

# Studies of seed dormancy and germination for conserving an endemic and vulnerable species, *Prunus taiwaniana*, in Taiwan

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## 【Abstract】

*Prunus taiwaniana* is an endemic species in Taiwan and is considered to be a vulnerable species on the criteria of the IUCN Red List of threatened species. Seeds of *P. taiwaniana* did not germinate at alternating temperatures of 25/15°C, but slightly germinated at 20/10°C, and germination percentage was 6.7–22.6% after 22 weeks incubation. Cold stratification at 5°C for 8, 12 and 16 weeks increased germination to 15.6, 12.2 and 14.4%, respectively, and the ungerminated seeds remained dormancy. Seeds stratified at 25/15°C for 2 weeks followed by cold stratification at 5°C for 4 weeks significantly increased germination to >80%. Result showed that warm plus cold stratification is an effective method for promoting seed germination of *P. taiwaniana*. Seeds of *P. taiwaniana* that required cold stratification for dormancy break exhibit physiological dormancy.

## Introduction

*Prunus taiwaniana* Hayata (Rosaceae) is a deciduous tree that is sparsely distributed in central Taiwan between 900 and 2400 m elevations and is considered to be a vulnerable species. It is an endemic species in Taiwan and produces white flowers in February–March and dark red mature fruits in April–May, making it a valuable ornamental plant. Freshly harvested seeds of *P. taiwaniana* are dormant at maturity and germinate to low percentage. The purpose of our study was to characterize a method to enhance seed germination of *P. taiwaniana*. Two dormancy-breaking pretreatments were used in the study: one is moist cold stratification at 5°C, whereas the other one is warm stratification at 20/10 or 25/15°C plus cold stratification at 5°C.



## Materials and methods

### Seed collection and preparation

Mature dark red fruits of *P. taiwaniana* were harvested from Wushe and Aowanda, Nantou county, central Taiwan, at elevation of 1100 m in April 2005. Fruits were macerated by hand in water and the empty seeds were floated off and discarded. The clean sunken seeds were used for subsequent pretreatments.

### Moist cold stratification at 5°C

Fresh seeds were mixed with moist sphagnum (small pieces) (water content of the sphagnum was about 400% of its dry mass), sealed inside polyethylene bags and stored at 5°C in darkness for 8, 12, and 16 weeks. The cold-stratified seeds were removed from storage room for germination test at 25/15°C (12/12 h).

### Warm plus cold stratification

Freshly harvested seeds were placed in sealable polyethylene bags with moist sphagnum and sealed and then they were warm-stratified for 2 and 4 weeks at an alternating temperatures regime of 20/10 and 25/15°C in light. The warm-stratified seeds were then given 4, 8, and 12 weeks of cold moist stratification at 5°C.

### Germination tests

Fresh, moist cold and warm plus cold stratification seeds were mixed with moist sphagnum and incubated at alternating temperatures of 20/10 and 25/15°C (12/12 h) with 12 hours of fluorescent light (80–100  $\mu\text{mol m}^{-2} \text{s}^{-1}$ , 400–700 nm) at the higher temperature. Germination, i.e. radicle at least 5 mm long, was recorded weekly for 12 weeks. Results are expressed as germination percentage (%) and as mean germination time (MGT) in days.

### Statistical analysis

Germination percentages (mean  $\pm$  SE) were calculated, and means were compared by analysis of variance (ANOVA) and by least significant difference (LSD) test at the 5% level of significance.

## Results

### Seeds

Freshly harvested seeds of *P. taiwaniana* from Wushe and Aowanda germinated to 20% and 7%, respectively, at 20/10°C, and to below 2% at 25/15°C after 22 weeks incubation (Figure 1). No fresh seeds germinated during the first 5 weeks of incubation.

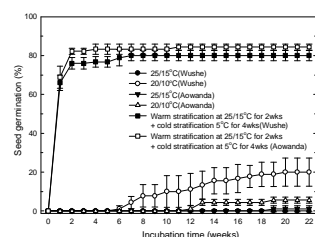


Figure 1. Effect of warm followed by 5°C stratification on the germination of *Prunus taiwaniana* seeds.

### Effect of moist cold stratification at 5°C on germination

Cold stratification at 5°C for 8, 12, and 16 weeks slightly increased germination from 2% to 16% at 25/15°C incubation (Figure 2).

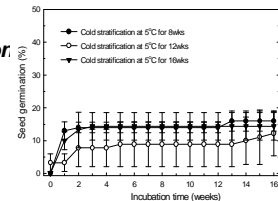


Figure 2. Effect of 5°C stratification on the germination of *Prunus taiwaniana* seeds. Seeds were a mixture of Wushe and Aowanda seeds. The cold-stratified seeds were incubated at 25/15°C.

### Effect of moist warm plus cold stratification on germination

Seeds stratified at 20/10 or 25/15°C for 2 weeks followed by 5°C for 4 or 8 weeks significantly increased germination percentage (Figure 1, Tables 1–3). Mean germination time (MGT) decreased (i.e. germination rate increase) drastically after warm and cold stratification. Meanwhile, we found that germination occurred during the stage of cold stratification for 8 or 12 weeks prior to incubation.

Table 1. Germination percentage of fresh-mature *Prunus taiwaniana* seeds that were collected from Wushe and stratified at 25/15°C followed by 5°C for various periods

Collection site (Wushe)	Fresh seeds	25/15°C 2 weeks +			25/15°C 4 weeks +		
		5°C 4 wks	5°C 8 wks	5°C 12 wks	5°C 4 wks	5°C 8 wks	5°C 12 wks
Germination (%)	0 <sup>b</sup>	80.9 $\pm$ 2.7	77.8 $\pm$ 7.9	74.4 $\pm$ 8.7	73.3 $\pm$ 7.2	72.2 $\pm$ 8.7	81.1 $\pm$ 4.2
MGT (day)	>196	9.7	6.4	3.5	16.4	7.2	0.9

\* Means ( $n = 3$ ) with the same letter do not significantly differ ( $p = 0.05$ ) by LSD test.

Table 2. Germination percentage of fresh-mature *Prunus taiwaniana* seeds that were collected from Aowanda and stratified at 25/15°C followed by 5°C for various periods

Collection site (Aowanda)	Fresh seeds	25/15°C 2 weeks +			25/15°C 4 weeks +		
		5°C 4 wks	5°C 8 wks	5°C 12 wks	5°C 4 wks	5°C 8 wks	5°C 12 wks
Germination (%)	1.1 <sup>d</sup>	84.9 $\pm$ 1.6	98.9 $\pm$ 1.6	91.1 $\pm$ 8.3	83.3 $\pm$ 7.2	93.3 $\pm$ 2.7	95.5 $\pm$ 4.7
MGT (day)	>196	9.3	6.4	0.2	16.5	7.3	1.4

\* Means ( $n = 3$ ) with the same letter do not significantly differ ( $p = 0.05$ ) by LSD test.

Table 3. Germination percentage of fresh-mature *Prunus taiwaniana* seeds that were collected from Aowanda and stratified at 20/10°C followed by 5°C for various periods

Collection site (Aowanda)	Fresh seeds	20/10°C 2 weeks +			20/10°C 4 weeks +		
		5°C 4 wks	5°C 8 wks	5°C 12 wks	5°C 4 wks	5°C 8 wks	5°C 12 wks
Germination (%)	6.7 <sup>d</sup>	88.9 $\pm$ 4.2	98.9 $\pm$ 1.6	91.1 $\pm$ 4.2	95.6 $\pm$ 3.1	95.6 $\pm$ 1.6	88.9 $\pm$ 1.6
MGT (day)	112	15.7	5.3	1.1	19.7	6.8	0.6

\* Means ( $n = 3$ ) with the same letter do not significantly differ ( $p = 0.05$ ) by LSD test.

## Discussion

1. No fresh seeds of *P. taiwaniana* had germinated after 5 weeks incubation. Cold stratification slightly increased seed germination. Warm followed by cold moist stratification completely released dormancy and promoted germination. We concluded that the seeds exhibit physiological dormancy.
2. In the present study, warm stratification given before a cold stratification period caused a significant increase in germination percentage. The moist warm stratification may allow continued seeds development and release the hard endocarp of *Prunus* seed, i.e. covering layers changes.
3. It is possible that the dormancy break is accompanied by a decrease in abscisic acid content of the covering layers and germination by an increase of embryonic gibberellins content.

## References

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