

# Effect of nitric oxide radicals on the proliferation of budding yeast

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We have investigated the effect of NO radical treatment on the proliferation of budding yeast and optimized treatment conditions. NO and O<sub>3</sub> densities were measured using UV absorption spectroscopy and the proliferation was evaluated with microscope with cell-counting chamber. From these results, we observed around 20 % increase of the number of yeast cells at a NO density of  $\sim 7 \times 10^{16} \text{ cm}^{-3}$ .

## 1. Introduction

Non-thermal atmospheric pressure plasmas (herein referred to as plasma) are gaining importance to use in biology, medicine, and agriculture. [1] The plasma is a mixed of electrons, ions, photons and neutrals. Recent our studies were focused on the correlation between microorganisms and plasma generated neutral reactive species. [2] Here, we focused on NO which is a well-known molecule in biomedical application. For examples, it is known that the NO enables to improve signal transmission between nerves, maintaining blood pressure, suppressing infection, and renewal tissue.

In this study, we report the effect of NO radical treatment on the proliferation of budding yeast and optimized treatment conditions.

## 2. Experimental

A commercially available atmospheric pressure plasma radical generator (Tough Plasma, Fuji Machine MFG Co., Ltd.) was used in this study. NO generated with mixture of O<sub>2</sub> and N<sub>2</sub> into buffered Ar (4 slm) through the radical generator. Flow rate of O<sub>2</sub> and N<sub>2</sub> was varied with a fixed total flow rate of 1 slm. The use of a large amount of Ar provides a high

electron density on the order of  $10^{16} \text{ cm}^{-3}$ . [3] The gas channel at downstream of the radical source is bended where high energy photons are intercepted and the electrically grounded electrodes on the gas channel and the nozzle exit (0.5 mm  $\times$  16 mm) terminate charged species.

The 3ml-yeast suspension was prepared in a 38-mm diameter petri dish and treated with a fixed distance of 1 cm between the slit exit of the radical source and the surface of the liquid suspension. The cells in counting chamber were counted by using a microscope.

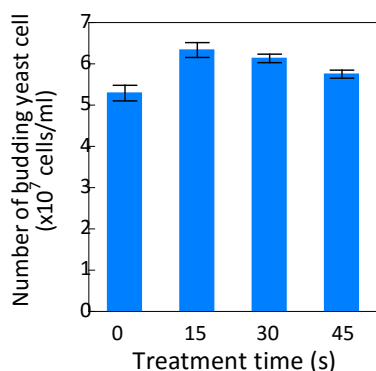
## 3. Results

Using UV absorption spectroscopy, we measured NO and O<sub>3</sub> densities, and decided a gas mixture condition, N<sub>2</sub> (30%)–O<sub>2</sub> (70%) in buffered Ar, to obtain high NO density but low O<sub>3</sub> density (lower than the detection limit). From our calculation, it was measure the NO density to be  $7.27 \times 10^{16} \text{ cm}^{-3}$ . With this known NO density, we obtained 20% increase of number of budding yeast cell for 15 s treatment. While, longer treatment times up to 45 s, the cell counting results also showed large number of yeast cells compare to the untreated ( $t = 0$ , control) but smaller than 15 s.

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## 4. References

- [1] A. Fridman, Plasma Chemistry, Cambridge (2008).
- [2] H. Hashizume *et al* Appl. Phys. Letts. **107** (2015) 093701.
- [3] H. Inui *et al* Appl. Phys. Express **3** (2010) 126101.



**Fig. 1** Number of budding yeast cell was increased about 20% with NO treatment for 15 s. Cells were counted after incubation time of 48 h.