# EFFECTS OF PRE-TREATMENT OF GIBBERELLIC ACID SOLUTION ON MUSA SIKKIMENSIS SEEDS

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#### ABSTRACT

Musa sikkimensis Kurz (Musaceae) is one of the most popular banana species in the western world. It is near-hardy, surviving temperatures down to 0°C, but its propagation and cultivation are little-researched. This study investigates the effects of gibberellic acid treatment on germination of Musa sikkimensis seeds. Enhanced germination rates will offer nurseries, botanic gardens and amenity growers the opportunity to increase their cultivation success. The findings are compared with those of other Musa species.

## BACKGROUND INFORMATION

Musa sikkimensis has a western to central spread in the Asian region throughout Bhutan, Nepal and India, in particular Sikkim, West Bengal and Mizoram. It is a perennial, clump-forming, rhizomatous mega-herb that produces suckers. The pseudostem grows up to 8m with arching oblong lime-green leaves to 60cm with red stripes at young age on the petioles. The inflorescence has a horizontal peduncle, with dark-violet bracts and cream-white fertile flowers. The fruits are yellow and banana-like, containing hard seeds. It thrives in warm conditions at 10–30°C in full sun in a very fertile light soil and large amounts of water. In botanic gardens throughout the world Musa sikkimensis is a magnificent perennial plant which represents the diversity of the family Musaceae as well as forming a very useful contribution to tropical displays in gardens (Fig. 1). Throughout the Musaceae it is difficult to predict what triggers germination because of the lack of research into the family.

## LITERATURE REVIEW

Musa sikkimensis is native to the foothills of the Indian Himalayas and has been grown in amenity horticulture in Britain from seed since it was collected by Wilhelm Sulpiz and described in 1878 (Singh et al., 2016). This species is particularly renowned by the popular striped-leaved Musa sikkimensis 'Red Tiger'. In temperate areas, the only seed available is of unknown provenance and age and this is a problem for growers, because it results in unreliable germination. Many close relatives of the species are commercially

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Fig. 1 *Musa sikkimensis* in glasshouse displays at RBGE. Photo: William Purdom.

valuable, for example *M. basjoo* and *M.* 'Cavendishii' (*M. acuminata*) and these have succumbed to new diseases. Therefore this species offers potential beyond its value as a relatively hardy ornamental species as a source of genetic material with which to strengthen these commercially valuable varieties. It has been known to hybridise readily with edible cultivars, such as the one parent of the Cavendish cultivar. Indeed, 'Helen's Hybrid', a cross between *M.* 'Chini Champa' and *M. sikkimensis*, is still grown and eaten in the foothills of the Himalaya and throughout Bangladesh (Mahdi *et al.*, 2014). Consequently, improved understanding of germination would be valuable for both amenity horticulture and food security.

There is a distinct lack of research on this species, although *Musa velutina*, a suspected relative, which belongs to the same x = 11 chromosome group (Li *et al.*, 2010), has been well studied. *M. velutina* is native to the same regions of India as *M. sikkimensis*, and is popular in tropical horticulture. At the University of Reading, a test was conducted in which fresh fruits from plants at the Royal Botanic Gardens, Kew (RBG, Kew) were harvested (Pancholi *et al.*, 1995). Half were sown immediately and half were stored at room temperature (18–23 $^{\circ}$ C) for one, two, three, seven, nine and ten months. At the same time an embryo germination test was conducted that used gibber-

ellic acid (GA) treatments to test germination success under different regimes. Seed germination of 70–85 per cent was recorded by month 10 in two flushes at months 4 and 9. The seed embryo germination over 14 days showed an average of 73.3 per cent germination with the optimum amount of GA at 0.1 µm in the dark conditions and 69.9 per cent germination with the optimum amount of GA at 0.1 µm in light conditions (Pancholi *et al.*, 1995). Seeds stored at room temperature for one year showed no germination.

In further research by a group from Osaka, Japan, seeds were collected from ripe fruit and stored in vermiculite at 25°C for zero, one, two and four weeks. After each of the periods the seeds were placed into bags of moist media to encourage the embryo to 'break'. They were then sown at a minimum of 10°C. Results showed that the seeds that were not stored germinated within a week. In correlation with prior experiments, this experiment concluded that *Musa velutina* loses viability after one week of storage unless conditions are moist (Nagano *et al.*, 2009). Clearly seed storage times are important in *M. velutina*.

Earlier research into germination by Nagano *et al.* (2008) has investigated the importance of temperature and storage. Fresh seeds from *Musa velutina* plants were sown on wet tissue at temperatures of 10, 15, 20, 25 and 30°C for 12 hours with a night-time fluctuation of 10–20°C, 15–25°C and 20–30°C. Embryos were also removed from the seeds for embryo germination. In contrast to studies carried out by Pancholi *et al.* (1995), no germination occurred among most of the treatments. Where germination did occur, it was in the temperature range of 20–30°C but this had low yields of only 6.7 per cent (Nagano *et al.*, 2008). It was suggested that seed had been harvested too early, and that seeds require a period in moist substrate to complete embryo maturity. This was supported by the study, in which fresh seed stored in moist compost for one to four months at air temperatures of 10–30°C produced germination successes of 80 per cent or above (Nagano *et al.*, 2008).

Interestingly, the storage period given in the Seed Information Database at Wakehurst Place suggests storage times of up to two years dry-stored (Abdelnour-Esquivel *et al.*, 1992). However, most growers will not be aware of the seed storage history or requirements when they obtain seeds.

Gibberellic acid has been used in other investigations into *Musa velutina*. Nagano *et al.* (2010) tried the following pre-treatments: scarification, control and GA. Seeds were collected from cultivated stock and subjected to combinations of scarification or not, and GA concentrations of 0, 1 or 10mg, then kept for 14 days at 25°C. By day 14, 76.7 per cent of the scarified seeds had germinated (mean 7.4 days to germination). None of the control had germinated and it was suspected that this was due to the embryos being immature. The application of 10mg GA after 14 days had the highest germination percentage of 100 per cent. The application of 1mg GA had a germination success rate of 97.7 per cent and with no GA 73.3 per cent germinated. Scarification sped up the germination time and GA application increased the yield from the seeds (Nagano *et al.*, 2010).

It is apparent that temperature, scarification, GA and freshness along with maturity are important factors in the germination of *Musa velutina*. The last of these is perhaps

the key to reliable germination. Unfortunately for growers obtaining seed from suppliers, there is no reliable way to tell how long or under what conditions seed has been stored, and therefore optimising the other factors becomes particularly important.

In *Musa sikkimensis*, the problem is compounded as there is no standing research which directly tested germination of this species. In order to rectify this, the authors carried out a series of germination tests with varying concentrations of GA.

## MATERIALS AND METHODS

Garden-origin seed of *Musa sikkimensis* was acquired from a reputable UK supplier, Jungle Seeds, and GA from Sigmalbrich (Jungle Seeds, 2017). The storage period of these was unknown. In the experiment 250 seeds were split into 5 lots, and soaked for 24 hours at 0, 0.125, 0.25, 0.5 and 1g of GA.

Seeds were sown 2cm deep in seed trays of John Innes seed compost. They were watered until the growing media was moist and placed in a greenhouse at a temperature set to 18–22°C. Results were recorded daily over 60 days (Fig. 2).



Fig. 2 *Musa sikkimensis* seedlings in trays. Photo: William Purdom.

## RESULTS AND DISCUSSION

Results are summarised in Fig. 3 and Table 1. First germination occurred in seeds with 0g GA treatment, after 22 days. This treatment produced eight seedlings. The batch treated with 0.125g produced 13 seedlings in total although they came up later than the first germination. The same results were obtained for 0.25g of GA treatment. Interestingly 1g GA treatment produced the most seedlings – 18 – but this batch was the last to germinate. These results can be compared to the existing research on *Musa velutina* (Nagano *et al.*, 2010) in which 1g of GA also produced the highest number of germinations (97%). The seed used by Nagano *et al.* was fresh rather than dried as in our experiment and the overall number of germinations was a lot higher.

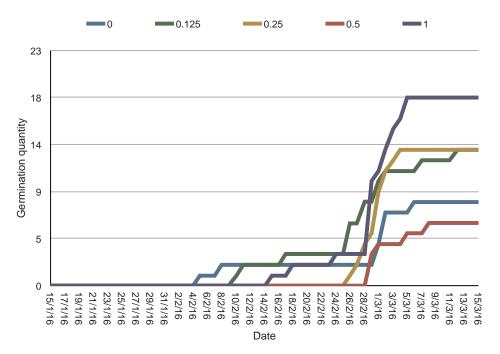


Fig. 3 Germination of Musa sikkimensis seeds with relative GA treatment.

GA (g/L)	Number of germinations	%
0	8	16%
0.125	13	26%
0.25	13	26%
0.5	6	12%
1	18	36%

Table 1 GA treatments and corresponding number of germinations in Musa sikkimensis.

## CONCLUSION

From the current data, we can conclude that GA does enhance the germination of *Musa sikkimensis* seeds, with the optimum amount being 1g/L. This is also what was found by Nagano *et al.* (2010) for *M. velutina*. The authors would recommend treatment of *M. sikkimensis* seeds with this level of GA to enhance germination success, particularly where the length and type of storage post-harvest is unknown. Further studies to investigate optimum storage conditions for *M. sikkimensis* are required, and could prove particularly interesting in light of the similar distribution and ecology between this species and *M. velutina*.

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