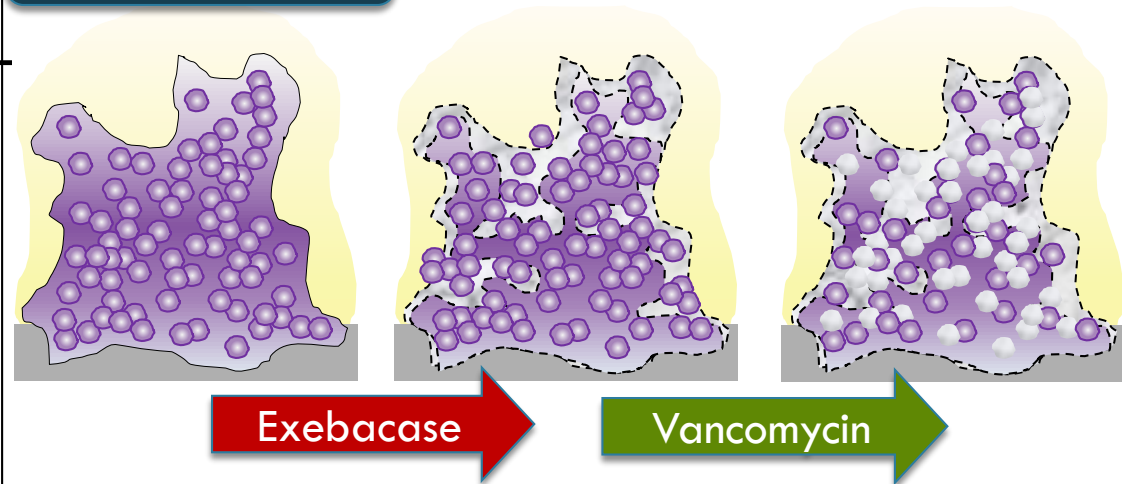


Isolates : n=19
except for rifampin: n=14

Synergies with
vancomycin

■ Max effect: -2,82 log CFU

Hypothesis



Bactericidal synergies on pre-formed biofilm ?

7

Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

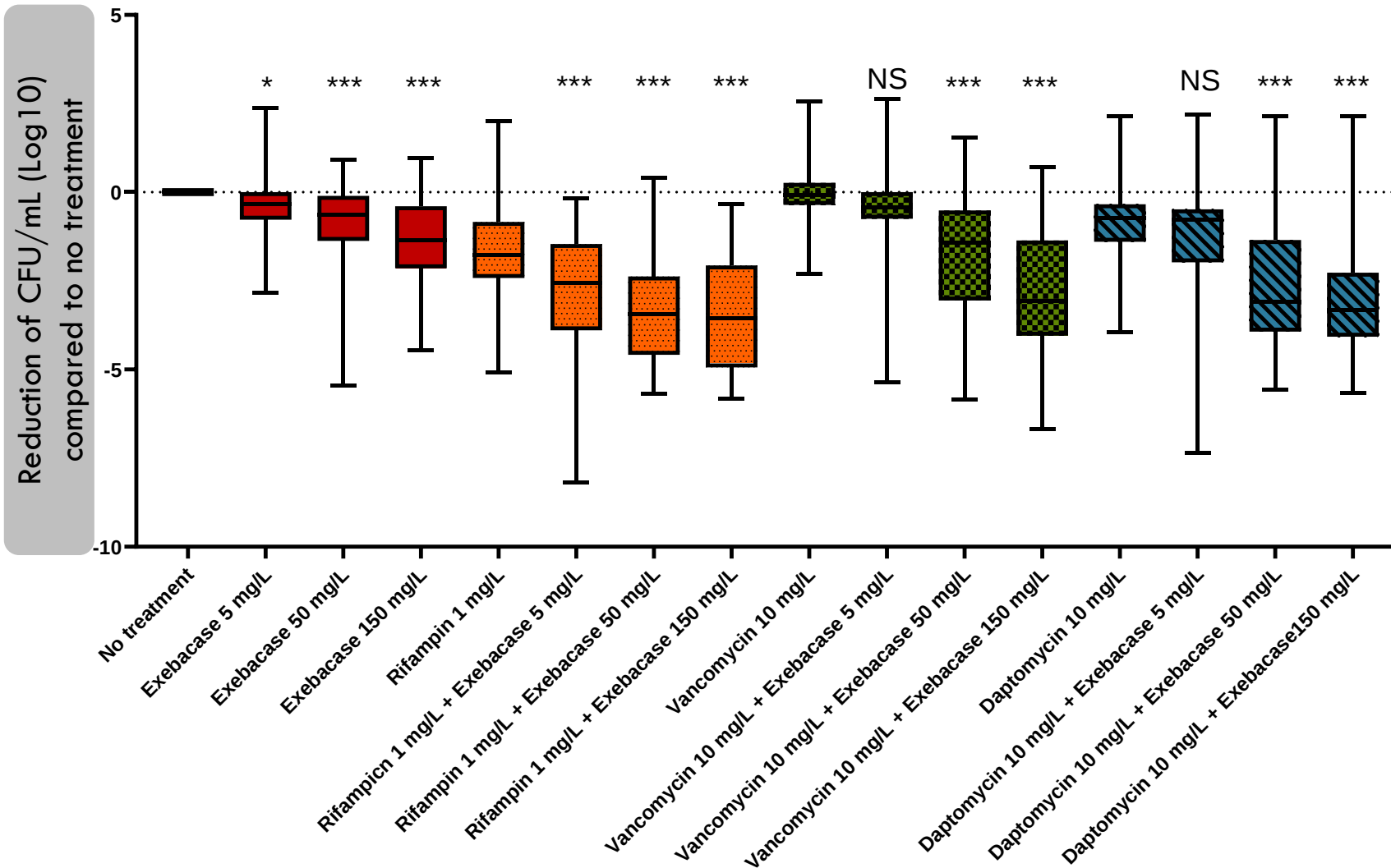
Conclusion

Synergy

Synergy

Synergy

Isolates : n=19
except for rifampin: n=14



Max effect: -3,1 log CFU

Anti-biomass synergies on pre-formed biofilm ?

8

Bone and Joint Infections

M & M

Bactericidal synergies ?

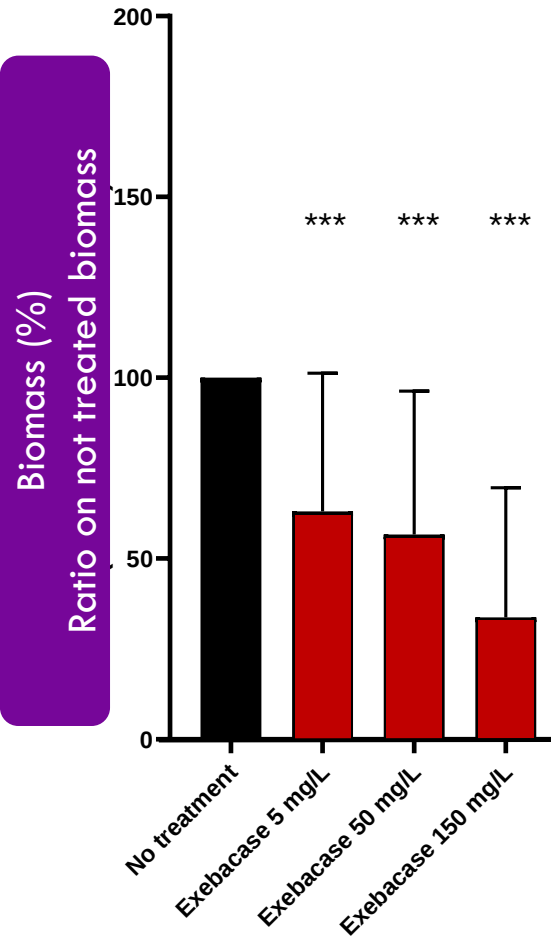
Anti-biomass synergies ?

Conclusion

Isolates : n=19
except for rifampin: n=14

Exebacase

- Dose dependant
- Max effect: -66%



Anti-biomass synergies on pre-formed biofilm ?

8

Bone and Joint Infections

M & M

Bactericidal synergies ?

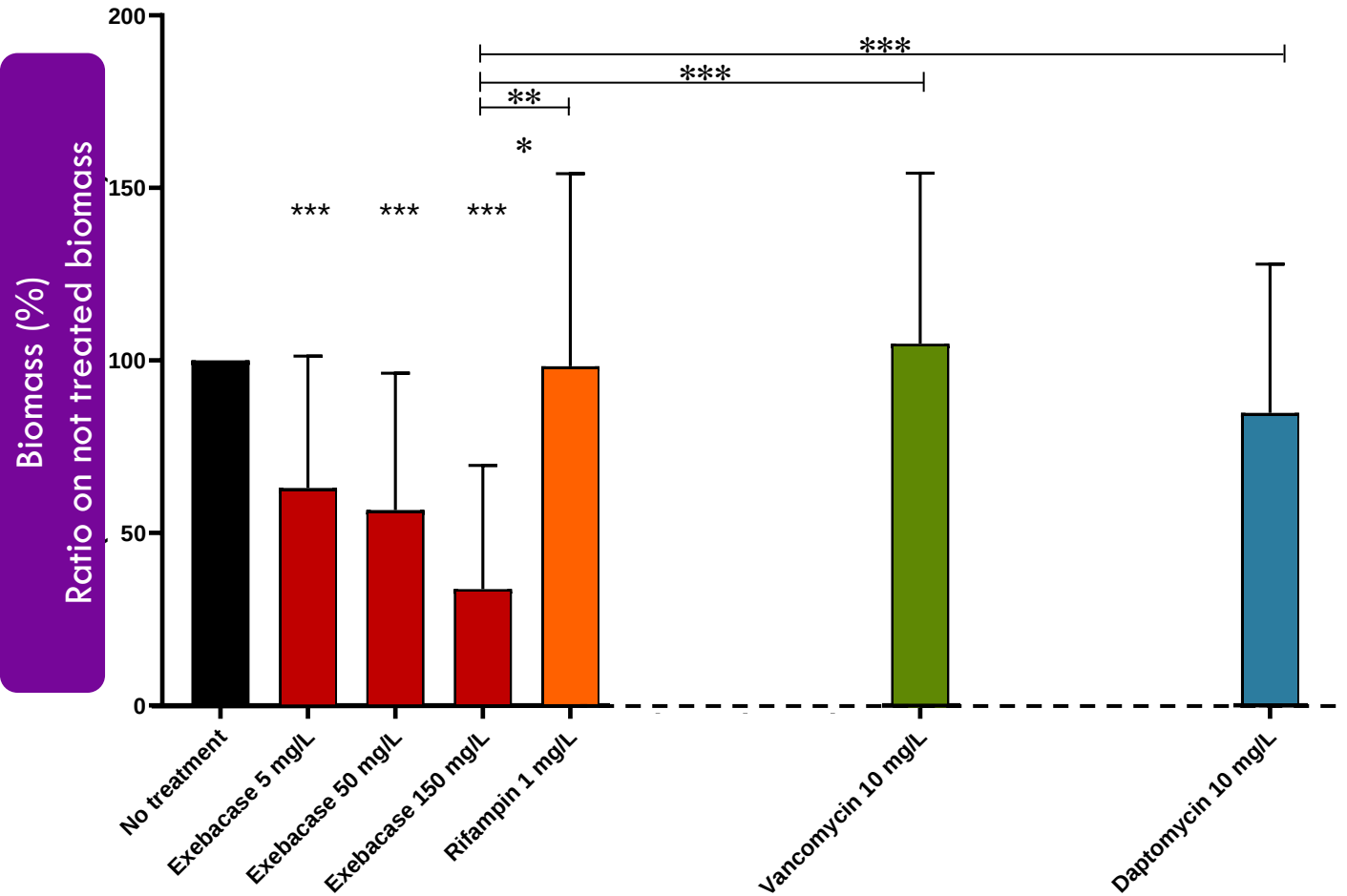
Anti-biomass synergies ?

Conclusion

Isolates : n=19
except for rifampin: n=14

Exebacase

- Dose dependant
- Max effect: -66%
- > rifampin ($p<0,001$)
- > vancomycin ($p<0,001$)
- > daptomycin ($p<0,001$)



Anti-biomass synergies on pre-formed biofilm ?

8

Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

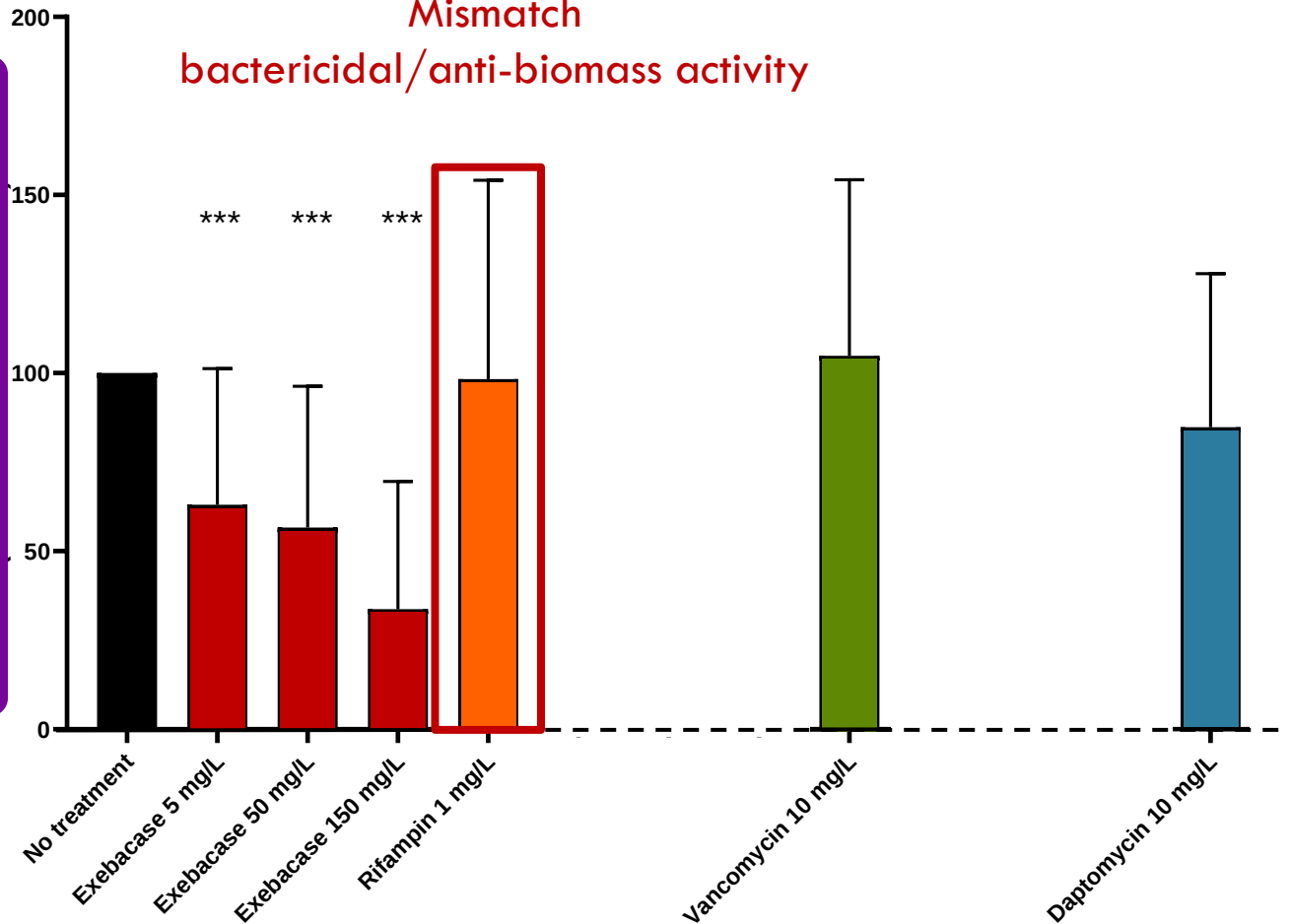
Conclusion

Isolates : n=19
except for rifampin: n=14

Exebacase

- Dose dependant
- Max effect: -66%
- > rifampin ($p<0,001$)
- > vancomycin ($p<0,001$)
- > daptomycin ($p<0,001$)

Mismatch
bactericidal/anti-biomass activity



Anti-biomass synergies on pre-formed biofilm ?

8

Bone and Joint Infections

M & M

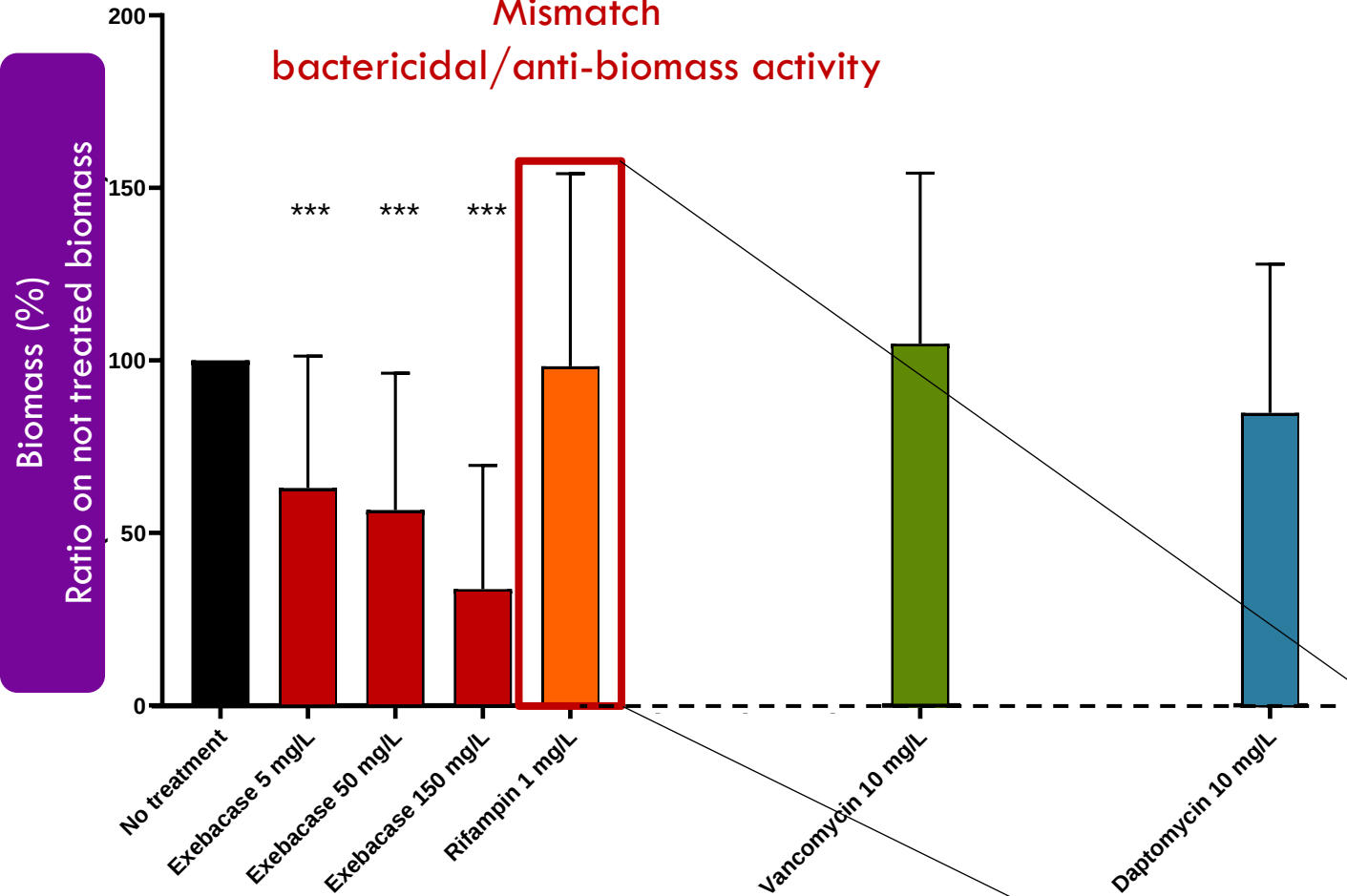
Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

Isolates : n=19
except for rifampin: n=14

Mismatch
bactericidal/anti-biomass activity

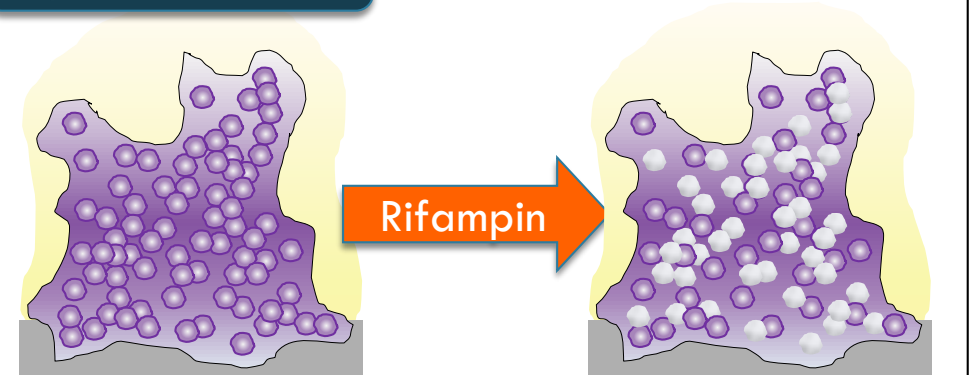


Exebacase

- Dose dependant
- Max effect: -66%
- > rifampin ($p < 0,001$)
- > vancomycin ($p < 0,001$)
- > daptomycin ($p < 0,001$)



Hypothesis



Anti-biomass synergies on pre-formed biofilm ?

8

Bone and Joint Infections

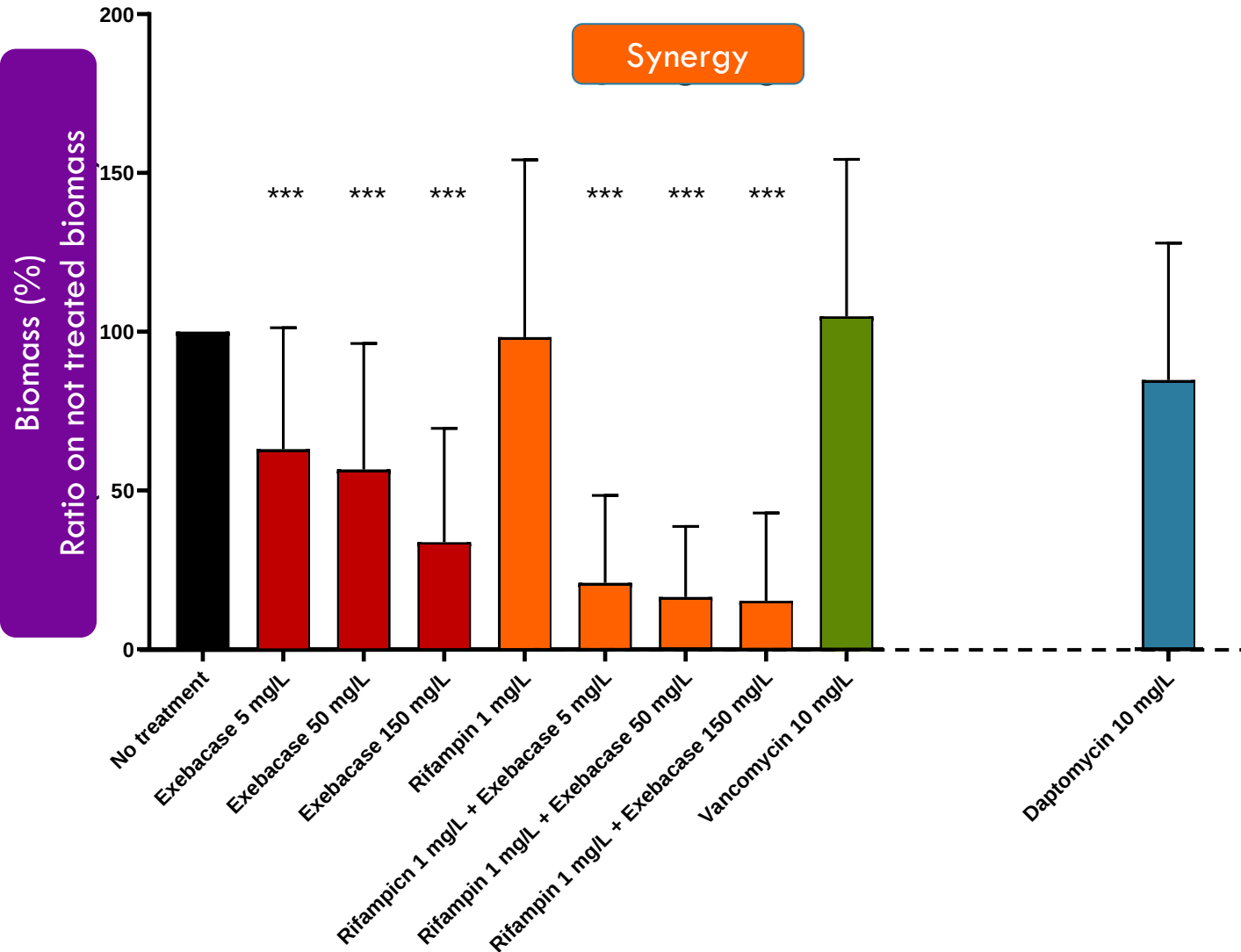
M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

Isolates : n=19
except for rifampin: n=14



Synergies with rifampin

■ Max effect: -84%

Anti-biomass synergies on pre-formed biofilm ?

8

Bone and Joint Infections

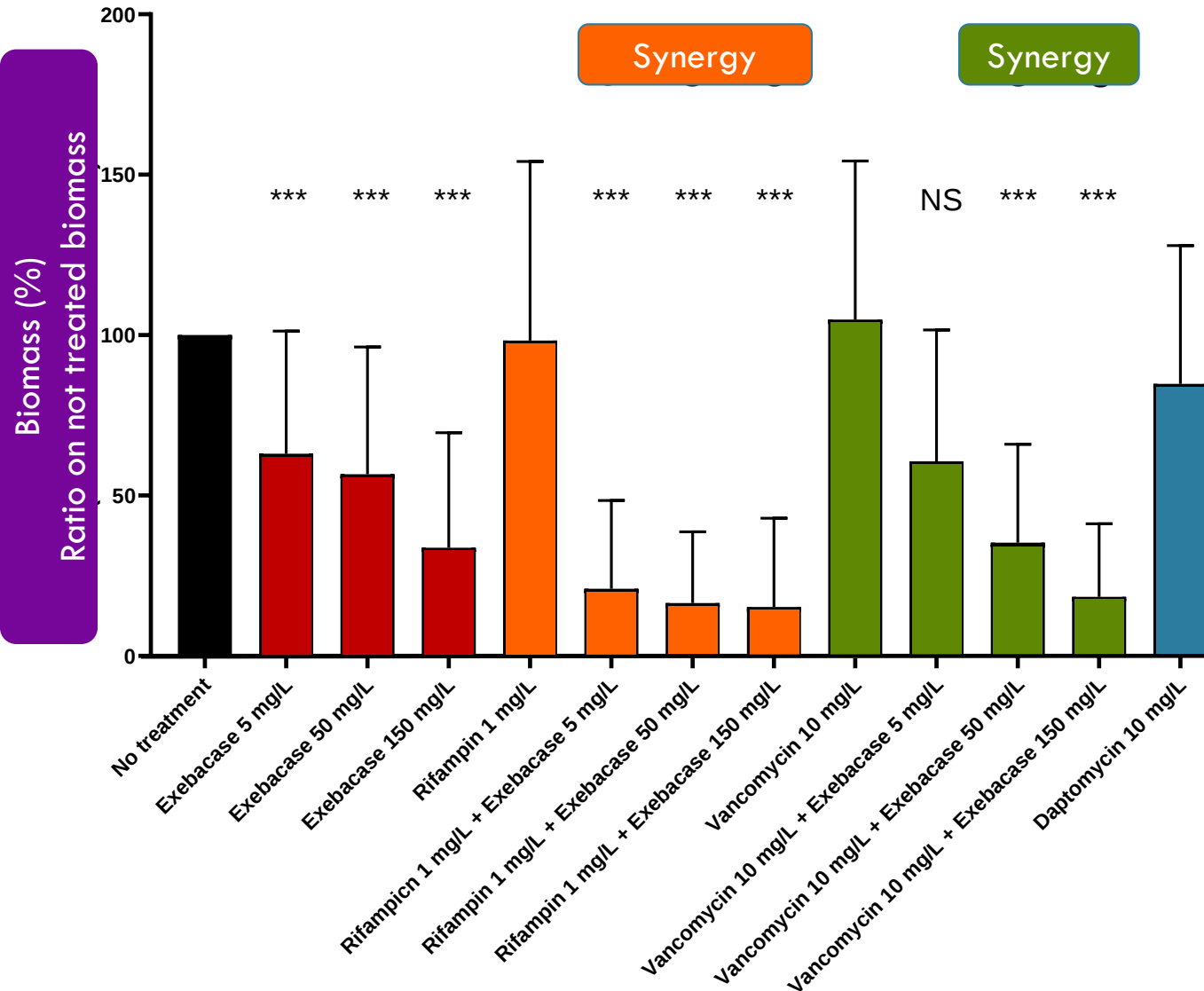
M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion

Isolates : n=19
except for rifampin: n=14

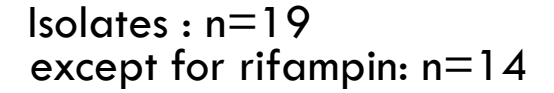


Synergies with
vancomycin

- Dose dependant
- Max effect: -81%

8

Anti-biomass synergies ?



- Dose dependant
- Max effect: -85%

Conclusion

9

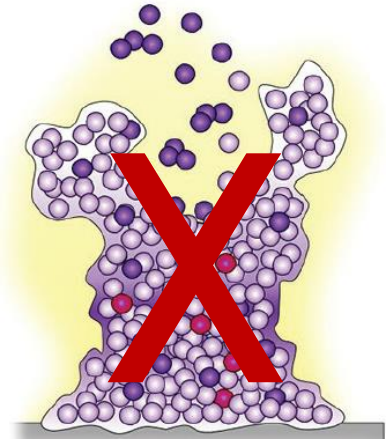
Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion



Anti-biofilm
activity



Synergies
against biofilm



Anti-
planktonics
activity

Conclusion

9

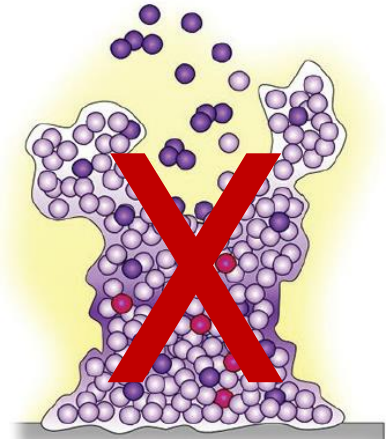
Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion



Anti-biofilm
activity



Synergies
against biofilm

Interest in BJI/PJI



Anti-
planktonics
activity

Conclusion

9

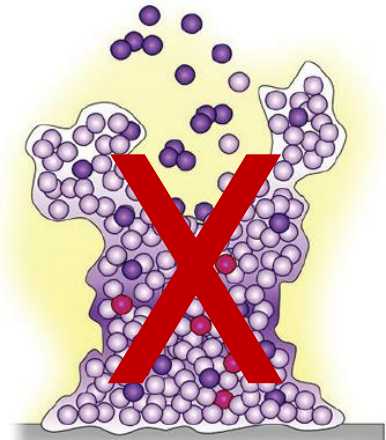
Bone and Joint Infections

M & M

Bactericidal synergies ?

Anti-biomass synergies ?

Conclusion



Anti-biofilm
activity

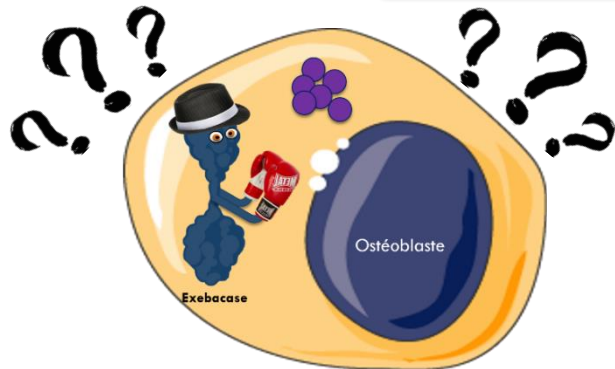


Synergies
against biofilm



Anti-
planktonics
activity

Interest in BJI/PJI



Intracellular ?



Strain dependant
activity ?



Thank you for your attention

Team Staphylococcal
pathogenesis



Contrafect Corporation

