Comparison of Point-to-Plane and Point-to-Point Corona Discharge for the Decontamination or Sterilization of Surfaces and Liquids

V. Scholtz,[1],* J. Julák,[2] & B. Štěpánková[1]

[1]Department of Physics and Measurements, Faculty of Chemical Engineering, Institue of Chemical Technology, [2]Institute of Immunology and Microbiology, 1st Faculty of Medicine, Charles University

*Address all correspondence to: V. Scholtz, Department of Physics and Measurements, Faculty of Chemical Engineering, Institute of Chemical Technology, Technická 5, 166 28, Praha, Czech Republic; scholtzv@vscht.cz

ABSTRACT: We studied the decontamination of surfaces and inactivation of bacteria and yeast in liquids by low-temperature plasma generated in the DC corona discharge of an open-air type in the point-to-plane or point-to-point arrangement. We found that the inactivation in liquid suspensions is slightly more efficient with the point-to-plane discharge in comparison with the point-to-point one. In the case of agar surface decontamination, two types of inhibition zones were observed, which indicates the different mechanism of action for the point-to-point and point-to-plane dicharges. This knowledge may be important in the future selection of microbicidal agents and in the development of efficient methods for low-temperature plasma decontamination or sterilization.

KEY WORDS: decontamination, inactivation, corona discharge, bacteria, yeast

I. INTRODUCTION

The action of the plasma generated by electric discharges is one of the possible methods of inactivation of bacteria and other microbes, mediated by the bactericidal action of UV light and reactive particles. The method is not yet frequently used in practice, but it is potentially important, especially for the decontamination or even sterilization of heat-labile or otherwise sensitive materials. The various experimental arrangements, advantages, and status of research in this field were reviewed in detail in many works, e.g.,.[1–5] This contribution is a part of our systematic study of properties and differences of various types and arrangements of DC discharges and their microbicial effects.

II. APPARATUS, METHODS, AND THE MICROORGANISMS UNDER STUDY

We studied the inactivation of microbial suspensions on wet surfaces and in liquids by low-temperature plasma generated in the DC corona discharge. The used simple apparatus of an open-air type enabling the point-to-plane or point-to-point arrangement was previously described in Ref. 6 and is schematically shown in the Figure 1.

The negative point-to-plane corona discharge was generated on the point electrode

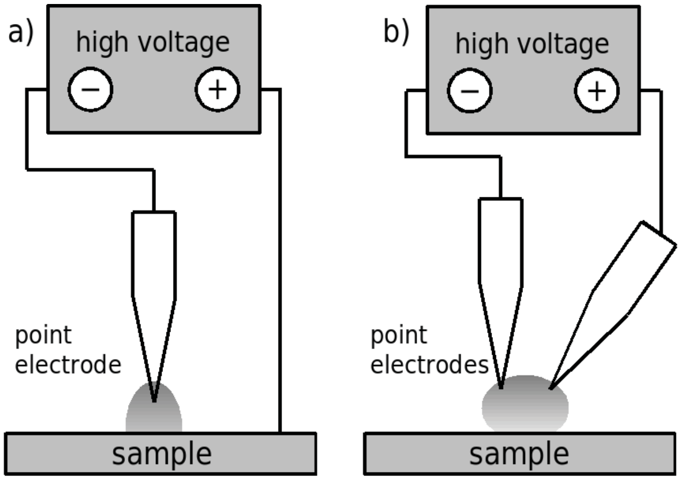

FIGURE 1. Schematical arrangement of used apparatus and the negative point-to-plane (a) and point-to-point (b) corona discharge.

represented by the tip of a hypodermic needle and situated 4 mm over the sample. The plane anode was the conducting surface of an agar cultivation medium. The bipolar point-to-point corona discharge was generated on a pair of hypodermic needles arranged at an angle of 30 deg with tips approximately 4–6 mm apart and situated 4 mm over the sample.

For the surface decontamination, 1 ml of the microbial suspensions were inoculated onto the surface of Sabouraud or Mueller-Hinton agar (Lab M, Ltd.), diluted to obtain a microbial concetration of 10^6 cfu/cm^2. After the suspension soaked, the samples were exposed to the corona discharge and incubated at 37 °C for 24 h, and the inhibition zones were measured. In the case of liquid samples, a 0.25 µl drop of an appropriate microbial suspension in physiological saline, adjusted to the concentration of ca. 10^6 cfu/ml, was placed on the hydrophobic surface of a sterile silicone disk and exposed to the discharge. The disk was then washed in 1 ml of physiological saline; this saline was inoculated onto Sabouraud or Mueller-Hinton agar, incubated as above, and the number of colonies was counted.

The microorganisms under study were "wild" strains of the following species isolated at the Institute of Immunology and Microbiology: a yeast, *Candida albicans*, a gram-negative bacterium, *Escherichia coli*, and a gram-positive bacterium, *Staphylococcus epidermidis*.

III. EXPERIMENTS

The experiments were performed under the following parameters. The point-to-plane discharge was adjusted to the current I = 0.05 mA and its voltage was U = 4.6 kV or

U = 9 kV for the surface or liquid sterilization, respectively. The point-to-point corona discharge was adjusted to the current I = 0.2 mA and voltage U = 10 kV. The different parameters for particular discharge types were determined by the different character and geometry of discharges. The values represent a compromise between discharge stability (transition into spark) and its energy, and enable the qualitative comparison of both discharge types.

The samples on the surface of agar were exposed for 8 min. The samples of liquid suspension were exposed for different times from 0.5 min up to 8 min, with the step of 30 s.

IV. RESULTS

For liquid samples exposed to the point-to-plane discharge, we found that all bacterial and yeast suspensions yielded no growth on Mueller-Hinton or Sabouraud agar after 2 min of exposure. In the case of point-to-point discharge, the complete inactivation was observed after 2 min for *S. epidermidis*, 2.5 min for *E. coli*, and 4 min for *C. albicans*. Although the parameters and power of both discharge types were different, for the liquid decontamination, a comparable inactivation effect may be concluded.

More interesting results were obtained after exposure of inoculated agar surfaces. In the case of point-to-plane discharge, we obtained circular and sharply bordered inhibition zones with diameters of 5–6 mm, which were completely clear and contained no growing microbes. However, after exposure to point-to-point discharge, we obtained two types of asymmetric fan-shaped zones of incomplete inhibition. In the case of *C. albicans* yeast and gram-positive bacterium *S. epidermidis*, the inhibition zones were well bordered and almost clear of any surviving colonies, whereas for the gram-negative bacterium *E. coli*, we obtained larger fan-shaped zones containing a reduced number of surviving colonies and very small, if any, zone of total inhibition only. The areas of total inhibition were approximately of 3 cm^2 for *C. albicans* and 1 cm^2 for *S. epidermidis*. The zone of incomplete *E. coli* inhibition was of 10 cm^2. The representative photographs of the zones obtained for point-to-plane and point-to-point discharges are shown in Figures 2 and 3, respectively. As contrasted to the inactivation in suspension, the inhibition effect of the point-to-point discharge differs from the effect of the point-to plane arrangement. This is probably caused by the different plasma-chemical processes in the discharge.

V. CONCLUSIONS

We found that the microbial inactivation in liquid suspensions is slightly more efficient using the point-to-plane discharge geometry than in the point-to-point arrangement. In the case of surface decontamination, it can be concluded that the mechanisms of microbial inactivation are different in these two discharge types. While in the case of point-to-plain discharge the inhibition zones are similar, in the case of point-to-point discharge, the appearance of two different inhibition zones occurs. This fact supports the conclusion that the sterilization agents are of a different nature in the point-to-point and

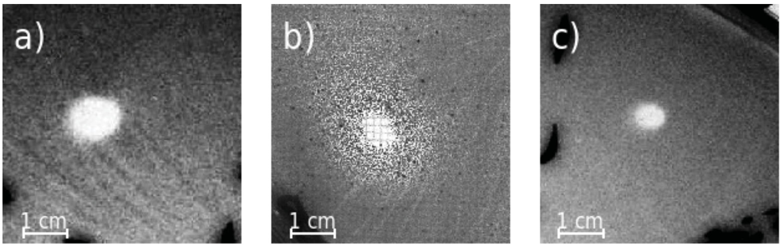

FIGURE 2. Representative photographs of the inhibition zones after 8 min of exposure to the negative point-to-plane corona discharge at *I* = 0.05 mA and *U* = 4.6 kV. Tested microorganisms were *C. albicans* (a), *S. epidermidis* (b), and *E. coli* (c)

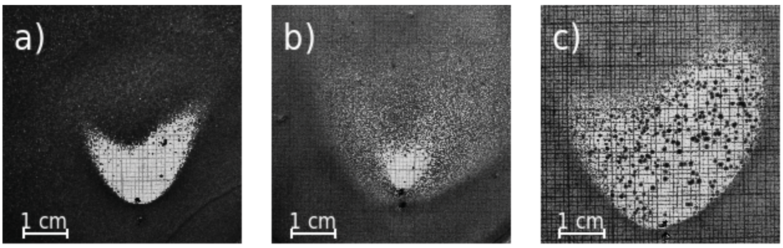

FIGURE 3. Representative photographs of the inhibition zones obtained after 8 min of exposure to the point-to-point bipolar corona discharge at *I* = 0.2 mA and *U* = 10 kV. Tested microorganisms are *C. albicans* (a), *S. epidermidis* (b), and *E. coli* (c)

point-to-plane discharges, displaying different efficiency and probably different mechanisms of action on microorganism's structures. We still have no explanation of this difference, which should be the subject of the future study, because it may be important for the selection of adequate microbicidal agents and for the development of efficient low-temperature plasma decontamination methods.

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