

Germination kinetics of plasma-activated seeds

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Global food shortages in the future are a concern. Seed germination and growth enhancement of seeds treated by nonthermal plasma are conceived a potential solution of declining world crop yields.[1] The germination and growth processes were enhanced 11% faster in the air plasma irradiation to seeds, as compared with that of the untreated seeds of *Arabidopsis thaliana*. [2] Early germinating processes, such as water uptake, imbibitional swelling, metabolic activation, seedling germination, and sprouting, are focused on the plasma irradiated seeds. Kinetic behaviors and cellular organic radicals of the seed germination have been studied.

Samples were seeds of *Raphanus sativus* L. The samples were first graded in sizes of 3.0-3.2 and 3.2-3.4 mm using a punched-hole mesh. Then, all the seeds were weighed and sorted according to their weights ranged between 17.05 and 17.30 mg. The graded samples were divided into two groups and each was used to prepare for the untreated and treated seeds. The plasma treatments were typically conducted using the air dielectric barrier discharge (DBD) for 3 min. [2] The seeds were planted on a sheet of filter paper and they were incubated in a chamber at 22°C with both dark and light conditions.

Photographs of the seed germination were kinetically taken at time intervals by the lapse camera [Fig. 1(a,b)]. The morphological information of all seeds was obtained from the acquired images and temporal variations were analyzed by producing histogram [Fig. 1(c,d)]. The seeds first swelled by physical water absorption and germinated after a period of time. The behaviors were parameterized by fitting the results to the Richard curves. The plasma treated seeds accelerated in the germination.

Moreover, the seeds were planted each in one well of 96-well microtiter plates Seed's organic radicals were measured by the electron spin resonance (ESR) method at temperatures below 0°C, preventing dielectric losses for liquid water.

ESR spectra for the seed were measured. Before water imbibition, the ESR signal appeared around g-value of 2 that is assignable to the organic free radicals. The signal changed irreversibly by water absorption. Behaviors of the ESR signals were different between the untreated and the plasma-treated seeds. This indicates probably that metabolic activation increases reactive oxygen species scavenging activity.

In summary, the kinetic behaviors and cellular organic radicals of the germination phase of the plasma-treated seed have been discussed.

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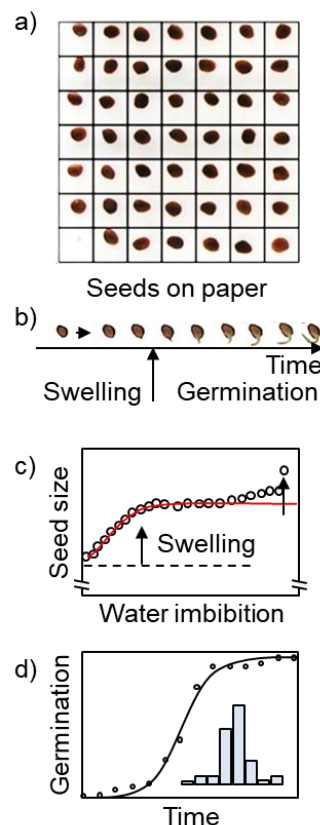


Figure 1. (a) An example of snapshot photographs of the lapsed observation of seed germination. Squares are a guide for extraction of each seed. (b) The time-lapsed morphological changes of one seed. (c) Temporal behavior of seed sizes for swelling induced by physical water absorption at the initial phase. (d) Cumulative germination rates as a function of water imbibition time and (inset) its probability density function. [3]

References

- [1] S. Kitazaki, T. Sarinont, K. Koga, N. Hayashi, and M. Shiratani, *Curr. Appl. Phys.*, 14, S149–S153, (2014).
- [2] K. Koga, T. Sarinont, T. Amano, H. Seo, N. Itagaki, N. Hayashi, and M. Shiratani, *Appl. Phys. Express*, 9, 016201, (2015).
- [3] K. Ishikawa et al., *JSAP-Spring* (2020) 13a-A201-4.