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Acclimation: An Effective Tool for Enhancing Low-Temperature Stress in Bitter Gourd

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Abstract

The impact of acclimation was investigated on growth and photosynthetic parameters in bitter gourd seedlings (Punjab-14 and PAUBG-56) under low temperatures. Fifteen-day-old seedlings were exposed to 5°C after acclimation for seven days and thereafter assessed for recovery at 25°C. For acclimation, the temperature was gradually brought to 5°C from 200C. Seedlings performed better after acclimation treatment in terms of survival percentage, root and shoot length, total chlorophyll, chlorophyll a, Chlorophyll b, Fv/Fm, and electrolyte leakage. PAUBG-56 showed higher resilience to low temperatures than Punjab-14. It is concluded that acclimation helped the plants to maintain growth probably through chlorophyll and membrane stability under low-temperature stress, thus an effective tool for developing resistance in plants.

Introduction

Bitter gourd (Momordica charantia L.) also known as karela, bitter melon, or balsam pear is a flowering vine in the genus Momordica of the family Cucurbitaceae. It is a climbing herb that thrives in tropical, subtropical, and temperate regions but is now cultivated all over the world because of the medicinal and dietary value of both unripe and ripe fruits. In India, bitter gourd occupied an area of 97 thousand hectares with a production of 1137 thousand metric tonnes during 2017-18. Generally, bitter gourd is an annual crop but can also grow as a perennial crop in areas having mild, frost-free winters. It is a tropical crop requiring warm conditions thriving from low land areas to altitudes of up to 1,000 m. Bitter gourd is an important vegetable cultivated in parts of Asia, East Africa, the Caribbean, and South America, where it is used both as food as well as a medicine. This species originated from a tropical/subtropical evolutionary background and does not have the genetic information for chilling tolerance. Low temperature (LT) or cold stress affects plant growth and crop productivity and can lead to substantial crop losses. Chilling stress produces injury without forming ice crystals in plant tissues, and in freezing stress ice formation occurs within plant tissues. Plants differ in their tolerance to chilling (0-15°C) and freezing (<0°C) temperatures. Both chilling and freezing stresses are together known as low temperature or cold stress. In general, plants originating from tropical and subtropical regions are sensitive to LT, whereas temperate plants showed chilling tolerance to variable degrees. Low-temperature stress may affect the survival rate of crop plants and also affect various processes including cell division, photosynthesis, plant growth, development, and metabolism and finally reduce the yield of crop plants, especially in the tropics and subtropics. In several species, the acquisition of freezing tolerance can be induced by exposure to low, non-freezing, and non-injurious temperatures. Acclimation may be defined as changes that occur in a plant in response to chilling temperatures which confer subsequent tolerance to the cold injury, especially during germination and early seedling growth.

Survival percentage, root length (RL) (cms), shoot length (SL) (cms), and RL/SL ratio

Low temperature affected the mortality percentage of the seedlings. Clear differences in survival percentages between the two gourd genotypes were evident after 7 days of recovery from stress (Fig. 1a). Cold acclimation significantly enhanced the survival rate of bitter gourd seedlings exposed to 5˚C. The survival percentage in Punjab-14 and PAUBG-56 after acclimation was 100%. In the PAUBG-56 genotype, a 10.7% higher survival percentage at direct 5˚C exposure was seen as compared to the Punjab-14 genotype. The Punjab-14 seedlings registered 64% survival, suggesting that it too exhibited good growth when exposed to low temperature. The higher survival percentage of the PAUBG-56 genotype is an indication of its better cold tolerance. Similarly, after acclimation surviving capability of both the genotypes enhanced. This might be due to enhanced cold tolerance mechanisms in seedlings during non-lethal temperature exposure.

The growth of seedlings was quantified in terms of changes in root and shoot length. LT stress on acclimated seedlings did not depict negative growth. PAUBG-56 registered a 69.81% increase in root length and a 36.43% increase in shoot length concerning control. While Punjab-14 registered a 16.45% increase in root length, whereas shoot length registered an increase of 73.62%. Direct exposure of seedlings at 5˚C caused a 48.48% and 70.58% decrease in root length in Punjab-14 and PAUBG-56 respectively in comparison to the control (Fig. 1ba). The decrease in shoot length was less in PAUBG-56 (39.27%) then Punjab-14 (69.94%) in comparison to control.

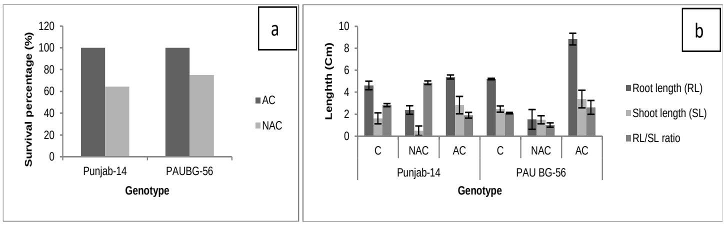

Figure 1: Effect of low temperature and cold acclimation on survival percentage (a), root length (RL) (cm), shoot length (SL) (cm), and RL/SL ratio (b) of bitter gourd seedlings (C- control, NAC-non acclimated at 5°C, AC-acclimated at 5°C) [values are mean±SD from 3 seedlings (CD=5%)].

Electrolyte leakage, chlorophyll pigments, and F_v/F_m

A look at chlorophyll (Chl) data (Figure 2a) showed that exposure to low temperature resulted in a greater decrease in Chl a and total Chl in PAUBG-56, although the basal level of these was higher in this genotype. The effect of LT stress on Chl b was not significant, however, after recovery, the values shifted toward normal. Acclimated seedlings of both genotypes performed better under LT stress. The chlorophyll a/b ratio increased by 97% and 98% in acclimated seedlings of Punjab-14 and PAUBG-56 respectively in comparison to the control (Figure 2b). Low-temperature stress increases the chlorophyllase enzyme activity and restrains the synthesis of total chl. So, plants need to maintain a sufficient level of Chl content to perform the photosynthesis process to some extent even under stress. Acclimation helps in maintaining a higher chl a/b ratio and a lesser decrease in total chlorophyll content.

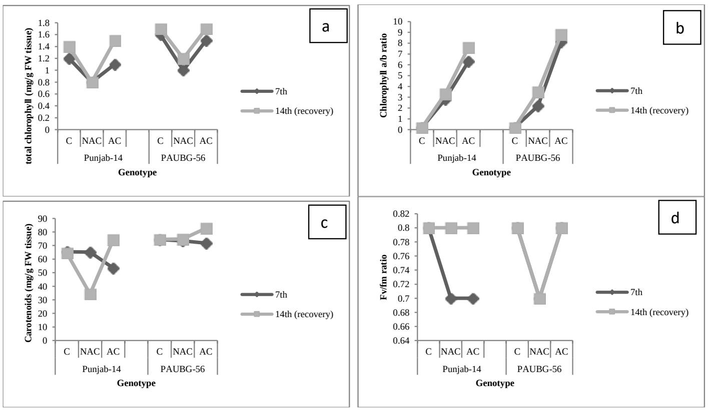

Figure 2: Effect of low temperature and cold acclimation on chlorophyll pigments in leaves of bitter gourd genotype seedlings, C- control, NAC-non acclimated at 5°C, AC-acclimated at 5°C, [values are mean±SD from 3 seedlings (CD=5%)].

Low-temperature stress at 5˚C caused a decrease in carotenoid content, which increased on the 7th day of stress. However, after recovery, Punjab-14 displayed a decrease in carotenoid content, but the content remained unchanged in PAUBG-56. Carotenoid content in acclimated seedlings of Punjab-14 and PAUBG-56 genotypes decreased by 18% and 3% respectively in comparison to the control. Post recovery the content in Punjab-14 and PAUBG-56 increased by 13% and 10% respectively (Figure 2c). Carotenoids are not photosynthetic pigments, but they are accessory pigments that play an important role in the protection of the photosystems and accumulate under low temperatures. Carotenoids act as natural antioxidants by quenching triplet Chl and singlet oxygen species which are potentially harmful to the chloroplast. However, some studies have reported a decrease under low-temperature stress. The possibility of low chlorophyll and carotenoid content could be due to oxidative stress caused by low-temperature exposure. It is well documented that photosynthetic apparatus is sensitive to several environmental stresses and PS II appears to be preferentially affected by chilling stress. The Fv/Fm ratio remained unchanged in response to LT stress in leaves of both the genotype seedling leaves as compared to control values (Figure 2d). Fv/Fm reflects the susceptibility to damage of the PSII. In the present study, the Fv/Fm ratio was not affected by cold stress, suggesting that low temperature did not affect the efficiency of PSII. Cold acclimation maintained the Fv/Fm ratio in plant.

Electrolyte leakage in Punjab-14 increased by 68.5% at 5˚C whereas in PAUBG-56 it increased by 40.5%. Electrolyte leakage was significantly (CD at 5%) lower in acclimated seedlings as compared to low-temperature exposed seedlings in comparison to control seedlings (Figure 3). Low-temperature stress is responsible for the accumulation of ROS viz. singlet oxygen ($^1O_2$), superoxide radical ($O_2^{\bullet -}$), hydrogen peroxide (H_2O_2), and hydroxyl radicals ($OH^{\bullet}$) through various chemical reactions. The ROS accumulated under LT stress leads to membrane lipid peroxidation causing an increased MDA content, electrolyte leakage, and finally membrane instability. Higher electrolyte leakage indicated that the Punjab-14 genotype was more affected by cold stress. Higher EL values reflected that LT stress caused membrane dehydration and ion leakage. Acclimation helped to lower electrolyte leakage.

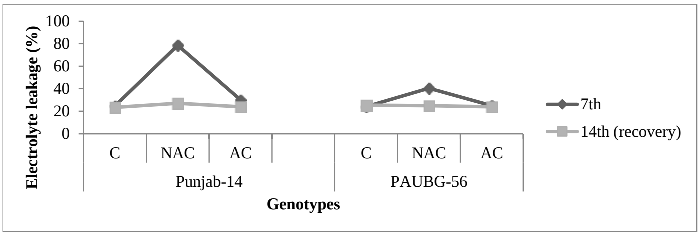

Figure 3: Effect of cold acclimation on electrolyte leakage in leaves of bitter gourd genotype seedlings, C- control, NAC-non acclimated at 5°C, AC- acclimated at 5°C, [values are mean±SD from 3 seedlings (CD=5%)].

Conclusions

Bitter gourd is an important vegetable crop cultivated in India mainly in summers but is in demand around the year due to its medicinal importance. Direct exposure to low temperature decreased survival percentage to a greater extent in Punjab 14. However, Chla and total Chl decreased more in PAUBG 56. Carotenoids and electrolyte leakage were less affected. PAUBG 56 was inherently high in these components than Punjab 14 seedlings. Acclimation improved the survival percentage in both genotype seedlings to 100% probably due to high Chla, total Chl and membrane stability.

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