IOCBE 2024 Conference
The 1st International Online Conference on Bioengineering
16–18 October 2024 | Online
MDPI

Use of Low-Level Electrical Stimulation for Bacterial Inhibition

Savannah Mosier[1], Jordan Gamble[2], Candan Tamerler[1, 2], Elizabeth Friis[1, 2]

[1]Bioengineering Graduate Program, School of Engineering, University of Kansas, Lawrence, KS, USA
[2]Department of Mechanical Engineering, School of Engineering, University of Kansas, Lawrence, KS, USA

INTRODUCTION & AIM

Introduction: Bacterial adhesion on medical devices can have severe consequences, such as prolonged inflammation and delayed tissue regeneration that results in chronic infections [1-3]. The most common treatment for bacterial infections by antibiotics, but this leads to the increased prevalence of antibiotic-resistant bacteria species. Recent reports in literature suggest the use of electrical stimulation (ES) to inhibit bacterial growth by the disruption of the bacterial membrane permeability [4]. However, results from these studies are largely inconclusive due to inconsistencies in the usage and reporting of stimulation parameters.

In this study, *S. epidermidis* (gram-positive) and *E.coli* (gram-negative) were used to determine the efficacy of low-level electrical stimulation against various bacterial wall types. In addition, custom fabricated petri dishes were used to tightly control the supplied electrical signals.

Study Aims:

- Provide controlled delivery of electrical stimulation and report all relevant stimulation parameters
- Explore EF distribution pattern in Tryptic Soy Agar via computational modeling
- Determine efficacy of clinically relevant low-level electrical stimulation against common causative agents of bacterial infections

METHOD

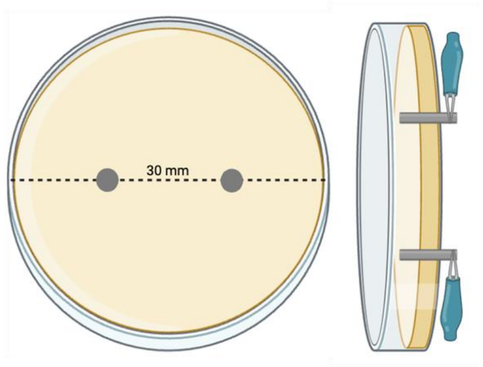

Figure 1a: (left) Top view depiction of petri dish fabrication with cpTi grade 5 rods separated 30 mm. (right) Side view of 10 mm overhang to allow for alligator clip connection. Images generated in BioRender.

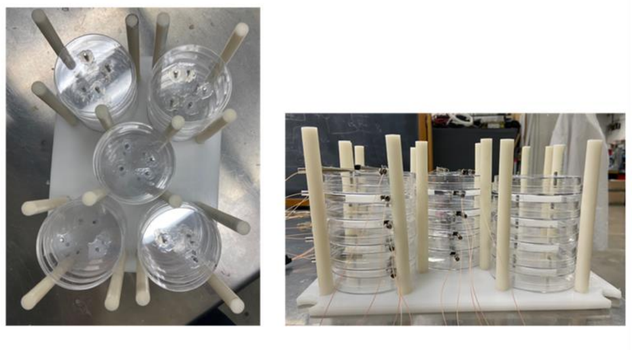

Figure 1b: Custom fabricated petri dish rack holders that were placed inside the incubator. (left) Top view to show five individual experimental group columns. (right) Side view to show stacking of petri dishes with Delrin spacers to keep petri dishes level.

Table 1: ES Parameter Combinations Tested on *S. epidermidis* and *E. coli.* Stimulation signals were created using the Analog Discovery 3 (Digilent®) Waveform Generator.

Group	Voltage (V)	Frequency (MHz)	Current Density at Electrodes (mA/cm^2)	Estimated Electric Field (mV/mm)
Control	0	0	0	0
Ti Control	0	0	0	0
Group 1	1.0	0.5, 0.75, 1, 3	12	1000
Group 2	0.3	0.5, 0.75, 1, 3	4	333
Group 3	0.1	0.5, 0.75, 1, 3	1.2	100

Simplified Experimental Protocol

- Bacteria Culture to Mid-Log Phase
- Swab Lawns then 30-minute rest
- 30-minute Electrical Stimulation
- 24-hour incubation at 37ºC
- Zone of Inhibition Images

RESULTS & DISCUSSION

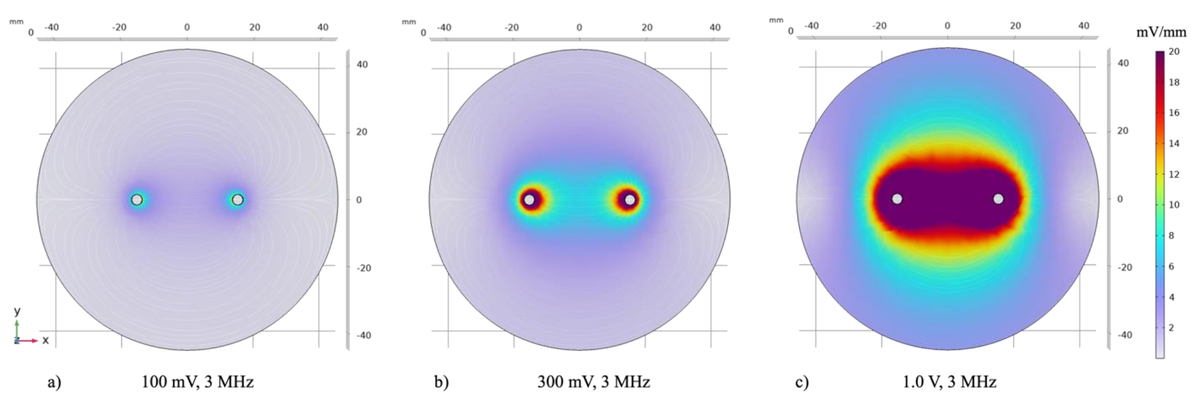

Figure 2: Top-down (XY) view of petri dish model with the peak Electric Field (mV/mm) distribution shown. a) 100 mV at 3 MHz, b) 300 mV at 3 MHz, and c) 1.0 V at 3 MHz Scale bar set to show distribution pattern across all supplied voltages. All models were generated in COMSOL Multiphysics and the AC/DC Module.

- Electric field scales proportionally with supplied voltage but is independent of frequency
- Gradual decrease in intensity as the radial distance from the electrode increases
- Provides insight to potential zone of inhibition sizes

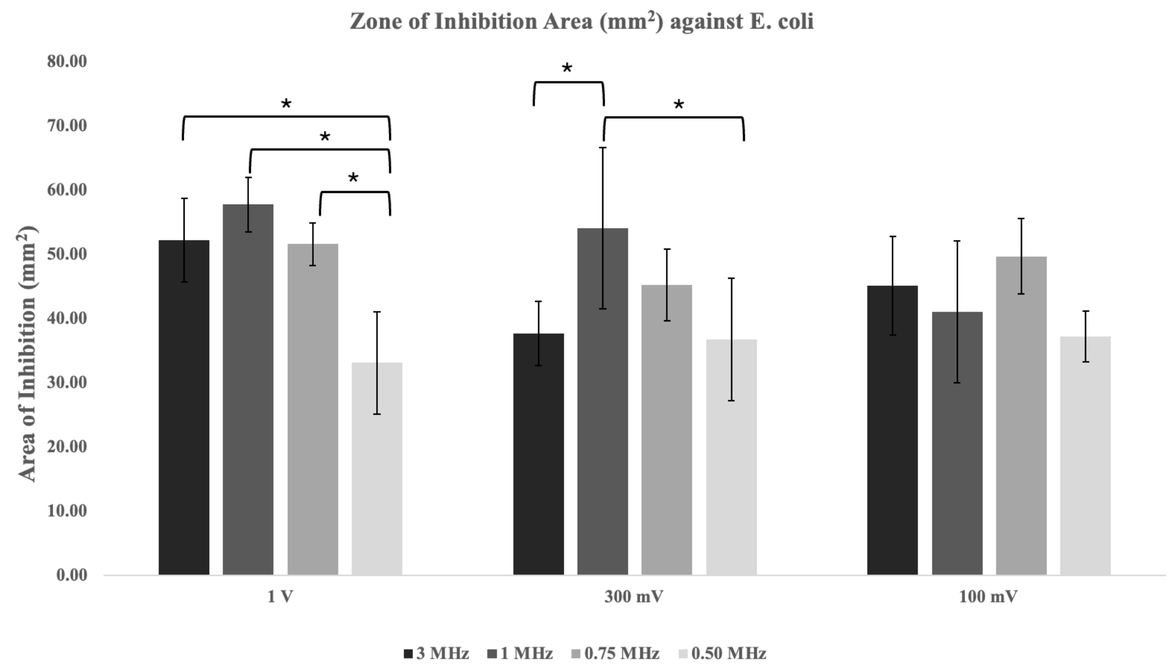

Figure 3: Zone of Inhibition around electrodes for E. coli after 30 minutes of electrical stimulation. * Indicates a significant difference based on Tukey-Kramer post-hoc test.

- *S. epidermidis* was not affected by any stimulation and showed zero zones
- All stimulation parameters proved effective against *E. coli*
- Overall largest average zone of inhibition area produced by 1V at 1 MHz
- No clear trend on if voltage or frequency is the key factor contributing to the inhibition zones

CONCLUSION

- Development of controlled electrical stimulation delivery
- Computational modeling aids in understanding the electric field distribution
- Ineffective inhibition against *S. epidermidis* but various levels of inhibition against *E. coli*
- Low-level electrical stimulation can effectively inhibit gram-negative species

FUTURE WORK / REFERENCES

Future Work will include studies against MRSA and *P. aeruginosa* bacterial species as they are common causative agents for post-operative infections. Additional studies should be done against biofilms formed on a material for a more clinically relevant environment.

References

[1] Vickery, K., Hu, H., Jacombs, A. S., Bradshaw, D. A. & Deva, A. K. 2013. A Review Of Bacterial Biofilms And Their Role In Device-Associated Infection. *Healthcare Infection*, 18, 61-66.
[2] Rouabhia, M., Park, H., Meng, S., Derball, H., Zhang, Z. 2013. Electrical Stimulation Promotes Wound Healing By Enhancing Dermal Fibroblast Activity And Promoting Myofibroblast Transdifferentiation. *Plos One*, 8.
[3] Rajendran, S. B., Challen, K., Wright, K. L. & Hardy, J. G. 2021. Electrical Stimulation To Enhance Wound Healing. *Journal Of Functional Biomaterials*, 12, 40.
[4] Czerwińska-Główka, D. & Krukiewicz, K. 2020. A Journey In The Complex Interactions Between Electrochemistry And Bacteriology: From Electroactivity To Electromodulation Of Bacterial Biofilms. *Bioelectrochemistry*, 131, 107401.