

SEED GERMINATION STUDIES IN *CRYPTOMERIA JAPONICA* D. DON.

S. PANDEY, OM PRAKASH AND P.K. NAGAR*

Division of Biotechnology, Institute of Himalayan Bioresource Technology, Palampur-176061 (H.P.)

Received on 9 Oct., 2002, Revised on 13 Aug., 2003

The present study investigates the germination requirements of the seeds of *Cryptomeria japonica* with respect to temperature, light and gibberellic acid. The study indicates that temperature optima of 22°C, darkness and GA₃ (25 mg l⁻¹) are factors promoting germination of this species. Poor germination at high temperature under both light and dark conditions may be an indication of thermo sensitive nature of this species.

Key words : *Cryptomeria japonica*, germination, gibberellic acid, seeds, temperature

Cryptomeria japonica D. Don (Taxodiaceae) is a fast growing exotic tree (40-45 m tall), used for making tea chests, wooden houses, bridges, furniture, boxes etc. Wood and leaves are also sources of essential oil which is used for incense sticks (Troup 1986). The tree is indigenous to China and Japan and in India it has been successfully grown at elevations from 900 to 2200 m., but appears to thrive best between 1200-1800 m. Seed germination of most plant species are affected by environmental factors, in which temperature plays an important role (Thomson and Grime 1983), with an alternating temperatures resulting in maximum germination in some cases (Kose 1998, Bhattacharya *et al.* 2000). Increased germination of *Lavendula angustifolia* (Aoyama *et al.* 1996) and *Sapindus trifoliatus* seeds (Naidu *et al.* 2000) have resulted from exogenous application of gibberellic acid (GA₃). Seed dormancy has also been broken by applying GA₃ in many crops like lettuce and wheat (Inoue 1991, Lenton and Appleford 1991).

The cones of *C. japonica* ripen in August-September at the lower elevations and somewhat later at the higher elevations due to low temperature. Under natural conditions seeds disperse from cones during December lay dormant on soil and germinate in April-May (when favourable temperatures arrivés), but exhibit poor germination ability (Troup 1986). So far no attention has

been given to study germination behaviour of *C. japonica* seeds. Therefore, in the present investigation, attempts have been made to understand the germination behaviour of *C. japonica* seed using different temperature regimes under both light and dark condition as well as the responses of seeds to different GA₃ treatments.

Mature freshly harvested seeds of *C. japonica* from cones were collected immediately after maturation from North-East region of India during first week of December from Shillong (1496 m above msl, 25.57° N, 91.87° E). For all studies conducted (or otherwise stated), seeds were sown in 9 cm diameter petridishes lined with two layers of Whatman number 1 paper moistened with 3.5 ml distilled water. These were sealed with parafilm to check evaporation and three replicates with ten seeds in each were maintained per treatment. All observations on germination were collected after 8 days and radical protrusion (1-2 mm) was considered as the criterion for germination. Seeds were allowed to germinate in the B.O.D. incubator under control condition, at either 10 h photoperiod of 20 μmol m⁻² s⁻¹, 400-700 nm white fluorescent light (10 h light: 14 h darkness) or complete darkness. The different temperatures regime tested were 18, 22, 26 and 30°C ± 1°C. Seeds were soaked for 1 hr in 3.5 ml each of different concentrations of GA₃, *i.e.*, 5, 10, 25 and 50 mg l⁻¹. For control, seeds were soaked only in

*Corresponding author: e-mail: nagar_pk2001@yahoo.co.uk

distilled water. These were then allowed to germinate both under light and dark at different temperatures as described earlier. Seeds were also subjected to a tri-phenyl tetrazolium chloride test. These were dissected longitudinally into two halves and were immersed in 0.1% tri-phenyl tetrazolium chloride solution in crucible for 24 h in complete darkness at 25°C. The embryos which become completely stained red were counted as viable, the ones, which were not stained were counted as non-viable. The standard errors of means for each treatment were determined statistically.

Maximum germination of *C. japonica* seeds was observed under dark condition as compared to light. Differences in germination under light and dark conditions at different temperature were highest (68.6%) under dark at 22°C followed by 61.3% at 26°C under similar conditions (Fig. 1). However, lower germination was observed under light than dark on entire range of temperature with least at 30°C.

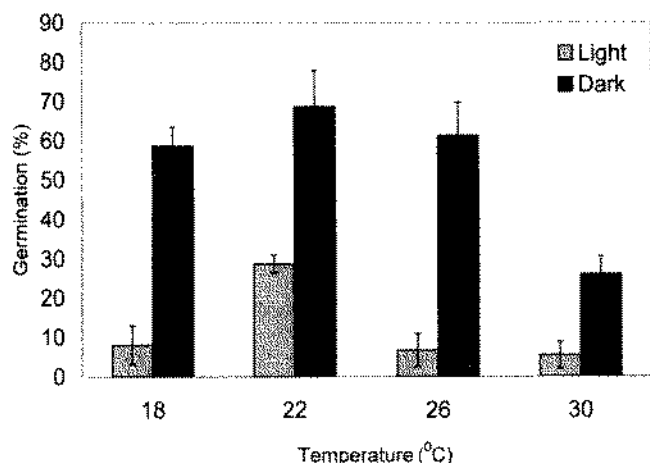


Fig. 1 Temperature response on seed germination of *C. japonica* L. under light and dark conditions. Standard error of means are represented by vertical bars. Data are based on average of all light and dark treatments at each temperature.

Seed germination was mostly higher by GA_3 treatments especially under dark condition. Highest germination (96.67%) was obtained when treated with 25 mg l⁻¹ GA_3 under dark at 22°C temperature (Table 1). Even at higher temperature (26°C), the germination was 83.3% but at 30°C, it decreased to 40%. In light, poor seed germination was observed under entire range of GA_3 treatment (Table 1). Tri-phenyl tetrazolium chloride test

indicated that the viability of freshly harvested seeds collected during December was very high (95%) but declined to 20% after 6-7 months (*i.e.*, by June).

Seeds, representing a precarious stage in a plants life and require a flexible strategy for germination coordinated by environmental signals. As in seeds of many species temperature is one factor which plays a major role in controlling germination (Thomas and Grime 1983, Karssen 1982). Mostly temperature and light will affect the germination behaviour in some way or other. (Thomas 1994). Germination was adversely affected by higher temperature of 30°C under both light and dark conditions (Fig. 1). This may be an indication of themosensitive nature of this species and probably explains why the *Cryptomeria* seeds produced in nature fails to germinate during winter from November to March. In nature, *C. japonica* seeds germinate during the summer months of April-May when the temperature is favourable for germination (Troup 1986). In *Tagetes minuta* high temperature is reported to contribute to the poor germination under hot regions (Drewes and Staden 1990).

In the present study, GA_3 (25 mg l⁻¹) was quite effective for germination of seeds at 22°C, however, beyond this it was ineffective in increasing germination. This indicates the inability of GA_3 in overcoming thermoinhibition. GA_3 was found ineffective to induce higher percentage of germination in *Solanum nigrum* at supraoptimal temperature of 40°C (Gvelberg *et al.* 1984). The seeds of *Florence fennel* germinates better in dark than in the light (Thomas, 1994) and high temperature inhibition was partially overcome by $GA_{4/7}$ indicating that GAs appear to be involved directly in the control of germination of both positively and negatively photo-inhibited seed. It is presumed that GA_3 increases the activation of enzymes or enhanced embryo growth by weakening the endosperm prior to radicle protrusion in *Cryptomeria* as in other seeds (Groot and Karssen 1987). The changes in the sensitivity of seeds to GAs and light may involve changes in the number of receptors, affinity of the receptor and in the signal-transduction pathways initiated by GAs or phytochromes (Weyers *et al.* 1991).

The authors are thankful to Dr. P.S. Ahuja, Director IHBT for providing necessary facilities and Dr. Amita Bhattacharya for critically going through the manuscript.

Table 1. Response of GA₃ and different temperatures on germination of *C. japonica* seeds under light and dark condition. Standard errors of means is represented by \pm and are based on three replications of 10 seeds each.

Treatments GA ₃	18°C		22°C		26°C		30°C	
	Light	Dark	Light	Dark	Light	Dark	Light	Dark
5 mg l ⁻¹	0.00	50.00	23.33	56.67	0.00	53.33	0.00	20.00
	± 0.00	± 1.15	± 3.53	± 7.05	± 0.00	± 2.40	± 0.00	± 1.15
10 mg l ⁻¹	0.00	53.33	26.67	66.67	0.00	56.67	0.00	26.67
	± 0.00	± 4.40	± 5.69	± 2.02	± 0.00	± 3.53	± 0.00	± 2.40
25 mg l ⁻¹	23.33	73.33	36.67	96.67	20.00	83.33	16.67	40.00
	± 3.33	± 3.52	± 4.05	± 2.40	± 1.15	± 2.40	± 0.88	± 2.65
50 mg l ⁻¹	16.67	66.67	30.00	80.00	13.33	76.67	10.00	30.00
	± 3.33	± 5.69	± 1.53	± 5.77	± 0.88	± 5.70	± 5.03	± 2.00
Control	0.00	50.00	26.67	43.33	0.00	36.67	0.00	13.33
	± 0.00	± 3.21	± 6.76	± 4.80	± 0.00	± 0.88	± 0.00	± 2.40

REFERENCES

- Aoyama, E.M., Ono, E.O. and Furlan, M.R. (1996). Germination study of lavender seeds (*Lavandula angustifolia* Miller). *Scientia-Agricola*. **53**: 267-272.
- Bhattacharya, A., Nagar, P.K. and Ahuja, P.S. (2000). Seed germination of *Rumex hastatus* D. Don. *Seed Sci. & Tech.* **28**: 67-74.
- Drewes, F.E. and Staden, J.V. (1990). Germination of *Tagetes minuta* L. II Role of Gibberellins. *Plant Growth Reg.* **9**: 285-291.
- Gvelberg, A., Horowitz, M. and Polja, K.M.A. (1984). Germination behaviour of *Solanum nigrum* seeds. *J. Exp. Bot.* **35**: 588-598.
- Groot, S.P. and Karssen, C.M. (1987). Gibberellins regulate seed germination in tomato by endosperm weakening: a study with gibberellin—deficient mutants. *Planta* **171**: 525-531.
- Inoue, Y. (1991). Role of Gibberellins in phytochrome mediated lettuce seed germination. In: N. Takahashi, B.O. Phinney and J. MacMillan (eds.), *Gibberellins*, pp. 199-210. Springer-Verlag, New York.
- Karssen, C.M. (1982). Seasonal patterns of dormancy in weed seeds. In: A.A. Khan (ed.). *The Physiology and Biochemistry of Seed Development, Dormancy and Germination*, pp. 243-270. Elsevier Biomedical Press, Amsterdam.
- Kose, H. (1998). Studies on the germination of some woody ornamental plants in the Turkish flora, *Arbutus unedo* L. and *Arbutus andrachne* L. *Anadolu*. **8**: 55-65.
- Lenton, J.R. and Appleford, E.J. (1991). Gibberellin production and action during germination of wheat. In: N. Takahashi, B.O. Phinney and J. MacMillan (eds.) *Gibberellins*, pp. 125-135. Springer-Verlag, New York.
- Naidu, C.V., Rajendrudu, G. and Swamy, P.M. (2000). Effect of plant growth regulators on seed germination of *Sapindus trifoliants* Vahl. *Seed Sci. Tech.* **28**: 249-252.
- Thomas, T.H. (1994). Responses of Florence fennel (*Foeniculum vulgare azoricum*) seeds to light, temperature and gibberellins A_{10T}. *Plant Growth Reg.* **14**: 139-143.
- Thomson, K. and Grime J.P. (1983). A comparative study of germination responses to diurnally fluctuating temperatures. *J. Applied Ecology* **20**: 141-156.
- Troup, R.S. (1986). *The Silviculture of Indian Trees* Vol. III. Oxford University Press, London.
- Weyers, J.D.B., Patterson, N.W. and A' Brook R. (1987). Towards a quantitative definition of plant hormone sensitivity. *Plant, Cell Environ.* **10**: 1-10.