Comparison of direct and indirect effects of cold air plasma on bacteria contaminated surfaces

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Résumé
Experimental work concerning non-thermal air plasma treatment of gram-negative bacteria *Salmonella typhimurium* on agar surfaces is presented here comparing direct and indirect exposition to the plasma for various times of exposition. The results are characterized by visible differences of inactivated areas between these two methods of treatment.

Introduction

Bacteria inactivation with non-thermal atmospheric pressure plasma in air is a highly complex process including many possible stress agents, such as charged particles, reactive neutral species, UV and electromagnetic radiation and heat. In recent years, the most discussed agents mainly contributing to efficiency of plasma induced bio-decontamination are charged particles and reactive neutral species [1, 2, 3].

Experiment

In our work, bacteria contaminated agar surfaces are treated by non-thermal atmospheric pressure air plasma induced by DC driven electrical discharges. Experimental setup was prepared the same way as in [4] to compare direct and indirect exposition of the agar surface on Petri dishes to the discharge. This was especially set to separate reactive neutral species and charged particles from the plasma.

Positive transient spark (TS), a DC-driven electrical discharge used in point to-plane geometry, is a transient streamer to spark discharge with self-driven pulse regime. TS is characterized by relatively high energy in very short pulses (10 – 100 ns) with temperatures ~ 550 ± 100 K [5]. The current pulses were of amplitude 2 – 3 A, with frequencies around 1 – 2 kHz and the average power of ~ 2 W.

Our biological sample *Salmonella typhimurium* was prepared by cultivation in liquid nutrient broth and then spread onto a solid agar over the whole surface in Petri dishes. Prepared samples were directly or indirectly treated with plasma and incubated for 12 h. Exposure time was set to 5, 10, 15 s, which is much shorter than in our previous work [4]. In the Petri dishes for direct exposition, a conductive wire was immersed into agar, acting agar surface as one electrode. In indirect exposition, we placed a grounded mesh over the agar surface. This mesh filters the charged species and enables only neutral active species to reach the agar surface. Schematics of the experimental set-ups is in figure 1. In both exposition methods we tried to ensure same electrical parameters. For this purpose, additional resistor simulating agar resistance was added between the mesh and the ground. The experiments were repeated 3 times with initial bacterial populations on agar surface of 10^4-10^5 CFU.

Results

The effects of plasma on contaminated agar are clearly visible as dark voids (Figure 2), whereas control samples were homogeneously covered by cultivated bacteria (bright). The results show that direct exposition has slightly stronger effect than indirect. However, even for small times of exposure, the effect of the indirect exposition is comparable with the direct exposition. Similar results were obtained previously for 1-2 min exposure times. This indicates that neutral reactive species generated in the discharge are crucial in bacterial inactivation even at very short times (5-15 s).

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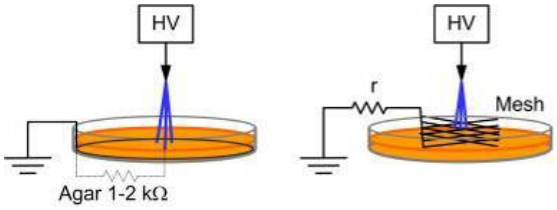

Fig. 1: Schematics of experimental setup, left: direct; right: indirect exposition.

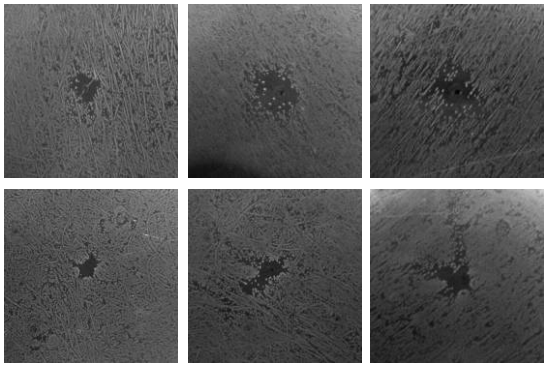

Fig. 2: Plasma treated agar surfaces with *Salmonella typhymurium* for various exposition times, top: direct, bottom: indirect exposition.

References

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