9th Asia-Pacific Conference on Plasma Physics, 21-26 Sep, 2025 at Fukuoka

Molecular Mechanisms underlying Cellular Responses to Plasma Irradiation in Fission Yeast

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In recent years, non-equilibrium atmospheric-pressure plasma has enabled the irradiation to living organisms and has been applied across a wide range of fields, including medicine and agriculture. However, the biological effects of plasma irradiation on living organisms remain largely unknown.

Plasma generates various stress factors, including chemically reactive species such as hydrogen peroxide. Hence, a single irradiation imposes multiple types of stress on cells at the same time. We set out to elucidate the cellular responses to compound stresses induced by plasma irradiation at the gene level by using the fission yeast *Schizosaccharomyces pombe*, a model organism extensively used to study cellular biological processes. Since most living organisms can only survive within a narrow temperature range — yeast cells, for example, proliferate exponentially at around 30°C — precise control of plasma gas temperature is essential for its use in biological studies. To this end, we have developed a plasma irradiation device with precise temperature control, in which helium gas is cooled by a Peltier device prior to discharge [1, 2]. This system can maintain the plasma gas temperature at a constant value of around 30 °C.

Using this device, we applied direct plasma irradiation to fission yeast cells and found that strong plasma irradiation inhibited cell growth. To identify factors involved in the cellular responses to plasma irradiation, we screened for plasma-resistant mutants. We introduced random mutations into wild-type cells and selected mutants that could survive after strong plasma irradiation, which allows little or no growth in wild-type cells. Through this screening, we found that mutants lacking either the *sep1* or *ace2* gene, each of which encodes a transcription factor essential for proper cell separation, were resistant to plasma irradiation (Figure 1A). Microscopic observations revealed that these mutants exhibited a defect in cell separation (Figure 1B).

We also performed RNA-seq analysis to examine gene expression profiles following plasma irradiation. We found that plasma irradiation downregulates the expression of genes that are regulated by the transcription factor Sep1. These results strongly suggest that the Sep1-mediated cell division regulatory pathway plays an important role in the cellular responses to plasma.

RNA-seq analysis also revealed that plasma irradiation downregulates the TORC1 pathway, a central regulator of intracellular nutrient signaling. The TORC1 pathway is crucial for regulating cell growth and proliferation and is conserved from yeast to humans. It is also of great interest due to its involvement in aging and various diseases. Microscopic observations and biochemical analysis confirmed that plasma irradiation reduces TORC1 activity. Altogether, these findings suggest that fission yeast cells respond to plasma irradiation through two distinct pathways: the Sep1-Ace2 cell separation regulatory pathway and the TORC1 nutrient response pathway [3].

References

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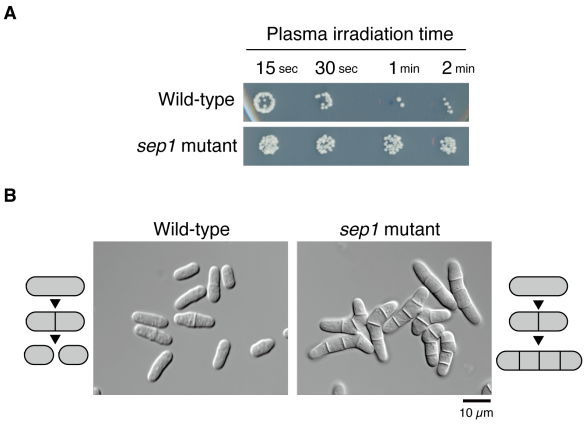

Figure 1. Isolation of plasma-resistant mutant
A. Growth of wild-type and plasma-resistant mutant cells after plasma irradiation. ~1000 cells were spotted on nutrient rich medium, irradiated with plasma for the indicated times and incubated at 30°C.
B. Wild-type and *sep1* mutant cells on nutrient rich medium were observed under DIC microscopy.