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STUDIES ON SEED GERMINATION AND INTRODUCTION OF *DUBOISIA MYOPOROIDES* R. Br.

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Optimum conditions for breaking the dormancy and storage of *Duboisia myoporoides* R.Br. seeds were studied. Gibberellin treatment at 100, 200 and 300 ppm gave positive responses. The treatment time with these concentrations was 24 hours. The effect of light on germination was negligible.

Key words: *Doboisia myoporoides*, Seed dormancy, Gibberellic acid.

Introduction

Recent experiments have shown that *Duboisia liechhardtii* F. Muell. native to Australia and a source plant of scopolamine and hyoscyamine can be cultivated successfully at Karachi [1]. The genus *Duboisia* is represented by three species viz. *D. leichhardtii* F. Muell, *D. myoporoides* R. Br. and *D. hopwoodii* F. Muell. Brown first collected *Duboisia myoporoides* B. Br. during 1802-05 and named the genus *Duboisia* in honour of French Botanist " Dubois " in 1810. He described *D. myoporoides* in his prodromus of 1810 [2]. The leaves of *D. myoporoides* contain 2-4 % of total alkaloid with more than 30% of hyoscyamine and 60 % hyoscine which are the main source of tropane alkaloids in the world today [3,4].

The species of *Datura* and *Physochlaina praealta* (Decne.) Miers had been the main source of these alkaloids. Seeds of *D. innoxia* Mills Contain 0.2–0.3% of total alkaloids (0.1% hyoscine) and leaves of *D. metel* L. which contain 0.4–0.5% of total alkaloid (0.25% hyoscine), were the source of hyoscine. Similarly, the leaves of *Physochlaina praealta* contain 0.6-0.7% alkaloids and *Datura stramonium* L. has only 0.3–0.8% hyoscyamine.

Due to its high alkaloidal contents and its tolerance to water logging, *D. myoporoides* has been introduced and cultivated for the first time in Pakistan at PCSIR Laboratories Complex, Karachi.

The drugs contained in Duboisia leaf are toxic. Most people who handle the crop are affected. A lot of dust (consisting of leaf particles containing high drug levels) is generated when dried leaves are handled and crushed. Drugs are readily absorbed from this dust through the eyes and inhalation as well as through the skin, especially when perspiring [5-7].

D. myoporoides (Fig. 1) is a shurb or a small peach-like tree and is extensively cultivated commercially in the South Burnett region of Queensland. The seeds were imported from Royal Botanic Garden, Australia. They are very small and difficult to germinate under ordinary conditions. They were specially treated with gibberellic acid before planting into seed beds, which were sterilized with fungicides and insecticides.

The experiments were designed to investigate the dormancy factors of *D. myoporoides* seeds, with a view to practical application.

Materials and Methods

Germination on filter paper: Four replicates each of 50 seeds were arranged on whatman No. 4 moist filter paper in 10 cm. petri dishes in the laboratory at different temperatures, e.g. 20,25,30,35°. The average time required for seed germination without any treatment was between 38-42 days with a standard error of 2. On 38th day only 2 seeds germinated

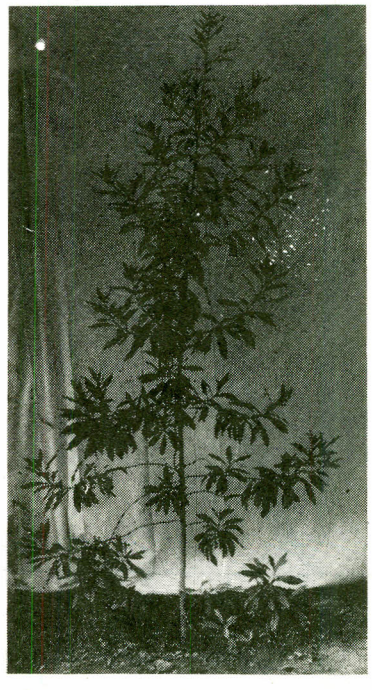

Fig 1. A plant of D. Myoporoides R. Bs. Grown in the experimental plot of PCSIR Laboratories Complex, Karachi

TABLE 1. OPTIMUM TEMPERATURE FOR GERMINATION (WITH OUT ANY TREATMENT)

Temperature (°C)	20	25	30	35
Starting days of germination	–	–	40	–
Germination %	–	–	2	–
Average no. of days required for germination	–	–	40±2	–
Fisnishing day of germination	–	-	42	–

and survived for a period of 5 days at 30° (Table 1).

Effect of gibberellic acid on germination: Before sowing, the seeds were treated with gibberellic acid for 24 hours in 100, 200 and 300 ppm concentrations at a temperature of approximately 40°. The temperature was maintained at a constant level by placing the container with the seeds in gibberellic acid in a water bath already brought to 40 °.

After treatment the seeds were washed thoroughly with clean water to ensure that the seed-coat was free of gibberellic acid. Excess of gibberellic acid causes elongation of stem at the time of germination. The promotion of germination by gibberellic acid had been reported for other hard-seeded plants [8]. The seed germination was recorded at 30°. The seeds were dried and stored for 1 and $1\frac{1}{2}$ month. Without this storage, the percentage of germination was found to be very poor. The treated seeds were kept in air-tight cellophane bags at 20° with 30% relative humidity in the incubator. The cumulative data of germination is incorporated in Fig 2..

Effect of light on germination. A set of 4 replicates of 50 seeds each was used to record the effect of light and gibberellic acid treatment on germination (Table 2). The seeds used in the experiments were stored for $1\frac{1}{2}$ month after the treatment with gibberellic acid. Following the above treatment all the replicates were left under a light of 400 ft candles from two pairs of mercury vapour tubes for a fixed duration of 8 hrs.

TABLE 2. EFFECT OF LIGHT AND GIBBERELLIC ACID TREATMENT ON GERMINATION

Treatment (G.A)	Light			Darkness		
Gibberellic acidn conc. (ppm)	100	200	300	100	200	300
Germination %	40	45	40	40	48	43
Starting day of germination	10	9	8	8	7	9
Finishing day of germination	40	41	40	39	40	40

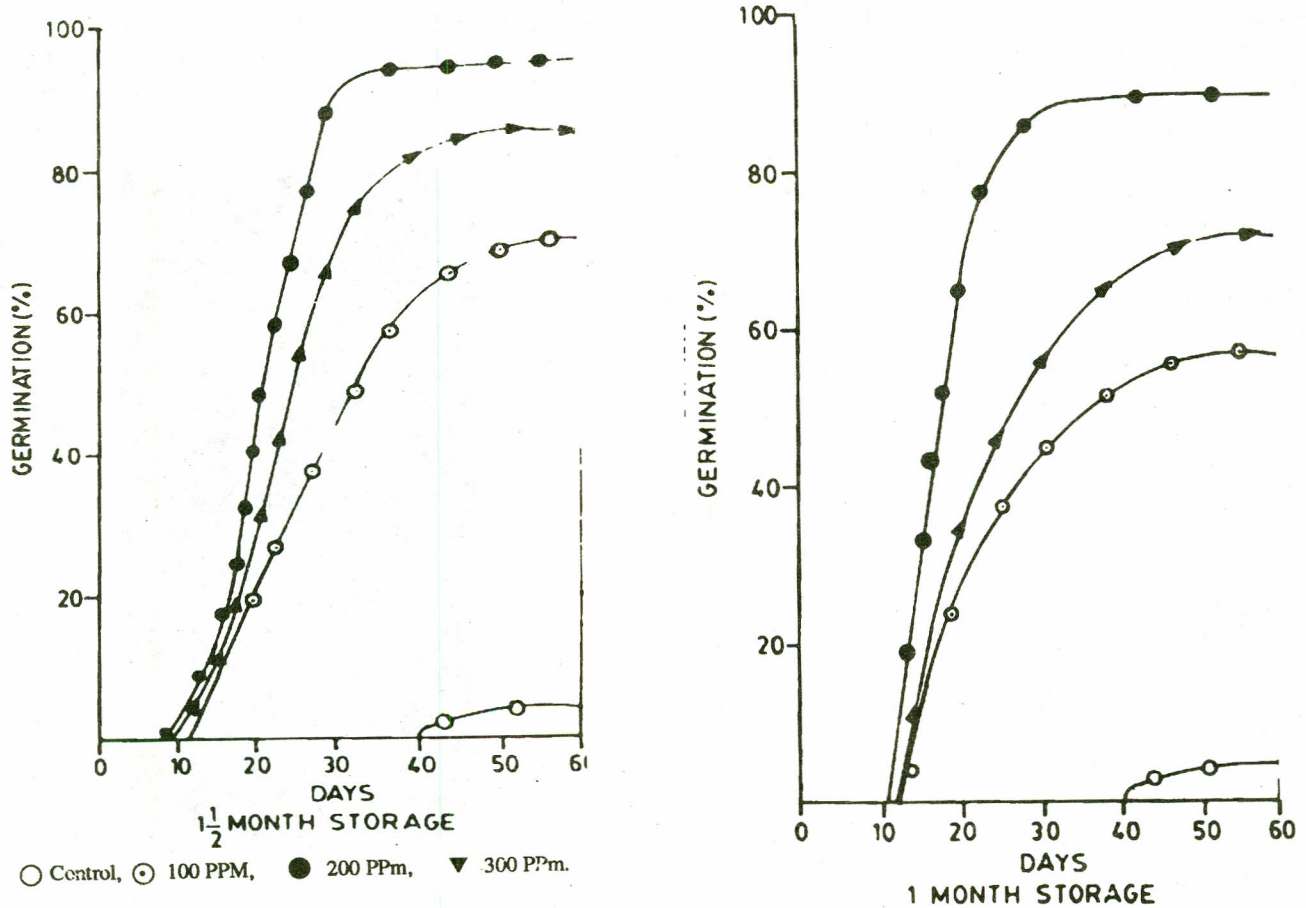

Fig 2. Change in germination with respect to methods and period of seed storage.

A second set of replicates was kept in the total darkness keeping in view that all the parameters such as air, temperature and humidity were the same as in the first experiment. The temperature in both the experiments varied from 28-30° with relative humidity of 60%.

Best results were obtained with seedling raised from gibberellic acid treated seed grown in 60% shade [9]. They were not planted out until the pigmentation of the stem was evident. To avoid nitrogen difficiency, ammonium nitrate was used with the ratio of 5 grams of mixture in 9 litres of water at weekly intervals.

Transplantation of the seedlings was carried out from the earthen pots to the sterilized soil bed after they attained a height of about 10 inches. The month of March was found favourable. The seedlings were planted at the same depth as they were in rursery, because they are subjected to crown rot. The roots of *Duboisia* are very tender and fine and if they were damage at the time of transplantation they are subjected to infection by crown rot organisms.

Results and Discussion

Although the untreated seeds were kept at different temperatures, viz. 20°, 25°, 30°, and 35° hardly any germination was observed as shown in Table 1. Only 2 seeds out of 200 germinated at 30°, but even these died soon after germination.

Germination percentage was slightly higher when the seeds were kept in the dark as compared to that when placed in light (Table 2.) e.g. the percentage of germination at 200 ppm of gibberellic acid treated seeds was 48% whereas those kept in the light it was 45%. Similarly at 300 ppm it was 43% and 40% respectively [10]. This shows that darkness favours germination marginally.

The comparative variation in the time requirement for germination of seeds treated for 24 hrs. with 100,200,300 ppm gibberellic acid subsequently stored for 1 and 1 1/2 months is represented by Fig 2. Combination of 200 ppm gibberellic acid treatment and 11/2 month storage had given the best results (Table 2). In control, the seeds whether stored for 1 or 1 1/2 month, they had germinated poorly. Precisely, gibberellic acid can significantly accelerate seed germination and bread the dormancy of seeds, and it was found concentration dependant [11].

In conclusion the results could be summarized as follows: (a) The percentage of germination increased slightly in darkness; (b) The treatment of *D. myoporoides* seeds with gibberellic acid was essential for germination; and (c) Storage of 1 1/2 month gave better results as compared to 1 month storage.

The 200 ppm of gibberellic acid treatment in combination with 1 1/2 month storage gave the maximum percentage of germination. This combination is therefore recommended for all practical purposes regarding germination of *D. myoporoides* seeds.

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