Sterilization process of bacteria by using atmospheric microplasma

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INTRODUCTION

In recent years, infective diseases such as new influenza caused by pathogenic organisms, were spread world wide. It is expected that alternatives to chemical sterilization methods such as new safety sterilizing process using nonthermal plasma to be developed.

We have investigated sterilization effect of bacteria at low voltage by using atmospheric microplasma.

METHODS

(1) Microplasma Electrodes

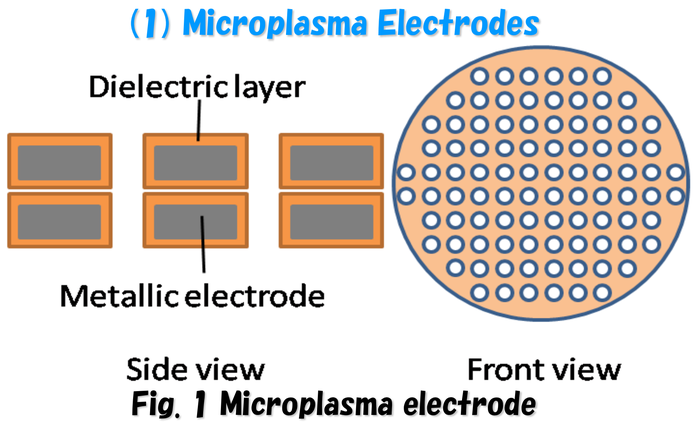


Fig. 1 Microplasma electrode

Pair of metal plates covered with dielectric materials is used as a dielectric barrier electrode.

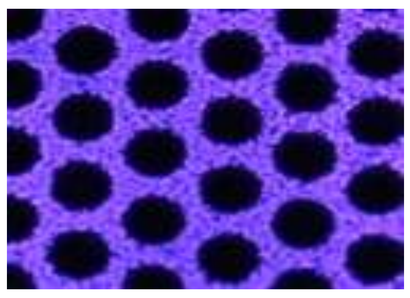


Fig. 2 Electrode surface

The image of microplasma electrodes during discharge at discharge voltage of 1 kV.

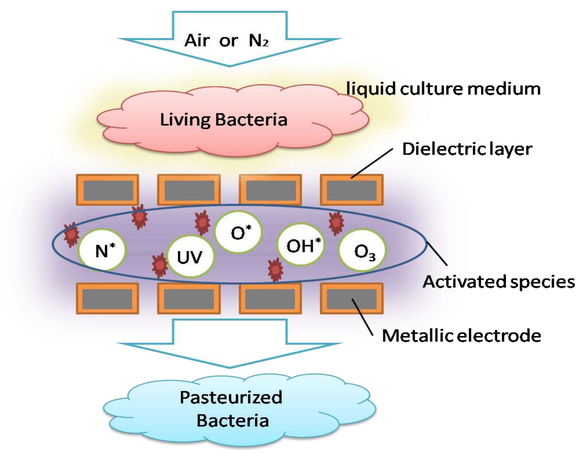


Fig. 3 microbicidal mechanism

Each bacteria could be attacked and sterilized by atomic oxygen and nitrogen species, UV, OH radicals and O_3 generated between the electrodes.

(2) Experimental Setup

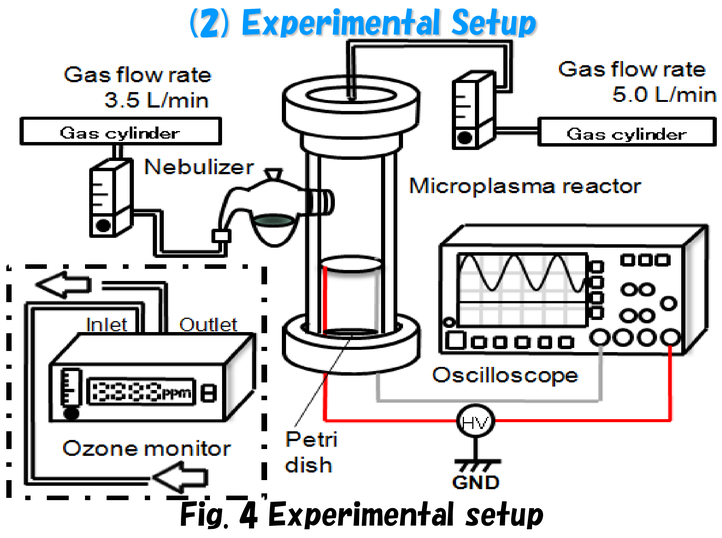


Fig. 4 Experimental setup

Table. 1 Experimental condition

Discharge voltage	**1.0～1.4 [kV]**
Gas flow rate	**8.5 [L/min]**
Carrier gas	**Air, N_2**
Target bacterias	***E.coli* , *B.subtilis***

The liquid culture medium was introduced in the microplasma reactor and sprayed at a gas flow rate of 3.5 L/min, through the electrode against a petri dish with culture medium. Total gas flow rate was 8.5 L/min. Sprayed bacteria with liquid culture medium was sterilized between microplasma electrodes. Discharge voltage and discharge current were measured by a digital oscilloscope.

RESULTS

(1) Electric Characteristics

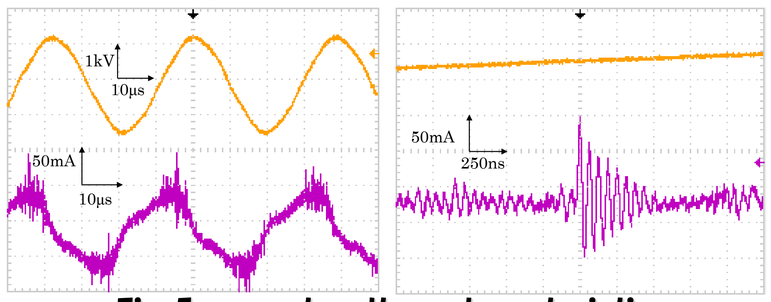


Fig. 5 current-voltage characteristic

Spike-like currents occured at the steepest slopes of the discharge voltage. This is a typical waveform of dielectric barrier discharge.
The microplasma reactor can generate atmospheric plasma at about 1 kV, since its discharge gap is narrow (about 50 to 100 μm).

(2) Ozone Generation

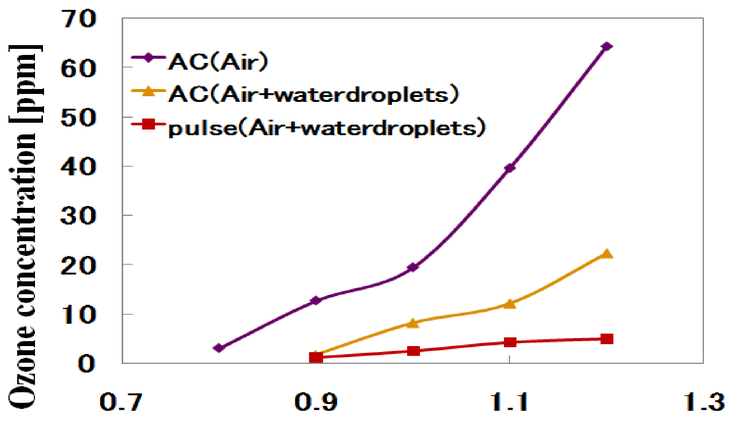
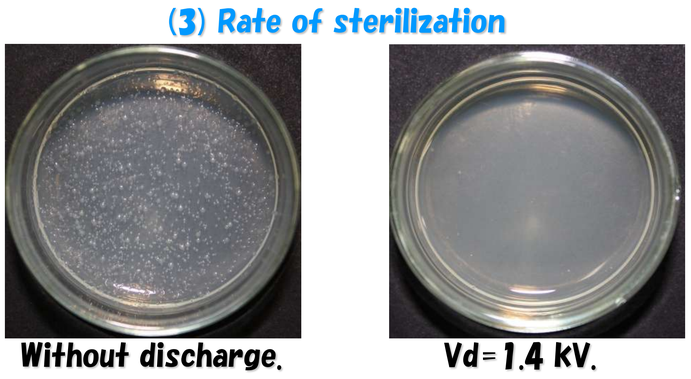


Fig. 6 Ozone concentration

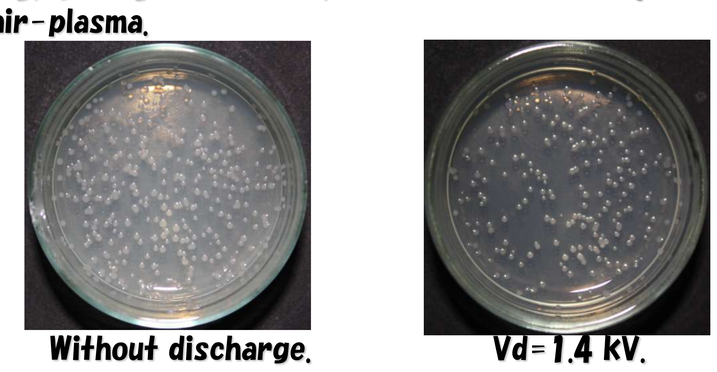
Generated ozone concentration by microplasma can be controlled by discharge voltage. It was observed that sprayed distilled water contributed to lower than ozone concentration with only air.

(3) Rate of sterilization



Without discharge. Vd=1.4 kV.

Fig. 7 Images of the *B. subtilis* treated samples by air-plasma.



Without discharge. Vd=1.4 kV.

Fig. 8 Images of the *B. subtilis* treated samples by nitrogen-plasma.

Sterilization rate was measured with only one pass process between the microplasma electrode.

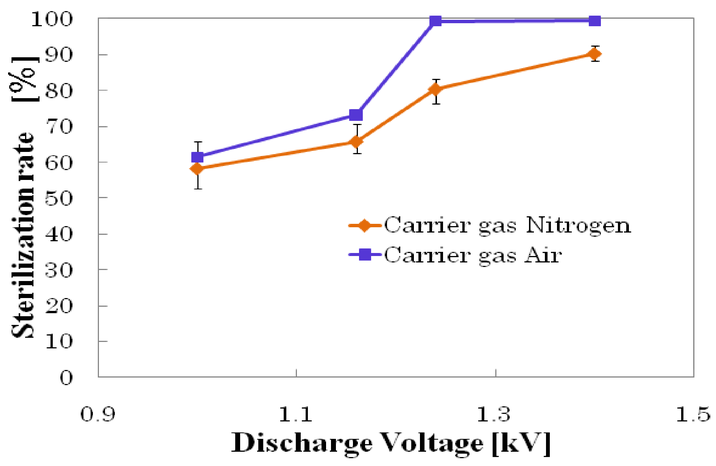


Fig. 9 Sterilization rate of *E.coli* by AC voltage application

The sterilization rate of *E. coli* versus discharge voltages for both carrier gases of air and nitrogen increased with discharge voltage . Sterilization rate with air was higher than with nitrogen plasma.

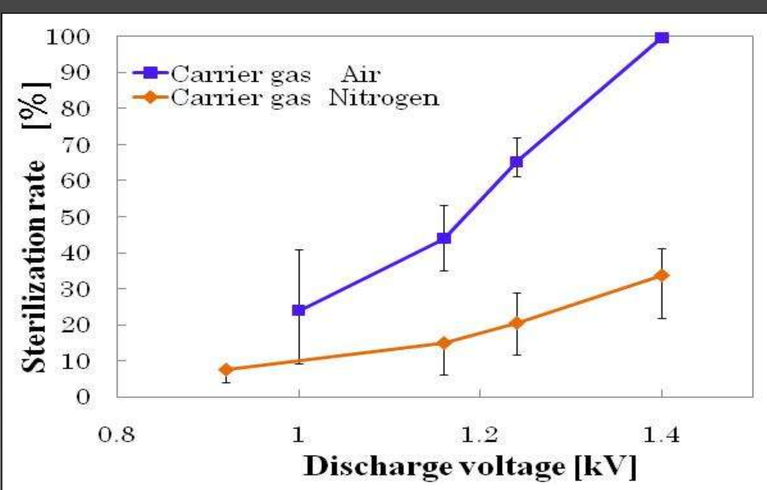


Fig. 10 Sterilization rate of *B.subtilis* by AC voltage application

Similar sterilization rates of *B. subtilis* were obtained for air as carrier gas. Sterilization rate by nitrogen plasma was lower compared to the rate of *E. coli*. The thickness of the cell wall could be considered the sterilization effect between *E. coli*

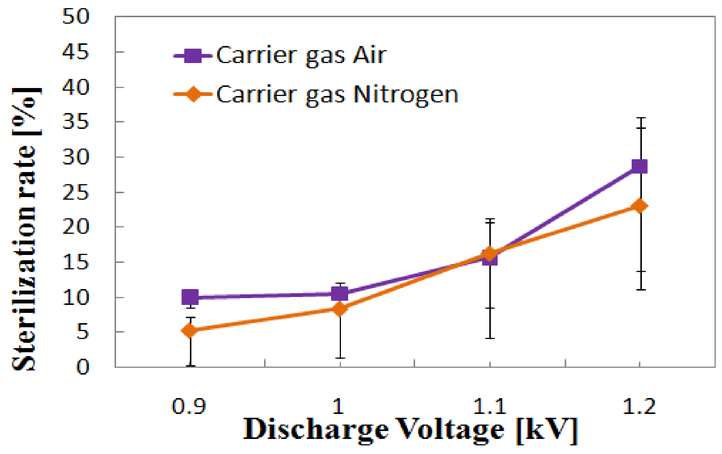
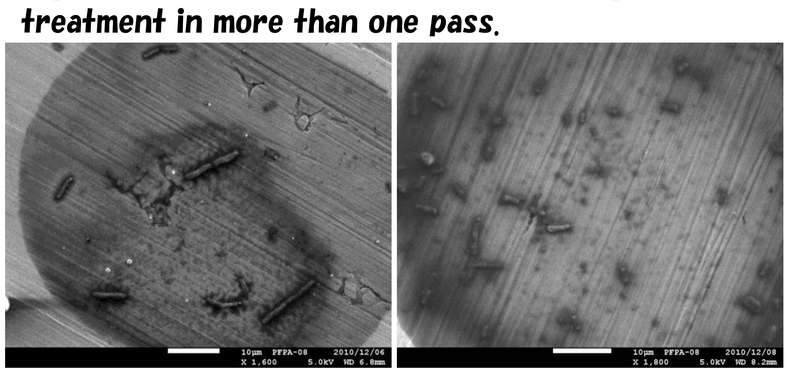
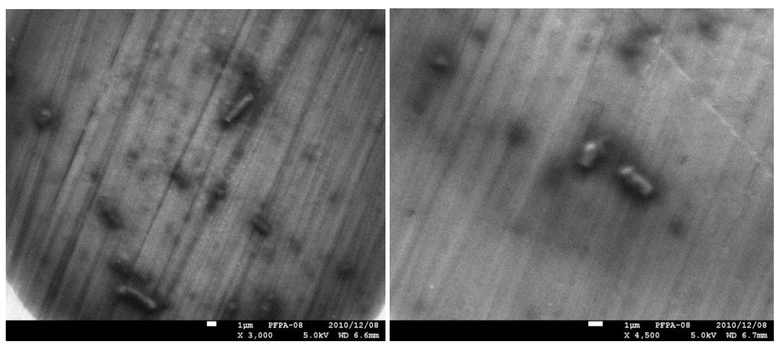


Fig. 11 Sterilization rate of *E. coli* with pulsed microplasma

Sterilization rate by pulse voltage application was lower than by AC voltage application. It is expected sterilization rate could be improved by treatment in more than one pass.



untreated(×1600) treated (×1800)



treated (×2300) treated(×4500)

Fig. 12 FE-SEM images of *B. subtilis* before and after the microplasma.

The photos of *B. subtilis* before and after treatment with nitrogen gas were taken by a SEM. *B. subtilis* bacterial could be identified as the cylindrical shape on the untreated photo which after the sterilization process transforms in a wider flat shape due to the effect of microplasma.

CONCLUSIONS

In this study, the following conclusions were obtained.

1) Sterilization by microplasma was confirmed at a relatively low discharge voltage of 1.0 kV for both carrier gases: air and nitrogen.

2) The sterilization rate by air plasma was higher than that of nitrogen plasma. It could be considered by the combination effect of high electric field, UV radiation and other radicals.

3) The sterilization rate of *E. coli* was higher than the results for *B. subtilis* due to the characteristics of the cell walls, which are different in thickness and chemical composition.