

Study of the effect of pulsed lavage on the antibiotic susceptibility of *S. aureus* biofilms

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Disclosures

- Interpulse devices gifted by Stryker Corp.
- Grants:
 - Fondation Saint-Luc, Belgium
 - Fonds pour la Recherche Scientifique – FNRS, Belgium

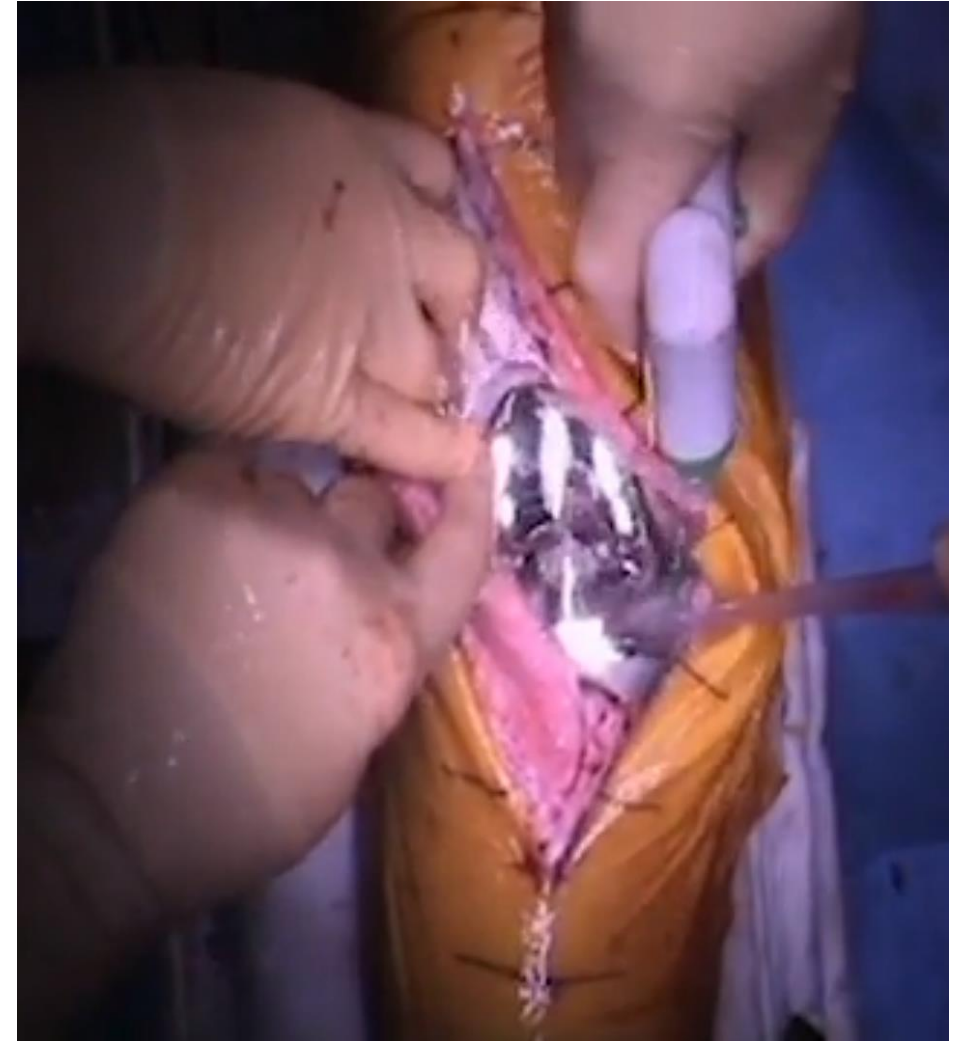


Introduction

- Pulsed lavage is frequently used during debridement surgery
- Expected effect: removal of bacteria
- Few experimental reports of its effects in the literature
- No report of the combined effects of pulsed lavage and clinically relevant antibiotics

Objective

Describe the effect of the combination of pulsed lavage and vancomycin



H. Clarke and M. Spangehl
<https://www.vumedi.com/video/two-stage-debridement-with-antibiotic-beads-for-acute-peri-prosthetic-joint-infection/>

Materials and Methods

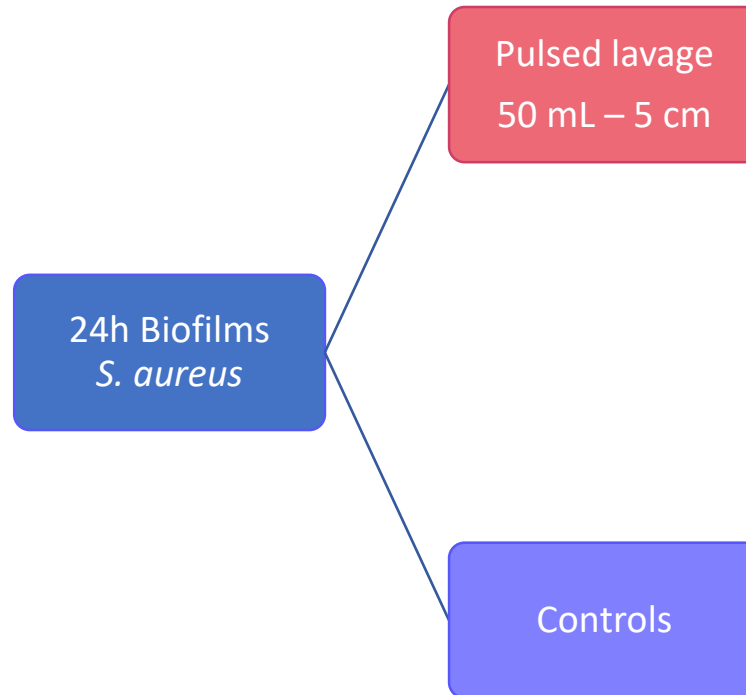
- 3 *S. aureus* strains
 - 1 reference (ATCC 33591)
 - 2 clinical isolates from ODRI
- Ti6Al4V coupons
- $6.5 \log_{10}$ CFU inoculum in TSB supplemented with 1% Glucose and 2% NaCl (TGN)
- 24h at 37°C, constant agitation (50rpm)



24h Biofilms
S. aureus

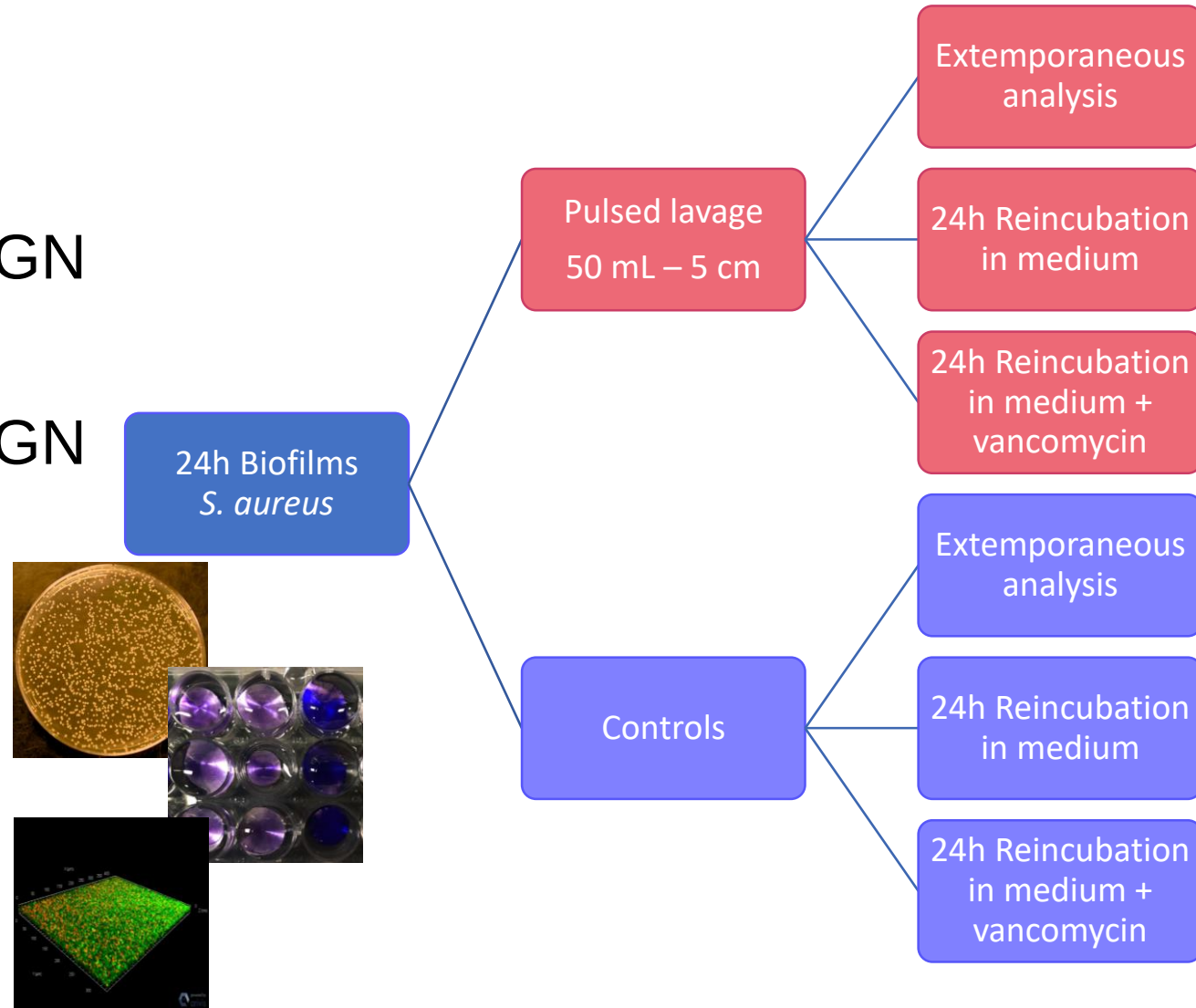
Materials and Methods

- 2 groups:
 - Pulsed lavage
 - Control
- Pulsed lavage performed with fixed volume and distance



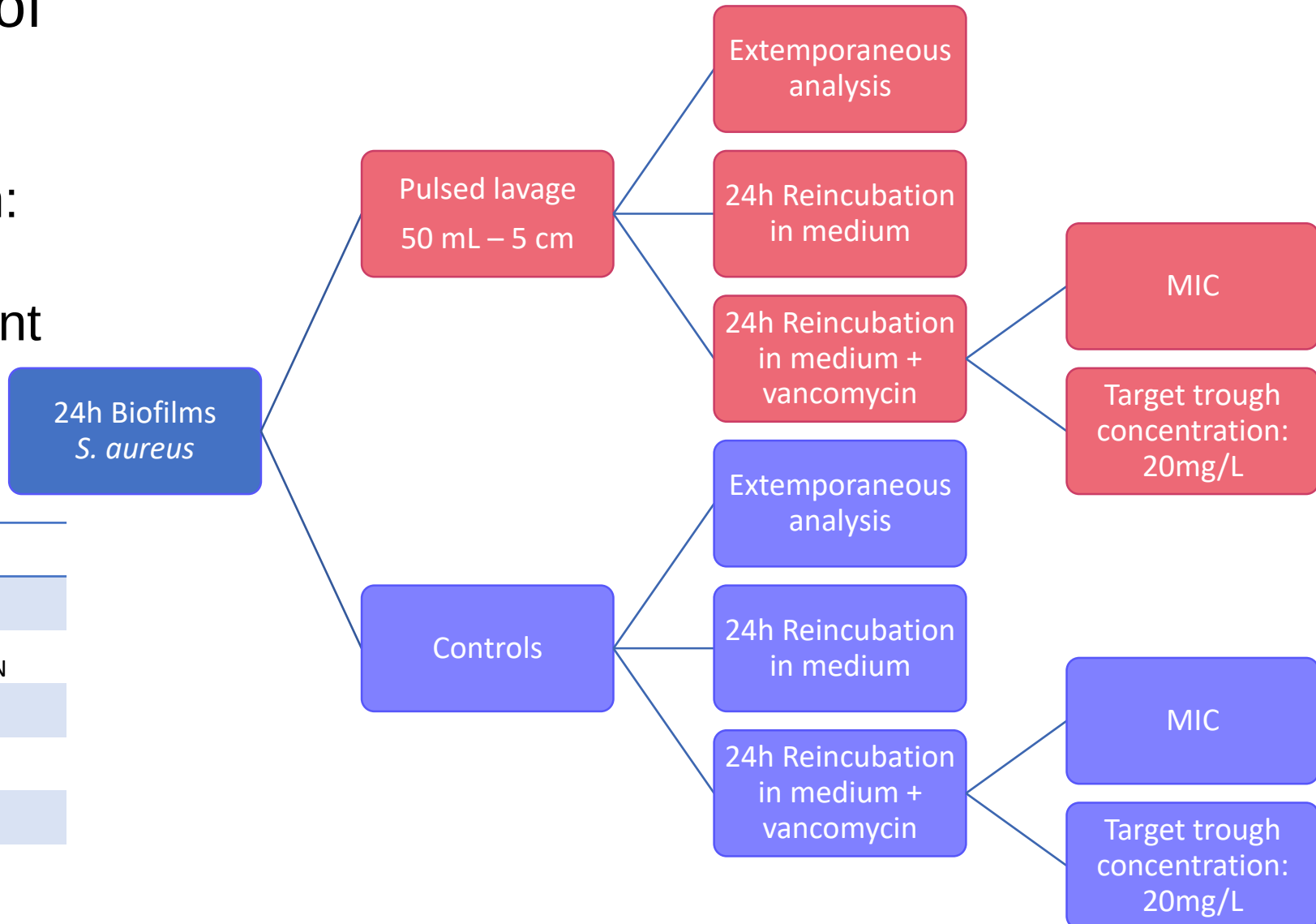
Materials and Methods

- Extemporaneous analysis
- OR
- Reincubation in TGN
- OR
- Reincubation in TGN with vancomycin
- Analysis:
 - CFU counts
 - Biomass assay
 - Fluorescence microscopy



Materials and Methods

- Two concentrations of vancomycin
 - Vancomycin target trough concentration: 20mg/L
 - MIC: strain dependant



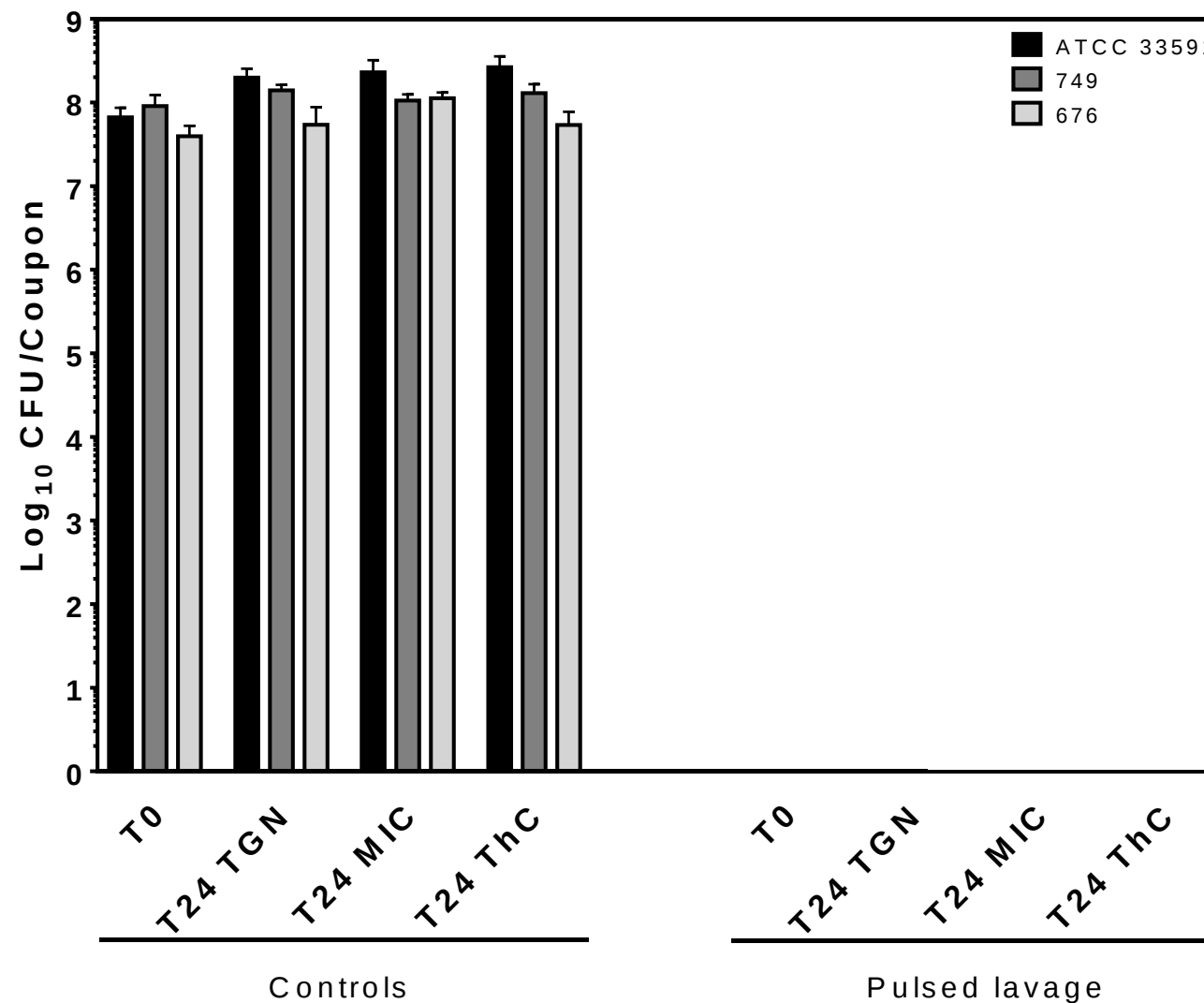
MIC (mg/L) values for the tested strains

	Strains	Oxacillin	Vancomycin	
		CA-MHB	CA-MHB	TGN
MRSA	ATCC 33591	>64	1	4
	749	>64	1	8
	676	>64	1	8

CLSI breakpoints values (in CA-MHB; mg/L): Oxacillin: S≤2, R≥4;

Vancomycin: S≤2, R≥16.

Results: CFU counts



24h Biofilms
S. aureus

Controls

Extemporaneous
analysis

24h Reincubation
in medium

24h Reincubation
in medium +
vancomycin

MIC

Target through
concentration:
20mg/L

N experiments = 4

Two-way ANOVA followed by Holm-Sidàk test

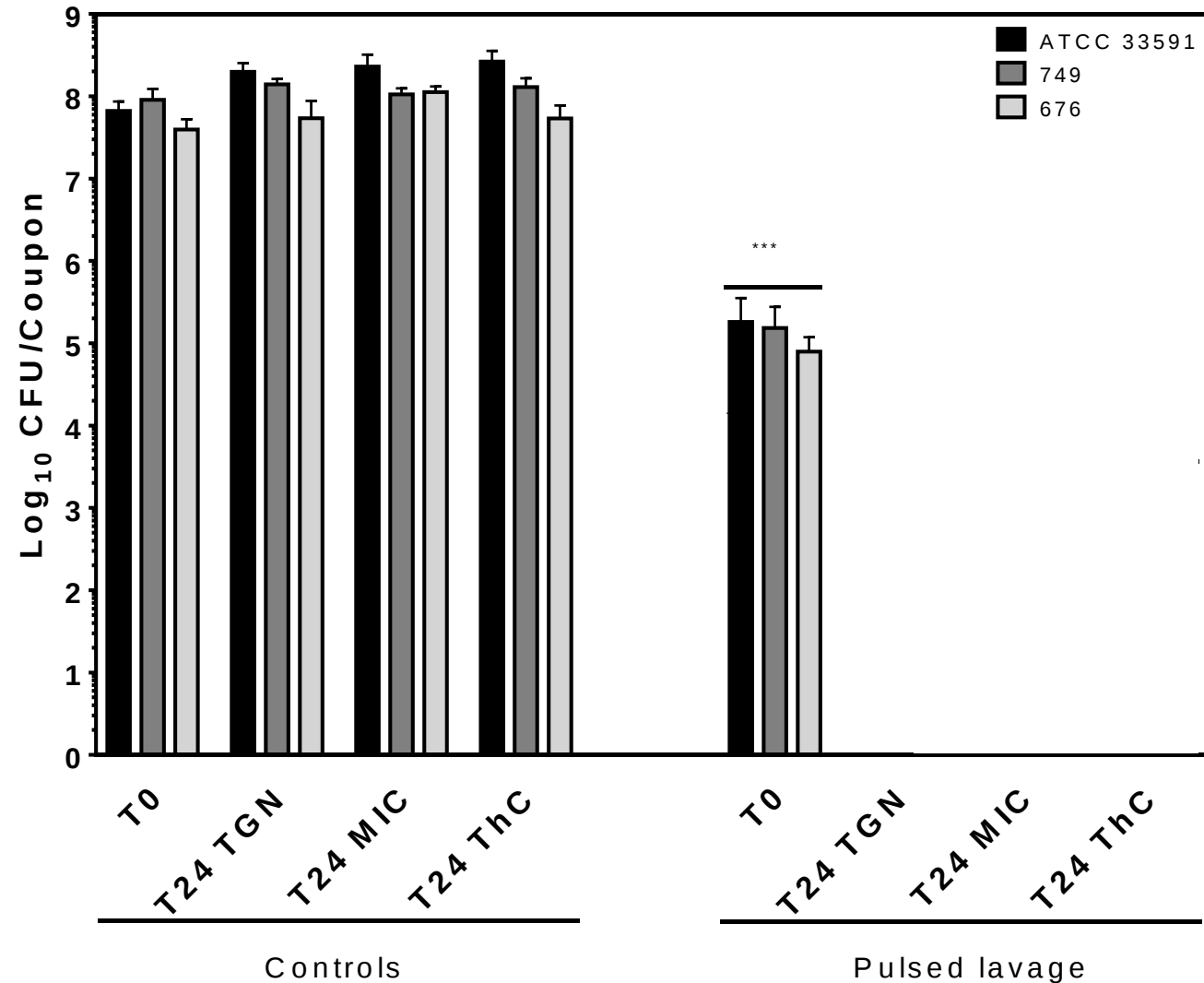
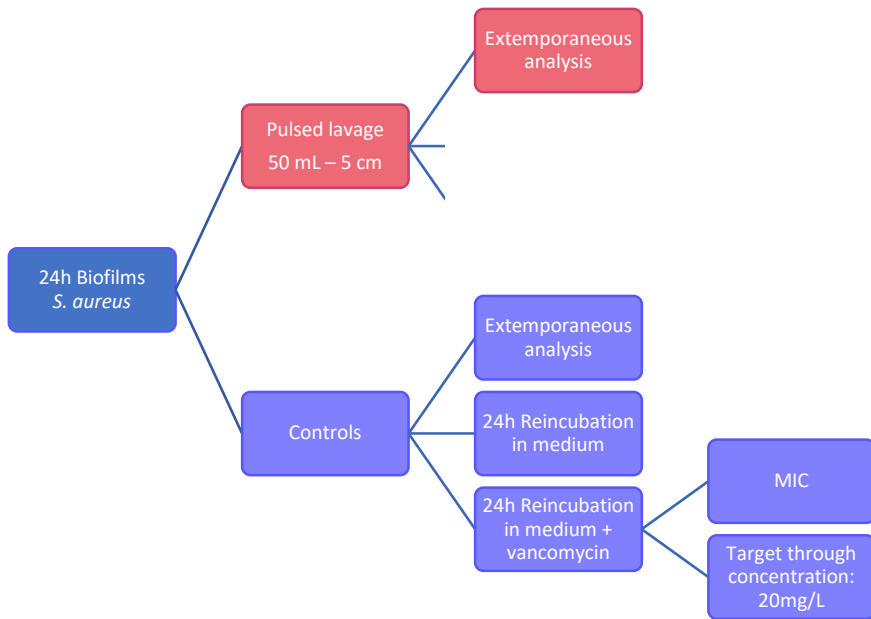
Comparisons with T0 control samples: *: $p < 0.05$;

: $p < 0.01$; *: $p < 0.001$

Comparisons with T0 pulsed lavage samples: †: $p < 0.05$;

††: $p < 0.01$; †††: $p < 0.001$

Results: CFU counts



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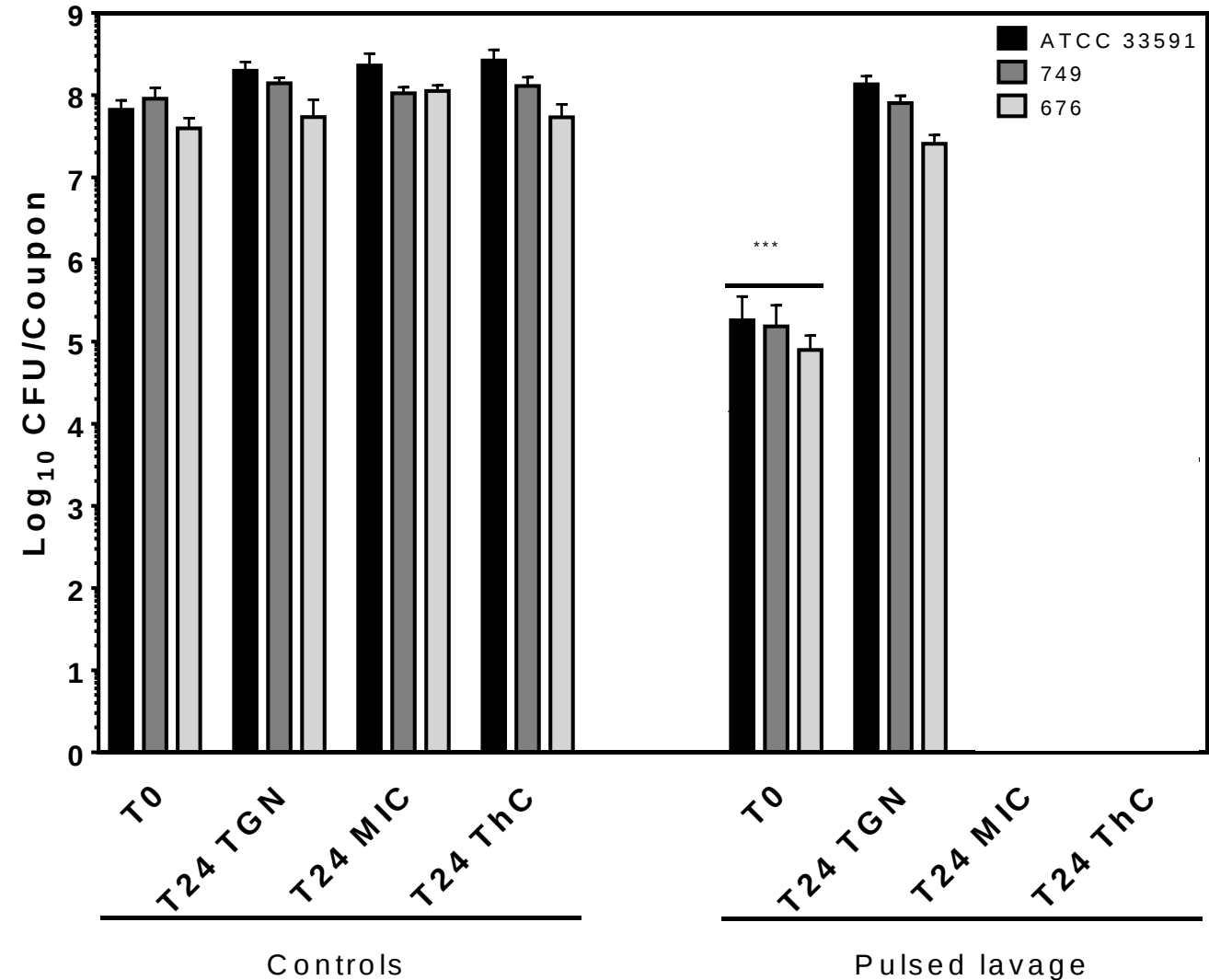
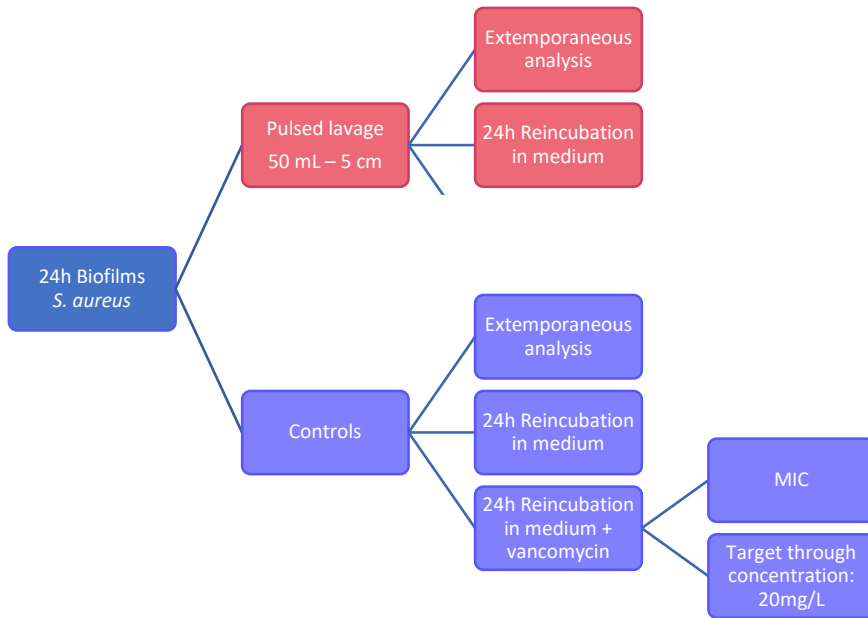
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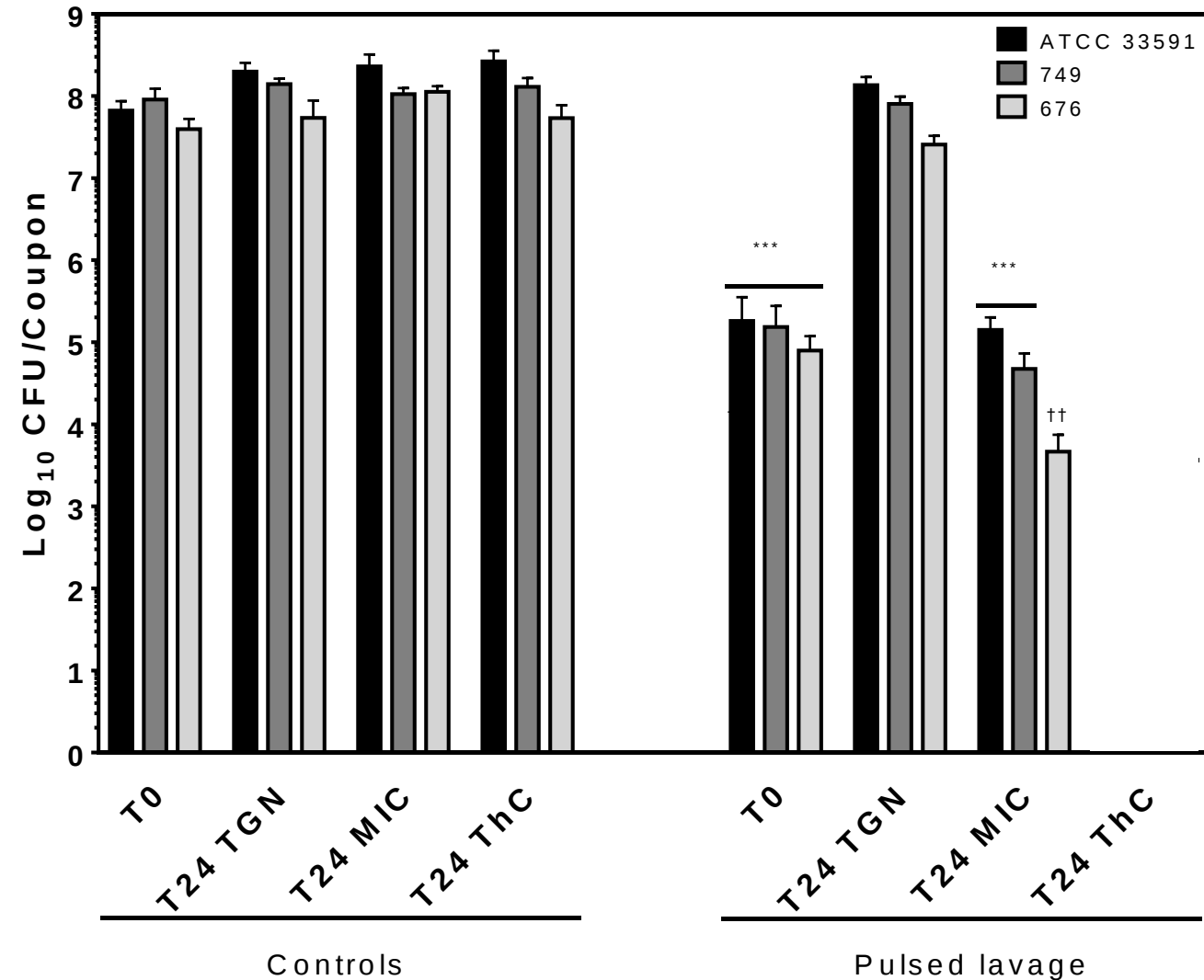
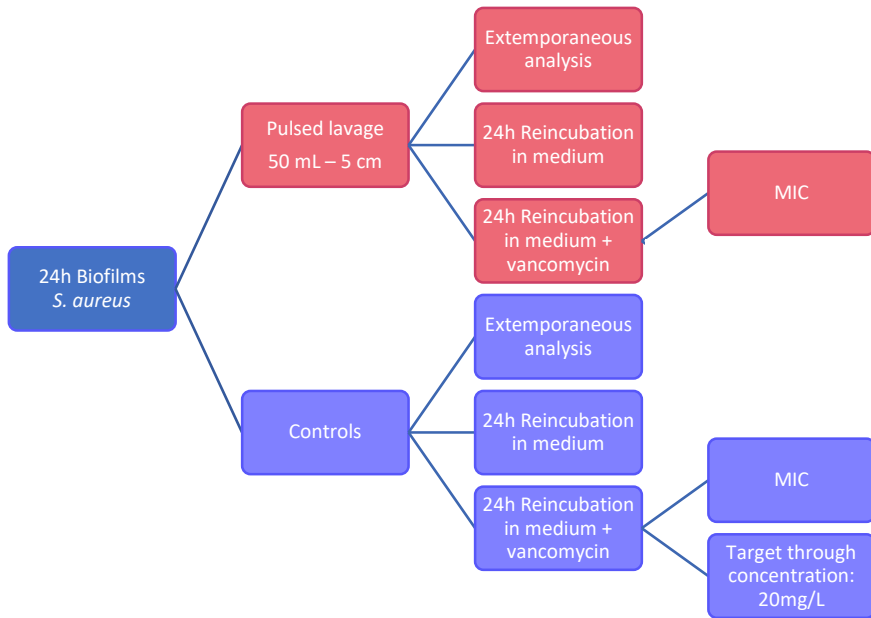
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Results: CFU counts



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Results: CFU counts



N experiments = 4

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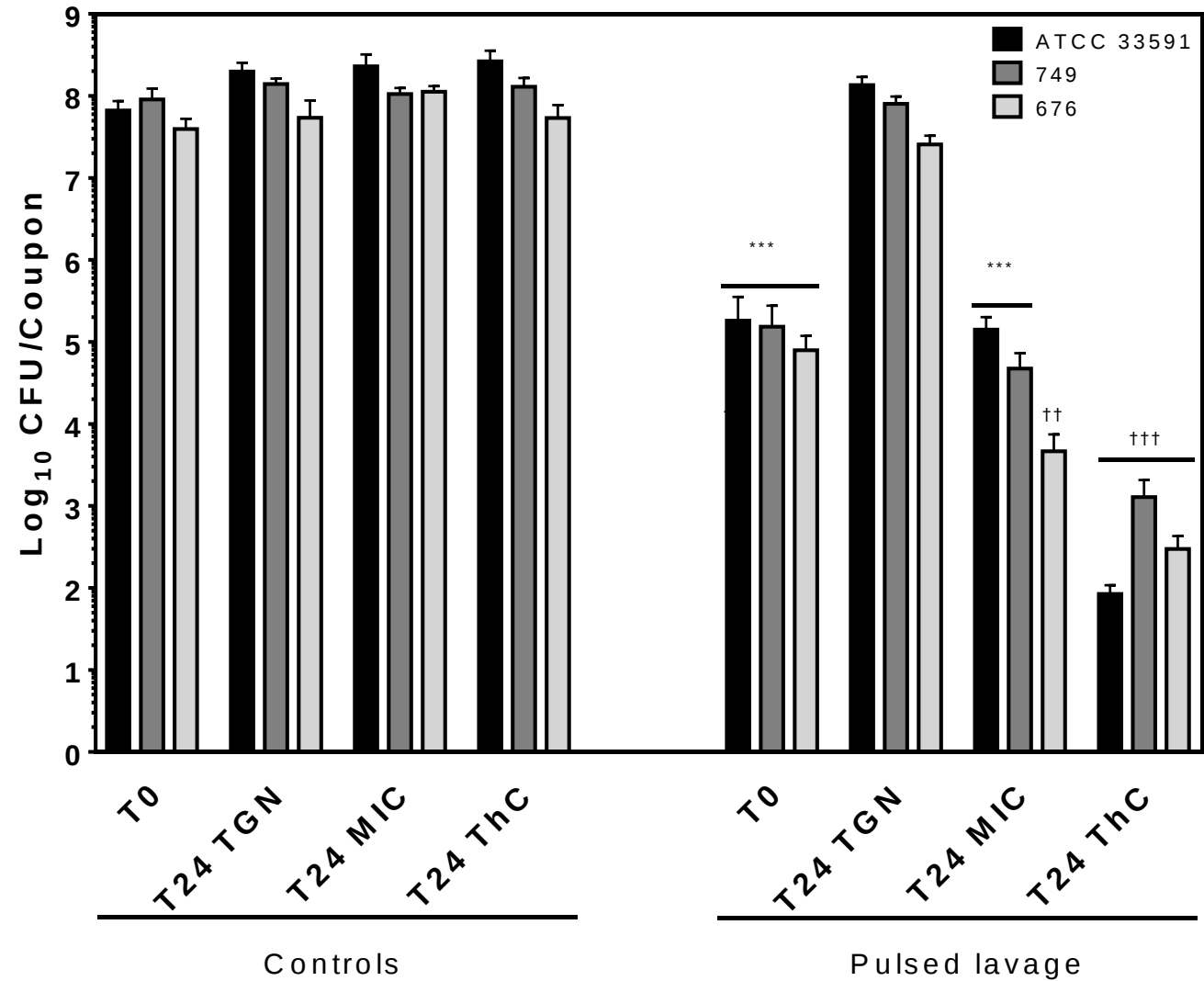
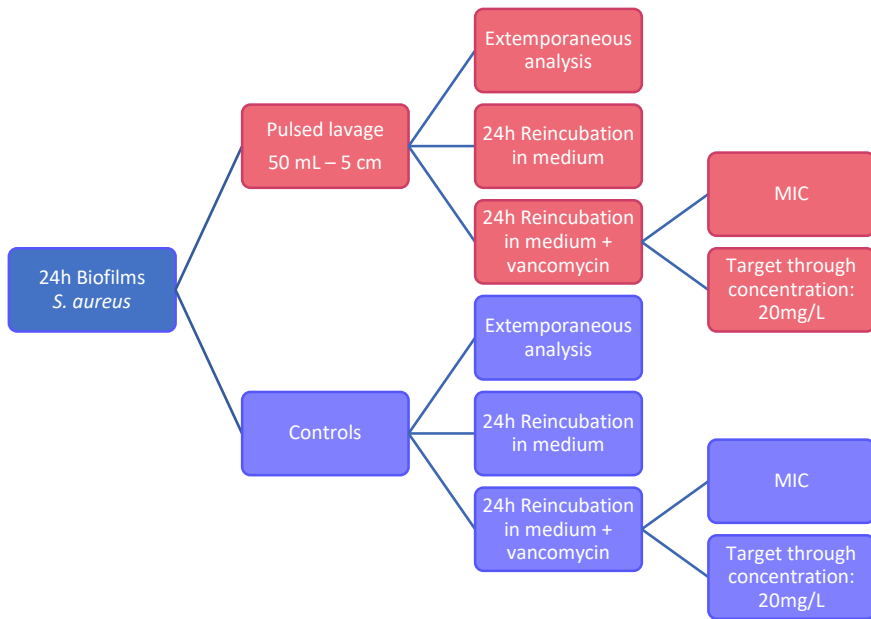
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Results: CFU counts



N experiments = 4

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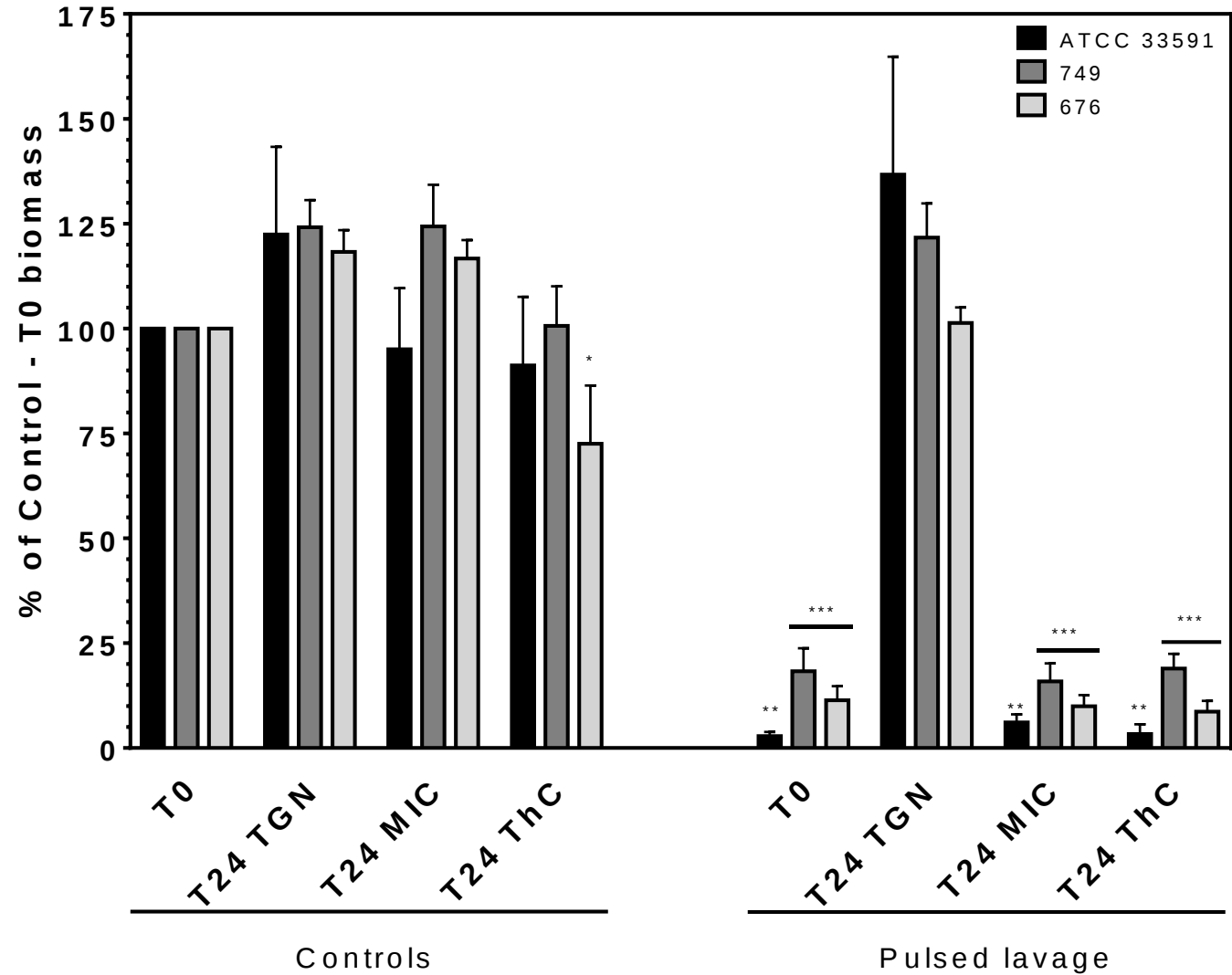
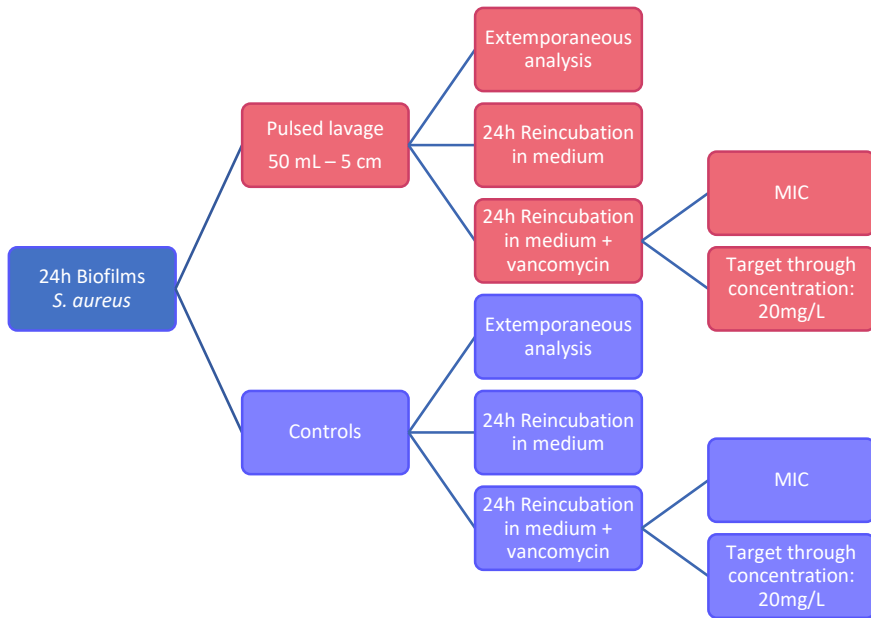
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Results: Biomass assay



N experiments = 4

Two-way ANOVA followed by Holm-Sidak test

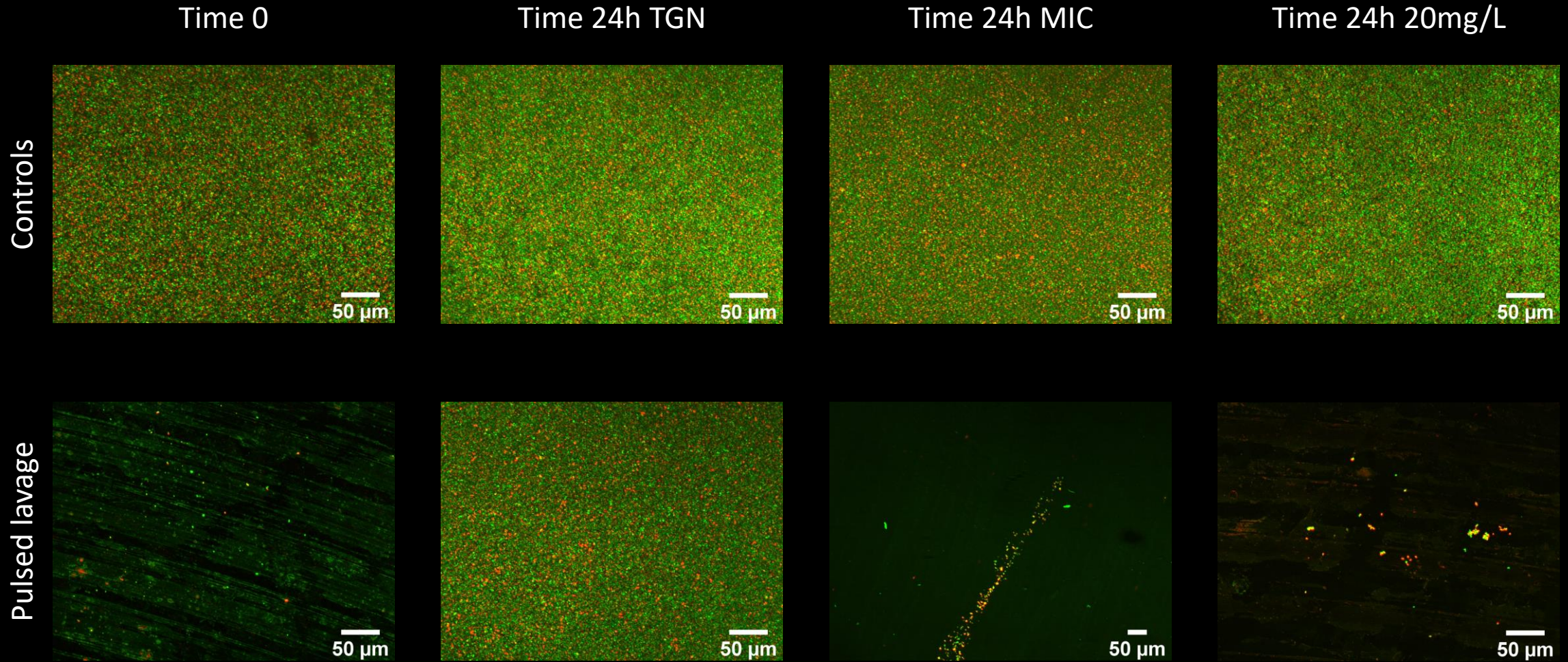
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Results: Live/Dead fluorescence microscopy



Discussion

- Pulsed lavage is an effective tool to reduce the bacterial load of MRSA biofilms on titanium surfaces
 - Previous studies hinted at a weaker effect of pulsed lavage in similar conditions [1-3]
- Vancomycin alone is ineffective against MRSA biofilms at clinically relevant concentration
 - Similar results in other models in the literature [4]
- Pulsed lavage and vancomycin at 20mg/L have a synergistic effect against MRSA biofilms
- A similar synergistic effect of pulsed lavage and flucloxacillin against methicillin susceptible *S. aureus* biofilms was observed

1. Schwechter, E., et al. *J. Arthroplasty* (2011) PMID: 21641757

2. Urish, K., et al. *J. Arthroplasty* (2014) PMID: 24439797

3. Smith, D., et al. *J Arthroplasty* (2015) PMID: 26105616

4. Diaz-Iglesias, Y., et al. *Antimicrob Agents Chemother* (2019) PMID: 31036685

Conclusion

Should we continue to use pulsed lavage?

Yes ... if followed by antibiotics at adequate dosages



MSSA strains results

