

Bioremediation of Cd polluted soil by two host plants treated with three different species of dark-septate root endophytic fungi originated from the subarctic-subtropical areas.

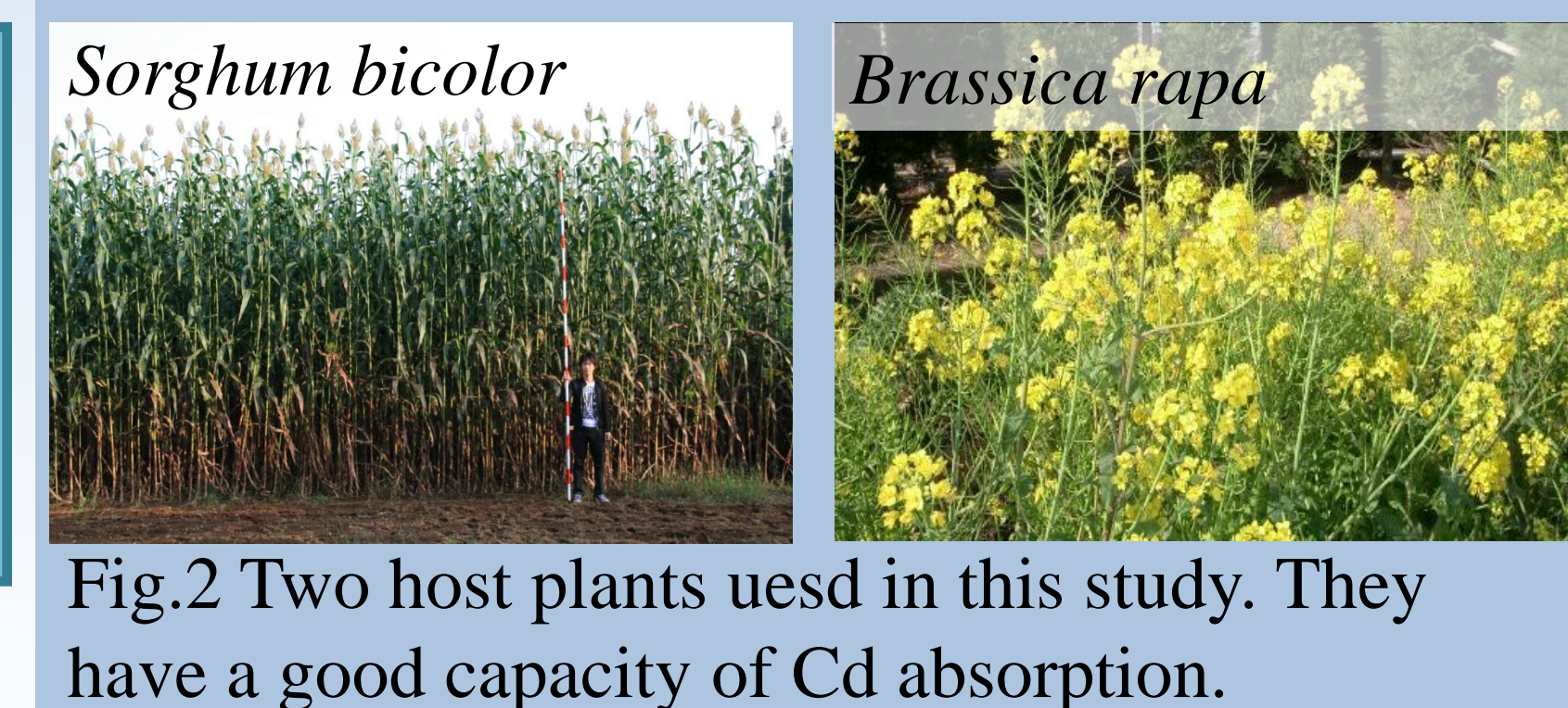
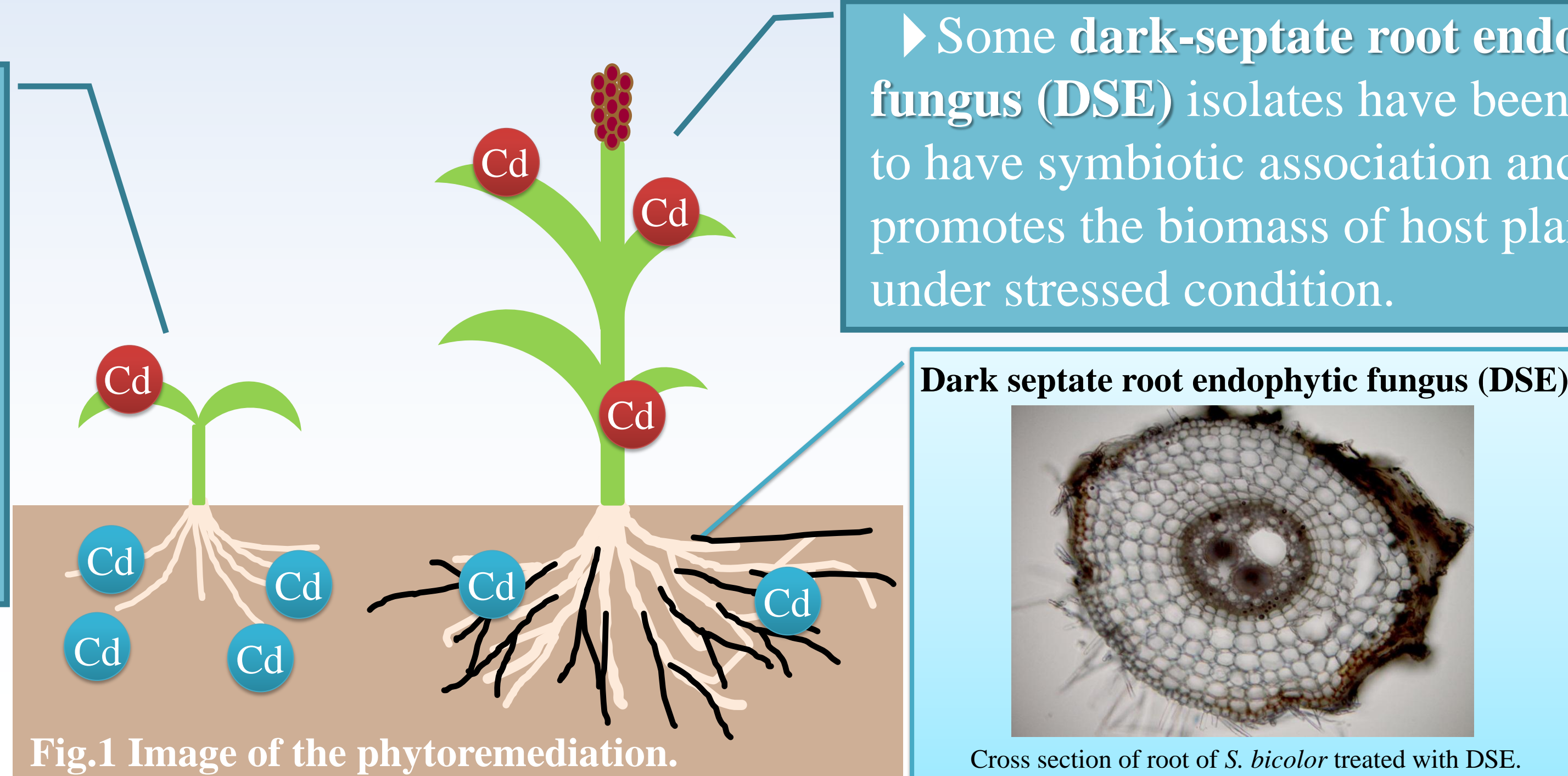
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Introduction

► **Cadmium (Cd)** polluted soil and water are easily discovered in Japan including agricultural land.

► To remove these pollutants, the **phytoremediation** is the most environmental-friendly and low cost method. However, the phytoremediation has not been accepted widely because plant growth is deficient under the stressed condition.

► Some dark-septate root endophytic fungus (DSE) isolates have been known to have symbiotic association and promotes the biomass of host plant even under stressed condition.



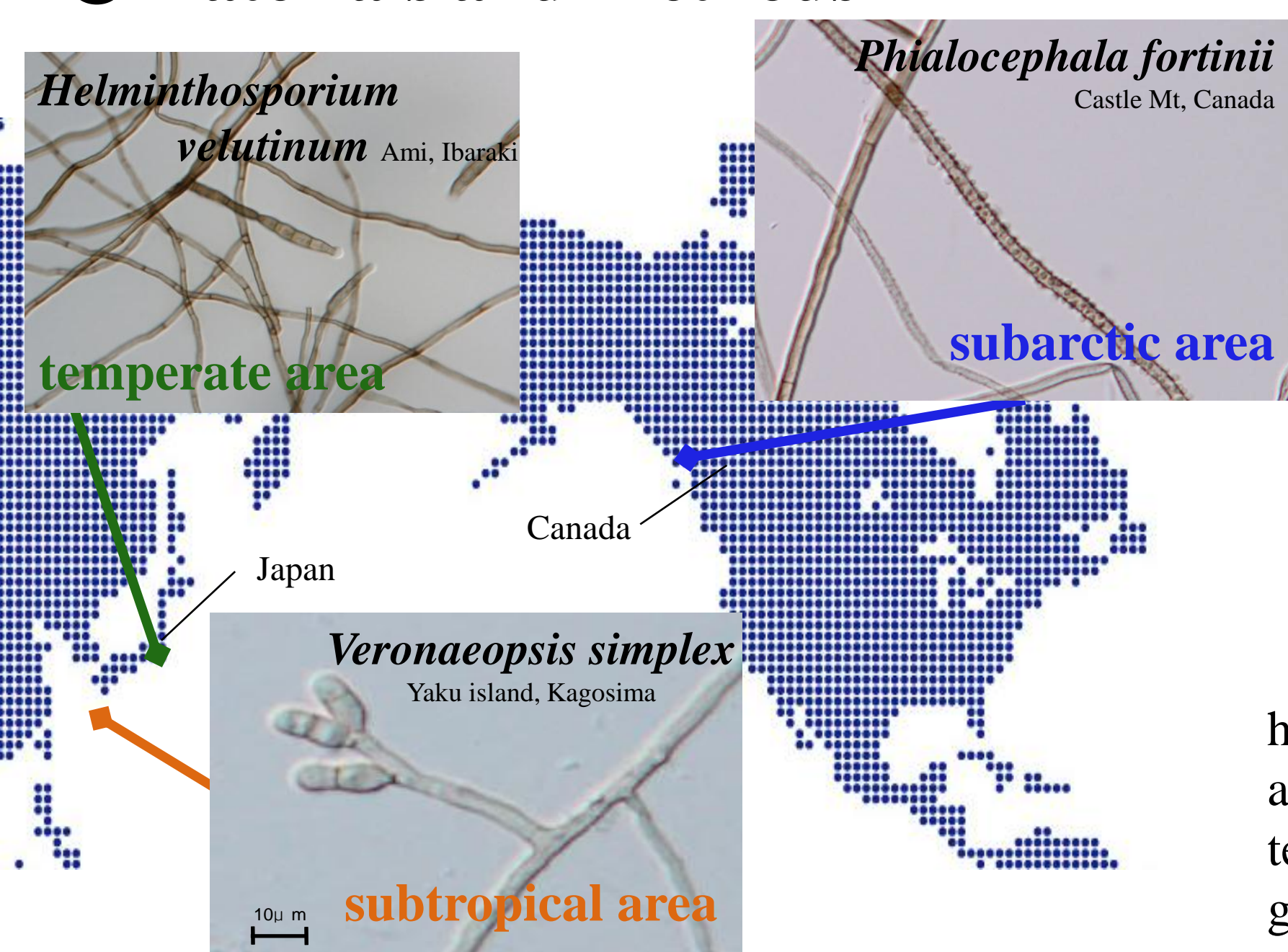
► We previously selected three different species of DSE originated from the **subtropical area** (*Veronaeopsis simplex*), **temperate area** (*Helminthosporium velutinum*) and **subarctic area** (*Phialocephala fortinii*) (Fig.3), which were able to increase the biomass of *Sorghum bicolor* and *Brassica rapa* (Fig.2).

Objective

► In this study, we examined the use of the DSE fungi to solve the problem of phytoremediation for Cd pollution.

1. Comparison with optimum growth temperature of selected DSE isolate

Materials and Methods



Temperature
4°C, 13°C, 23°C, 30°C and 37°C

DSE isolates
V. simplex, *H. velutinum* and *P. fortinii*

Selected DSE isolates were cultured on half-strength cornmeal-malt-yeast extract agar (1/2CMMY)¹ under five different temperatures to determine the optimum growth temperature.

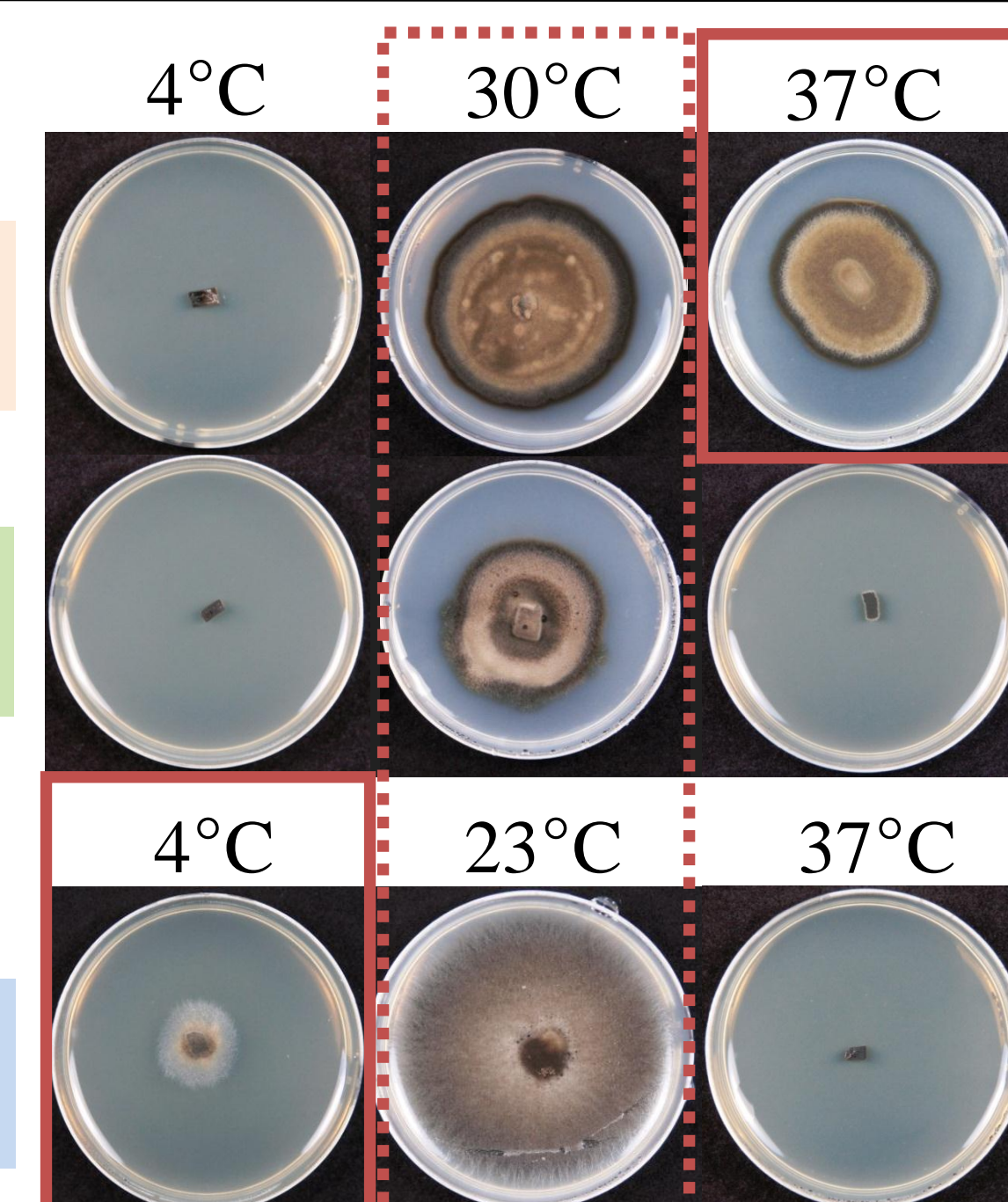
1. Cornmeal 8.5 g, Bact agar 7.5 g, Malt Extract 10.0 g, Yeast Extract 2.0 g L⁻¹

Results

V. simplex

H. velutinum

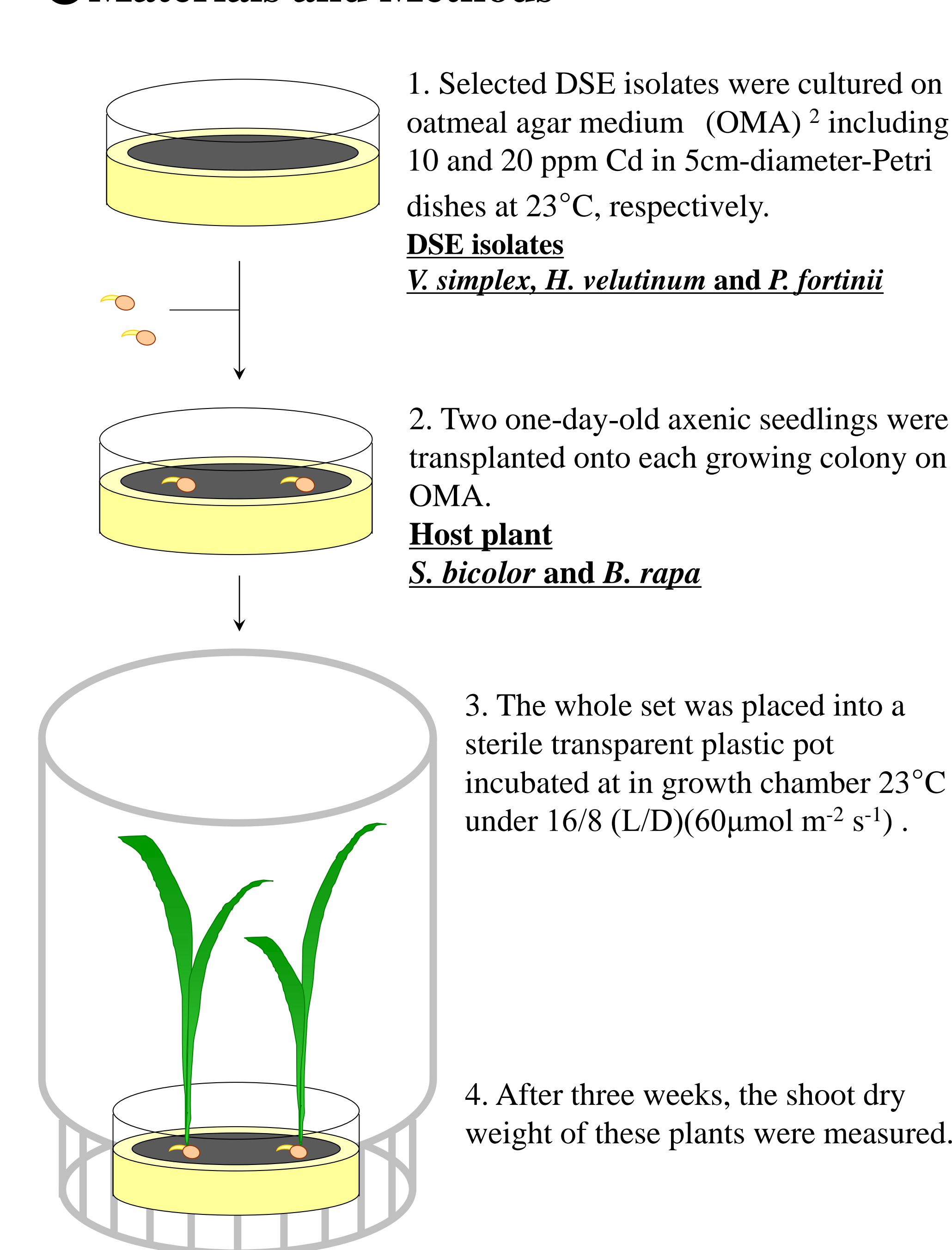
P. fortinii



Optimum temperature for the growth of both *V. simplex* and *H. velutinum* was 30°C (Fig.4), the subtropics originated isolate of *V. simplex* merely could grow under high temperature condition at 37°C (Fig.4). Even though optimum temperature for the growth of *P. fortinii* was 23°C (Fig.4), the subarctic originated isolate of *P. fortinii* could grow at 4°C (Fig.4).

2. Effects of inoculation of DSE isolates on the growth of two host plants seedlings

Materials and Methods



Results

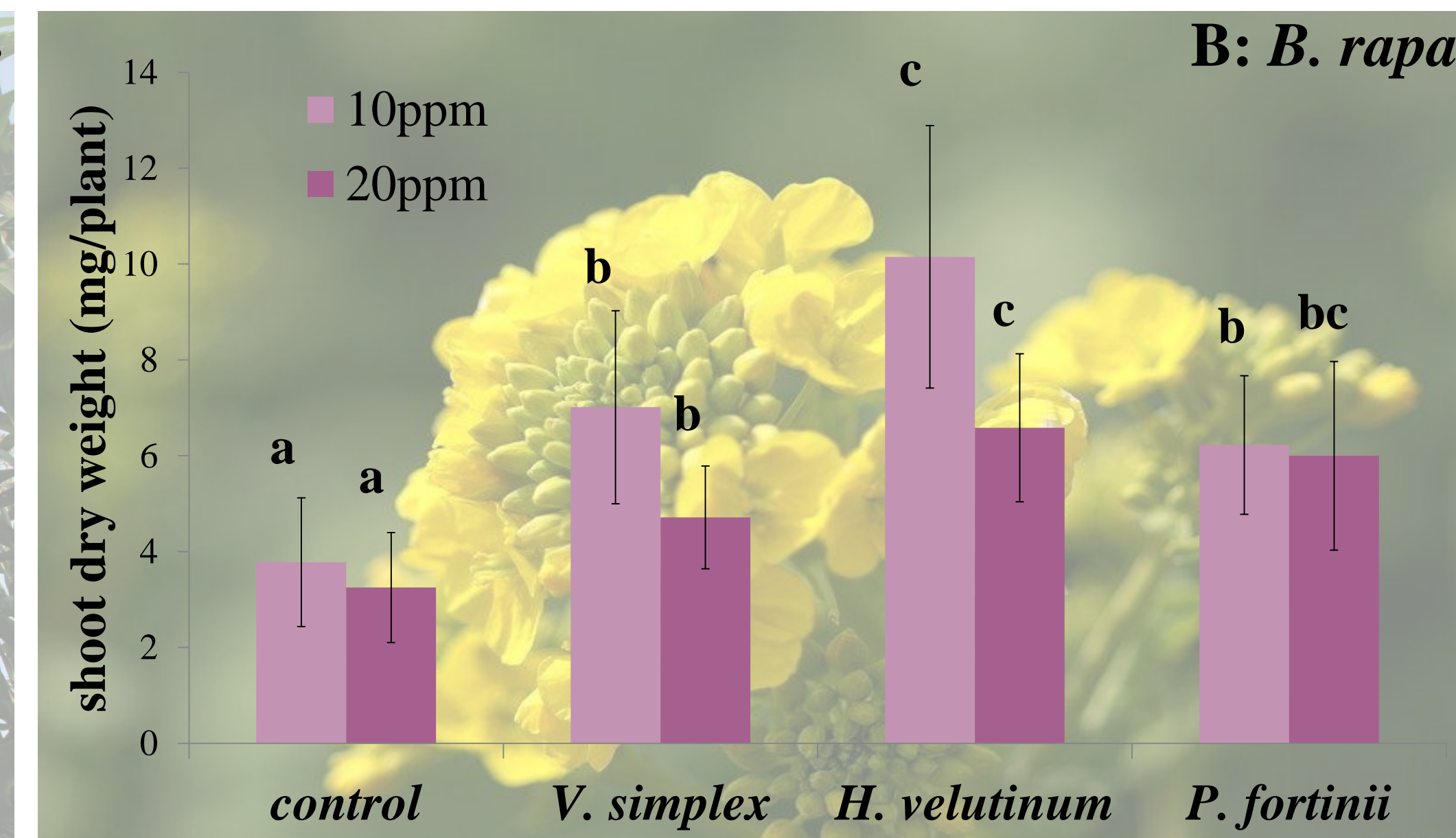
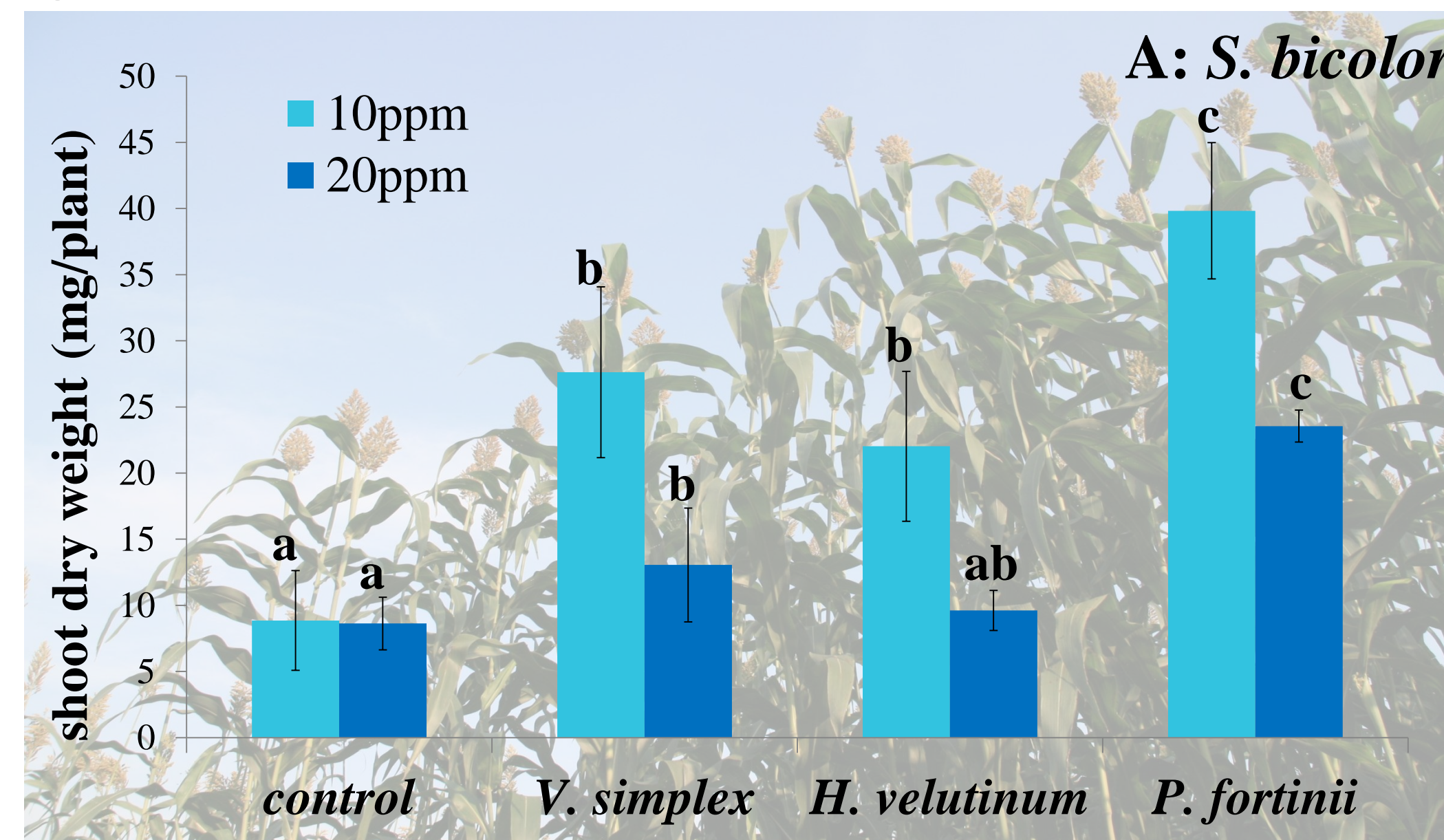
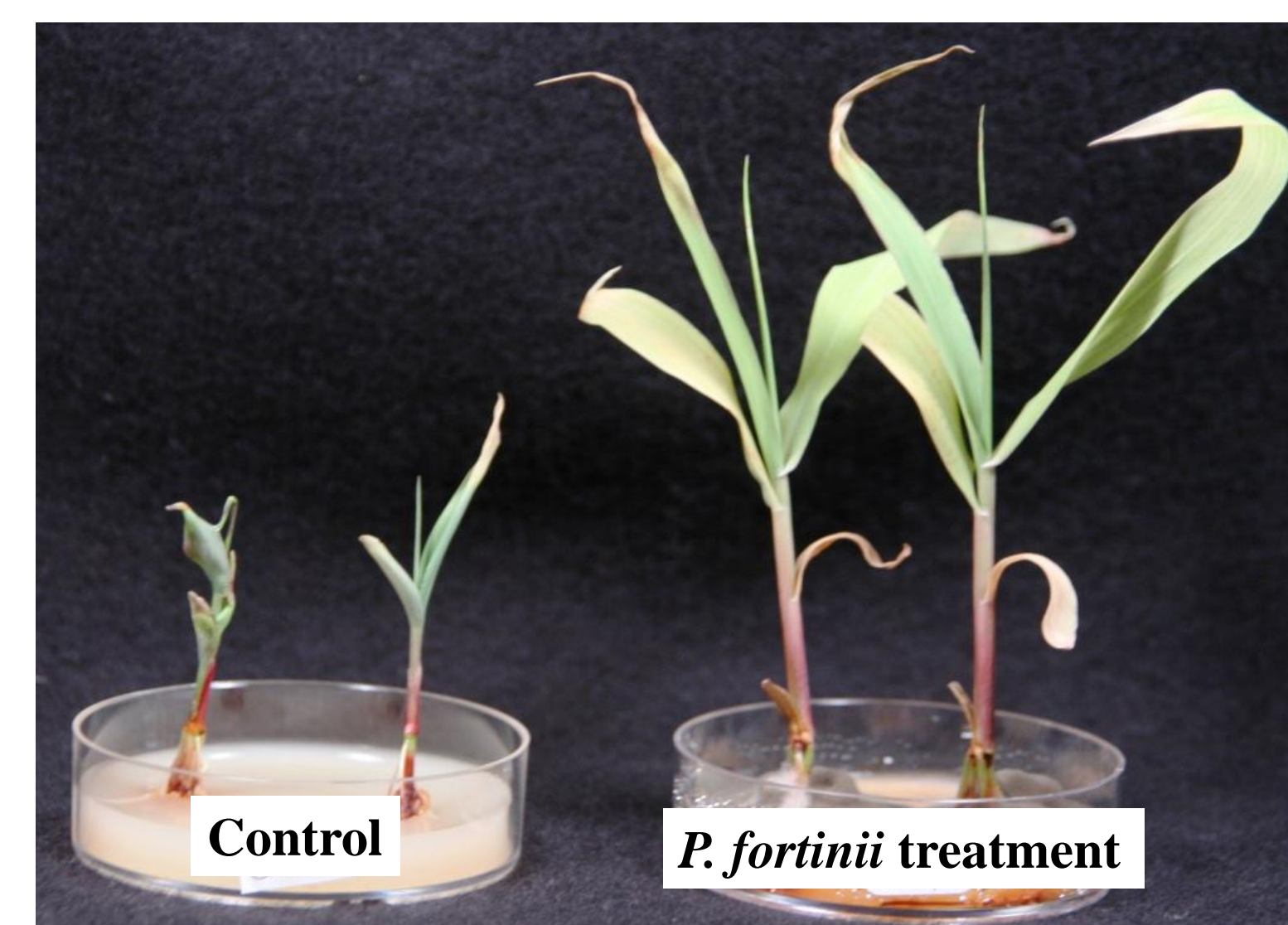


Fig.5 The shoot dry weight (mean ± SD) of host plants treated with DSE isolates after 3 weeks incubation at 23°C and 16 h/8 h (light/dark). A: *S. bicolor*, B: *B. rapa*. Bars represent standard deviation and different letters indicate significant differences range test at $P < 0.05$.



All tested DSE isolate significantly promoted in comparison with control the shoot growth of *S. bicolor* under 10 ppm Cd concentration. Under 20 ppm Cd concentration, *V. simplex* and *P. fortinii* significantly promoted the shoot growth. Among them, especially, the average shoot dry weight treated with *P. fortinii* under 10 ppm and 20 ppm Cd were the highest (Fig.5 A and Fig.6).

In *B. rapa* experiment, all tested DSE isolate significantly promoted the host biomass under both of 10ppm and 20ppm. Especially, the average shoot dry weight treated with *H. velutinum* under 10 ppm and 20 ppm Cd were the highest (Fig.5 B).

Conclusion

► The subtropics origin, *V. simplex*, merely could grow under high temperature condition, in the contrary the subarctic origin, *P. fortinii*, could grow under low temperature condition.

► The isolates of *V. simplex* and *P. fortinii* significantly promoted the shoot growth of *S. bicolor* under 20 ppm Cd concentration *in vitro*. Especially, treatment of *P. fortinii* showed best growth promotion effect on *S. bicolor*. In terms of *B. rapa*, all tested DSE significantly promoted the biomass under both of 10 ppm and 20ppm Cd concentration.