Water bio-decontamination by spraying through cold air DC discharge plasma

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Résumé

Two types of positive DC discharges in atmospheric air pressure (streamer corona and transient spark) were tested for bio-decontamination of bacteria in water solution. Both generate cold non-equilibrium plasma. The highest efficiency was achieved with the transient spark in flowing regime. Streamer corona was efficient when the treated solution flew through the active corona region. Electro-spraying was observed and resulted in fast bio-decontamination. Bacteria were handled and their population evaluated by standard microbiological cultivation procedures. Oxidation stress measurements in the cell membrane indicated that radicals and reactive oxygen species have the major role among the bio-decontamination mechanisms.

Introduction

Non-equilibrium plasma at atmospheric pressure finds numerous biological and bio-medical applications thanks to their reactive nature. It has been tested on a large variety of bacteria, spores, yeasts, viruses for their sterilization and interactions of plasma with live tissues, e.g. skin disinfection, blood coagulation, wound healing, dentistry.

Experimental set-ups

We investigated the DC discharges in point-to-plane geometry: a high voltage hollow needle electrode enabling the treated medium flow directly through the discharge and a mesh electrode. The gap between electrodes was 10 mm. DC high voltage was applied through the ballast resistor R (20 MΩ for SC, 6,6 MΩ for TS). The discharge voltage was measured by a high voltage probe Tektronix P6015A and the current was measured on a 50 Ω for SC and 1 Ω for TS. Current and voltage signals were processed by a digitizing oscilloscope Tektronix TDS 2024.

Treated microorganisms, microbial handling and cultivation procedure

Bio-decontamination effect of DC discharges was tested on Gram-positive *Bacillus cereus* in saline (physiological) solution. Initial population was 10^5-10^7 colony forming units per ml (CFU/ml). The treated water was collected in a sterile Petri dish. All steps of microbial of microbial cultivation were carried out in a sterile environment. For statistical evaluation, 3 Petri dishes were taken and incubated 12 h in a thermostat at 37 °C. The grown CFUs of each sample were counted and finally evaluated for inactivation efficiencies.

Measurement of oxidative stress

Reactive oxygen species interact with the bacterial cell membranes and result in the peroxidation of membrane lipids. The final product of lipoperoxidation is malodialdehyde (MDA), which is quantifiable by VIS spectrophotometry after the reaction with thiobarbituric acid (TBA) at 90-100 °C. This method of thiobarbituric acid substances (TBARs) was applied to measure the oxidative stress induced in bacteria in water exposed to SC and TS. We assigned the TBARs concentrations from the absorbance of MDA at 532 nm from Lambert-Beer's law with absorption coefficient $1{,}57.10^5$ $mol^{-1}.l.cm^{-1}$.

Results

Bio-decontamination of water solution contaminated by bacteria (*B. cereus*) was investigated in two types of positive DC discharges (TS and SC) at atmospheric pressure air in point-to-plane geometry. The fact that the treated water flows directly through the active zone of discharge region made these discharges very effective. The efficiency of transient spark was higher (median 99,68 %) than streamer corona

(median 81,26 %). TS decreased the initial microbial population roughly by 3 logs and SC by 1 log. We use a new parameter E-value to express the combined energy requirements and efficiency of the process (Joule per treated water volume and one log reduction of microbial population). Streamer corona is less energy-demanding (median E-value = 1.32 J/ml.log reduction) than TS (median 231.07 J/ml.log reduction), as shown in Figure 1 left. Figure 1 right shows the inactivation efficiencies for both discharges with the measured concentrations of TBARs compared with UV irradiation. Concentration Δc(TBARs) correlated with the inactivation efficiency of discharges. Comparison with UV that resulted in strong inactivation but no Δc(TBARs) indicates that the oxidations of cell membranes by reactive oxygen species are important in microbial inactivation.

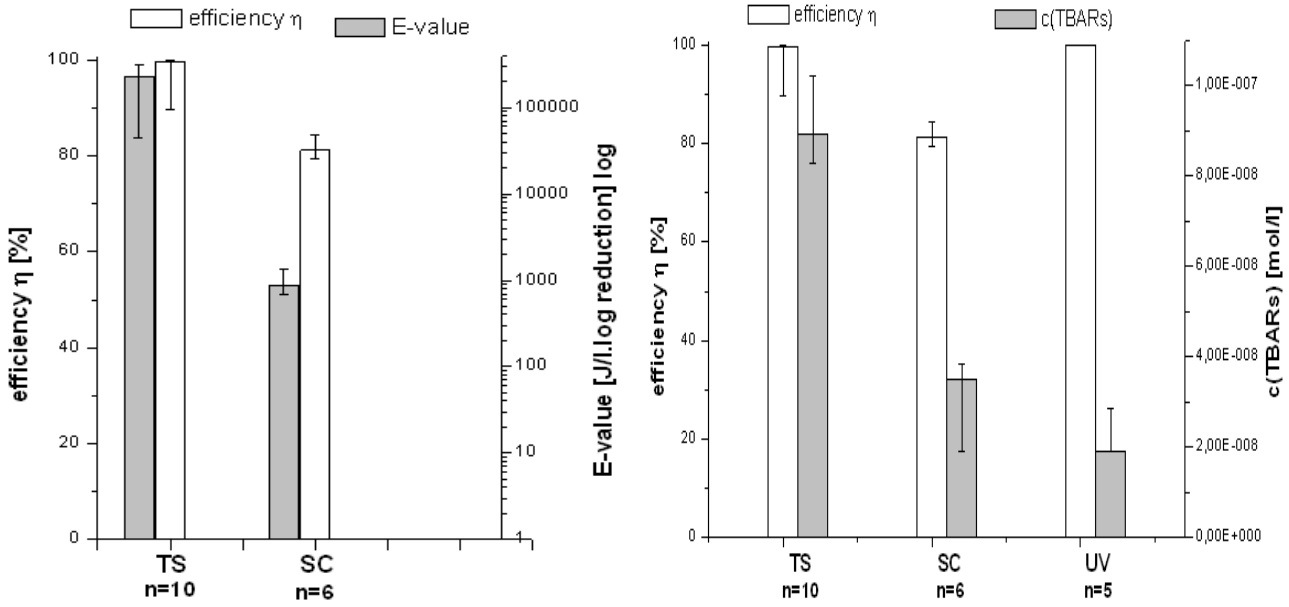

Fig. 1: Comparison of inactivation efficiency with E-value (left) and Δc(TBARs) (right) for positive TS and SC. Medians with 1st and 3rd quartiles. Results from UV irradiation are added in the right figure; n is the number of repeated experimental sets.

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