

## EFFECT OF BRASSINOSTEROIDS ON ABA-INDUCED GERMINATION INHIBITION

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Effect of 28-homobrassinolide and 24-epibrassinolide on ABA - induced inhibition of germination and seedling growth was studied. Both the brassinosteroids reversed the inhibitory effect of ABA.

**Keywords:** Abscisic acid; Brassinosteroids; Germination; Seedling growth.

### Introduction

Brassinosteroids are novel type of growth promoting substances<sup>1</sup>. As a result of extensive investigations, brassinosteroids were found to show characteristic physiological action on growth of plants in microquantities<sup>2</sup>. In an earlier study brassinosteroids were found to stimulate seed germination<sup>3</sup>. In the present study the effect of brassinosteroids on abscisic acid induced inhibition of germination and seedling growth is being investigated.

### Materials and Methods

28-Homobrassinolide and 24-epibrassinolide were purchased from M/s Beak Consultants Inc. Brampton, Ontario, Canada. Seeds of *Trigonella foenum-graecum* Linn. were surface sterilized with 0.1% (W/V) mercuric chloride and washed thoroughly several times with sterile distilled water. Twenty five seeds were distributed in each petriplate (10 cm diameter) provided with Whatman No. 1 filter paper. Each plate contained 5 ml of either of the test solution distilled water;  $0.5 \times 10^{-4}$  M ABA;  $10^{-4}$  M ABA;  $0.5 \times 10^{-4}$  M ABA along with  $1 \mu$  M/3  $\mu$  M brassinosteroids;  $10^{-4}$  M ABA along with  $1 \mu$  M/3  $\mu$  M brassinosteroids. The plates were kept in dark room whose temperature was maintained at  $25 \pm 1^\circ\text{C}$ .

Emergence of radicle was taken as the criteria of seed germination. Germination counts were recorded at the end of 24 and 36 hours. After 48 hours only 5 seedlings were retained in each petriplate and 3 ml more of test solution was added. On the 6th day (from the start of the experiment) seedling length and fresh weight were recorded. The seedlings were dried in oven at  $110^\circ\text{C}$  for 24 hours and dry weight were recorded.

### Results and Discussion

Effect of brassinosteroids on the ABA-induced inhibition of seed germination and seedling growth is shown in Table 1. Brassinosteroids acted as germination promoters and reversed the inhibitory effect of ABA by enhancing the percentage of seed germination. Homobrassinolide at  $3 \mu\text{M}$  concentration was found to be most effective in decreasing the inhibitory nature of ABA.

Brassinosteroids stimulated the growth of *T. foenum-graecum* seedlings by counteracting the inhibitory effect of ABA. The growth of seedlings were enhanced in terms of length, fresh and dry weight. 28-Homobrassinolide at  $3 \mu\text{M}$  concentration was most effective in reversing the effect of ABA. The results obtained in the present study

Table 1 : Effect of abscisic acid and its interaction with brassinosteroids on germination and seedling growth of *T. foenum-graecum*.

Treatment	Percentage of seed germination*		Seedling length (cm)*	Fresh weight of seedling (mg)*	Dry weight of seedling (mg)*
	24 hours	36 hours			
Control	73.3 ± 1.08	81.3 ± 1.08	6.4 ± 0.25	263.6 ± 6.9	17.9 ± 0.34
0.5x10 <sup>-4</sup> M ABA	9.3 ± 1.08	38.6 ± 2.17	2.03 ± 0.17	92.4 ± 5.4	9.8 ± 0.31
10 <sup>-4</sup> M ABA	1.3 ± 1.08	30.6 ± 1.08	1.8 ± 0.14	82.6 ± 2.76	7.8 ± 0.24
0.5x10 <sup>-4</sup> M ABA + 1 µM 28-HB	46.6 ± 1.08	82.6 ± 1.08	5.4 ± 0.20	221 ± 3.40	13.5 ± 0.97
0.5x10 <sup>-4</sup> M ABA + 3 µM 28-HB	68.0 ± 1.88	82.6 ± 1.08	5.9 ± 0.18	238.1 ± 2.04	18.3 ± 0.24
0.5x10 <sup>-4</sup> M ABA + 1 µM 24-EB	36.0 ± 2.00	67.0 ± 0.82	4.48 ± 0.16	213 ± 1.93	13.3 ± 0.30
0.5x10 <sup>-4</sup> M ABA + 3 µM 24-EB	42.0 ± 1.00	73.0 ± 1.65	4.79 ± 0.11	219 ± 2.30	17.0 ± 0.29
10 <sup>-4</sup> M ABA + 1 µM 28-HB	60.6 ± 1.96	77.3 ± 2.17	4.69 ± 0.12	218 ± 0.61	12.8 ± 0.90
10 <sup>-4</sup> M ABA + 3 µM 28-HB	68.0 ± 1.88	80.0 ± 1.88	4.98 ± 0.10	228 ± 9.7	17.2 ± 0.30
10 <sup>-4</sup> M ABA + 1 µM 24-EB	29.3 ± 2.17	53.3 ± 1.08	4.12 ± 0.05	214 ± 2.39	12.4 ± 0.83
10 <sup>-4</sup> M ABA + 3 µM 24-EB	37.3 ± 2.17	65.3 ± 1.08	4.49 ± 0.10	219 ± 1.73	16.3 ± 0.27

ABA : abscisic acid, 28-HB = 28-homobrassinolide

24-EB = 24-epibrassinolide

\*Mean ± SE.

indicate that brassinosteroids are capable of reversing the inhibitory effect of abscisic acid on seed germination and seedling growth. In an earlier study<sup>4</sup> it was reported that brassinolide stimulated growth of etiolated squash (*Cucurbita maxima*) hypocotyls was associated with decrease in the levels of ABA.

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**References**

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**Introduction**

The bell-shaped berry fruit of *E. jamborum* yellowish when it ripens. It develops from the bicarpellary inferior ovary with axile placentation. Anomocytic and paracytic types of stomata on the foliar epidermis of Myrtaceae have been reported<sup>5,6</sup>. The present investigation is undertaken to trace out the epidermal features and stomatal complexes in the developing fruit wall of *E. jambor*.

**Materials and Methods**

The different developmental stages of *E. jambor* fruit starting from the ovary to mature fruit were collected and fixed in F.A.A. Epidermal peels from basal, middle and terminal regions of all developmental stages of fruit were obtained by treating the fruit walls with 60% HNO<sub>3</sub> and stained with iron haematoxylin or 1% aqueous safranin. Starch grains and oil granules were localized with I KI and Sudan Black B respectively. Drawings were made using Austin Projection Microscope (REichert). The

frequencies of epidermal cells and stomatal complexes were counted under Olympus Microscope by taking field area in 400 X (Table 1) and the stomatal index has been calculated<sup>7</sup>.

**Observations**

The epidermal cells of the ovary wall are small, polygonal or isodiametric (Fig. 1A). However, occasionally they appear elongated and irregularly arranged. As the fruit grows, the size of the epidermal cells increases and cell walls become thick and beaded in nature. An abnormal wall thickening is also noticed in the epicarp of different parts of the fruit. Epidermal cells are found to contain dense cytoplasm with prominent and spherical nuclei along with starch grains and oil granules. Epidermal cells of the developing epicarp undergo divisions but the frequency of division is found to be decreased gradually towards maturity due to the submergence and elongation of epidermal cells (Table 1).

**Stomata:** The epicarp of developing fruit possesses predominantly six types of stomata