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# Enhancement of antibacterial effect of plasma activated water with pulsed electric field

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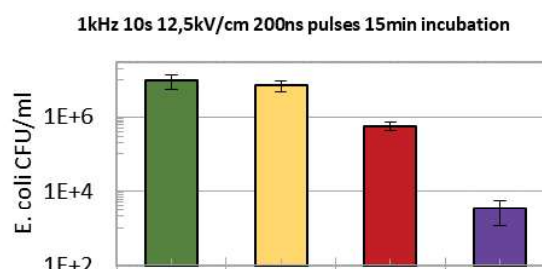
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Operative bacterial infections are still responsible for several thousand deaths per year, requiring the use of disposable medical equipment or more reliable sterilization. Non-thermal plasma of Transient Spark (TS) discharge induced by several kilovolts drop during 20 ns and driving a current of several tens of amperes demonstrated a significant antibacterial effect when combined with water electrospray. However, the mechanisms have not been clearly identified yet. Previously we observed significant bacteria reduction after direct exposure of *E. coli* bacterial suspension to plasma compared to only Plasma Activated Water (PAW) produced by TS with water electrospray. The difference between the results may be due to different plasma products, such as electrons, ions, UV, acidic condition, electric field, heating, Reactive Oxygen and Nitrogen Species (RONS), which were clearly identified as having strong antibacterial effects individually.

However, there are synergies between the different plasma products, which explain the stronger effect of direct exposition. Here we focus on the antibacterial effect of RONS in PAW coupled with the effect of electric field. RONS are known to damage the cell membrane, proteins and DNA that explains this antibacterial effect of PAW. Applying a high electric field moves the lipid bilayer of the cell membrane; which expands the membrane pore size depending on the length of the pulse and the electric field intensity. These pores allow the penetration of molecules, such as RONS, DNA or drugs into the cell.

To highlight this antibacterial effect accentuation due to the coupling between the RONS and electric field we have carried out repeated experiments with 4 conditions:

- 1) control where bacteria are incubated for 15 minutes in Deionized water (DW).
- 2) bacteria in DW are exposed to pulsed electric field and incubated for 15 minutes
- 3) bacteria are incubated for 15 minutes in PAW
- 4) bacteria in PAW are exposed to pulsed electric field and incubated for 15 minutes



**Figure 1: *E. coli* antibacterial effect of electric pulses, PAW and their combination. CFU/ml in log scale and standard deviations as error bars.**

After incubation, the liquids containing the bacteria after treatments were deposited on petri dishes containing agar gel mixed with bacterial nutrient (Lauria-Bertani broth). After one-night incubation, counting of the survived bacteria is done, each of which is a macroscopic colony. The results (Figure 1) are presented for 5kV/cm pulses length for a duration of 200 ns at 1 kHz during 10 s.

The results illustrate a stronger antibacterial effect when RONS are coupled with high electric field pulses.

## References

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