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Research Article

Stress-Induced Transmissible Flowering Stimulus In Phyllanthus, Acalypha

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ABSTRACT

Many plant species can be encouraged to blossom by responding to stressors. Phyllanthus niruri and Acalypha indica both short-day plants, blossom on long days in response to stress caused by insufficient nutrition or low-intensity light. Grafting tests with two P. niruri types demonstrated the presence of a transmissible flowering stimulus in stress-induced blooming. The stress-induced flowering of P. niruri and A. indica plants resulted in anthesis, fruit, and seed production. These seeds germinated, and the progeny of the stressed plants grew normally. Phenylalanine ammonia-lyase inhibitors decreased stress-induced flowering, but the inhibition was alleviated by salicylic acid (SA), indicating that SA plays a role in stress-induced blooming. PnFT2, a P. niruri ortholog of the Arabidopsis thaliana flowering gene FLOWERINGLOCUS T (FT), was expressed when A. indica plants were induced to flower under poor-nutrition stress conditions, whereas PnFT1, another ortholog of FT, was not, implying that PnFT2 is involved in stress-induced flowering.

Key words: *flowering, stress, phenylalanine ammonia-lyase, salicylic acid, Phyllanthus niruri, Acalypha indica*

Introduction

In numerous plant species, flowering is governed by environmental conditions, including photoperiodic night length and temperature during vernalisation. Conversely, a short-day (SD) plant like P. niruri can be prompted to flower under long-day (LD) conditions when cultivated in environments characterised by inadequate nourishment, cold temperatures, or high-intensity light. The flowering prompted by these conditions is accompanied by an elevation in phenylalanine ammonia-lyase (PAL) activity.[1] Collectively, these results imply that the flowering generated by these conditions may be governed by a same mechanism. Poor nutrition, cold temperatures, and high-intensity light are considered stressors, leading to an increase in PAL activity under these conditions. Consequently, we hypothesised that the long-day flowering in A. indica may be triggered by stress. Non-photoperiodic flowering has been intermittently documented in many plant species aside from P. niruri, and a review of these studies indicated that the majority of factors [1]influencing flowering can be classified as stressors. Several examples of these factors are summarised. Consequently, the data supporting stress-mediated flowering is mounting. Consequently, we designate this phenomenon as 'stress-induced blossoming.' P. niruri can bloom under long-day conditions when cultivated in tap water (nutritional deficiency stress), at temperatures between 12 to 15°C (cold stress), or under light intensities of 15,000 to 20,000 lux (high-light stress). [2]The responses to various stressors vary according to the cultivars. For instance, cv. Tendan did not initiate flowering while experiencing nutritional stress from cultivation in tap water, although vegetative development was markedly suppressed, demonstrating that the plants were genuinely under stress. Tendan did not bloom under high-intensity light stress either. Cv. Kidachi did not flower due to inadequate nourishment or excessive light intensity. Conversely, the white-flowered mutant of cultivar Violet exhibited heightened sensitivity to low-temperature stress. A.indica seedlings react to these stressors as the cotyledons grow and also to short-day treatment during photoperiodic flowering. The removal of cotyledons postponed flowering, suggesting that cotyledons are essential for stress-induced flowering, akin to photoperiodic flowering.[3]

Recently, we discovered that the short-day plant, Perilla frutescens var. crispa, was prompted to flower under long-day circumstances when cultivated under low-intensity light (30 μmol m^{-2} s^{-1}). Fourteen The suppression of vegetative growth during the induction of flowering exemplifies stress-induced blooming. The flowering response was more pronounced in the red-leafed variant compared to the green-leafed variant.[4] We subjected the red-leafed variant to stressors aside from low-intensity light. The plants were cultivated in tap water or a diluted mineral nutrient solution (nutritional deficiency stress), at temperatures ranging from 5 to 15°C (cold stress), with sodium chloride concentrations of 50 to 400 mM (salinity stress), or with inadequate irrigation (hydration stress). None of these variables stimulated flowering; rather, they inhibited vegetative development.[5]Consequently, not all forms of stress can stimulate flowering. The red-leafed P. frutescens, subjected to low-intensity light throughout the expansion of their cotyledons, were induced to flower after a 3-week treatment, achieving 100% flowering during a 4-week treatment. The plants may react to low-intensity light promptly following the expansion of the cotyledons. The blooming response diminished with increasing plant age, and flowering was not stimulated when the low-intensity light treatment commenced two weeks following cotyledon expansion or at any subsequent time.[5]

METHODOLOGY & DISCUSSION

Production of Progeny by Plants that Flowered Under Stress Conditions

Plants can alter their growth to acclimatise to stressful situations. Stressed plants may bloom as a survival mechanism to propagate the next generation. under this manner, plants can sustain their species, even under adverse environments. For this reaction to confer a biological advantage, plants stimulated to blossom by stressors must yield viable seeds, and the offspring must develop appropriately.[6]

Phyllanthus niruri Violet was cultivated in a nutrient-rich solution or tap water for its whole lifespan. The plants subjected to flowering induction through nutrient-deficient stress conditions attained anthesis, bore fruit, and generated seeds. All of these seeds successfully germinated, and the offspring of the stressed plants exhibited normal development. The offspring reacted to SD therapy and developed flower buds.

Additionally, a typical second-generation from the stress progeny was generated. A. indica plants were cultivated under long-day circumstances with low-intensity illumination from the stage at which the cotyledons expanded. The plants stimulated to blossom under these conditions attained anthesis and produced seeds.[7]

The seeds germinated, developed properly, and were prompted to blossom in response to short day treatments. The results suggest that stressed plants do not have to wait for a season with favorable photoperiodic conditions for flowering, and this early blossoming may aid in species conservation.

Consequently, stress-induced flowering may confer a biological advantage and should be regarded as equally significant as photoperiodic flowering and vernalization.

Transmissible Flowering Stimulus Produced by Stress

Stress-induced flowering of P. niruri is inhibited by the PAL inhibitor aminooxyacetic acid (AOA), and therefore it is hypothesized that some compounds in the metabolic pathway regulated by PAL act as flowering stimuli. However, a transmissible flowering stimulus like florigen, which is involved in photoperiodic flowering, has not been reported in stress-induced flowering. To investigate this possibility, we performed grafting experiments to detect the transmission of stress-induced flowering stimuli in A.indica.

Violet and Tendan were combined through grafting in various configurations, and the resulting plants were cultivated in tap water under long-day circumstances. The Violet scions, when grafted onto the Violet rootstocks, produced flowers. The flowering may have resulted from the rootstocks' impact, as all leaves were excised from the scions. This indicates that a transmissible flowering stimulus plays a role in stress-induced flowering.[8] We anticipated that Tendan would not generate a flowering stimulus, as it did not bloom under conditions of nutritional deficiency. Nonetheless, the defoliated Violet scions grafted onto Tendan rootstocks bearing cotyledons were stimulated to blossom. The Tendan scions grafted onto the Violet rootstocks did not produce flowering. The data imply that Tendan generates a transmissible flowering stimulus but does not react to it.

Involvement of Salicylic Acid (SA) in Stress-Induced Flowering

As previously stated, certain compounds in the metabolic pathway regulated by PAL may function as blooming stimuli in P. niruri. Chlorogenic acid, a phenylpropanoid, was a significant candidate in earlier studies. Nonetheless, externally administered chlorogenic acid did not stimulate flowering. Besides chlorogenic acid, several chemicals such as SA and anthocyanin are synthesised from t-cinnamic acid, a process catalysed by PAL from phenylalanine. 11 A component in the metabolic pathways originating from t-cinnamic acid may play a role in stress-induced blooming. We stimulated blooming in P. nil through low-temperature or nutrient-deficient stress, suppressed flowering with AOA, and concurrently administered several metabolic intermediates in the pathways. Among the intermediates, t-cinnamic, benzoic acids, and SA were demonstrated to counteract the inhibitory impact of AOA, although p-coumaric and caffeic acids did not. These findings indicate that SA has a role in the stress-induced blooming of P. niruri. Stress facilitates the conversion of t-cinnamic acid to salicylic acid through benzoic acid. Salicylic acid (SA) performs multiple physiological functions in plant development. 18 The application of benzoic acid, salicylic acid, or benzoic acid derivatives to P. niruri and A. indica before exposure to low temperatures amplifies the flower-inducing impact of the low-temperature therapy.[9]

Moreover, several derivatives of benzoic acid and salicylic acid stimulate flowering in Phyllanthus niruri, while benzoic acid promotes the flowering of cultured plumules excised from photoinduced Phyllanthus niruri seedlings. SA promotes flowering in numerous species. to the Lemnaceae, where it has been associated with the stress-induced blooming of A. indica. These facts substantiate the aforementioned conclusions. Nonetheless, SA alone did not trigger blooming in P. niruri under non-stress conditions. Stress situations may stimulate not only salicylic acid biosynthesis but also other critical components that promote flowering.[10]
Previous observations indicated that the leaves of red-leafed P. frutescens exhibited a rich green colouration when stimulated to blossom under low-intensity light. The chlorophyll development in the leaves resulted from a reduction in anthocyanin levels. A negative association existed between anthocyanin content and the percentage of flowering. PAL is a crucial enzyme in anthocyanin biosynthesis; thus, we can infer that low-intensity light promotes flowering by inhibiting PAL activity. This, however, contradicts prior knowledge. Stress typically elevates PAL activity and stimulates anthocyanin production; furthermore, PAL activity is enhanced during the stress-induced blooming of P. niruri, as previously noted. Consequently, we investigated the impact of the PAL inhibitor on low-intensity light-induced blooming in A. indica. AOA and the PAL inhibitor L-2-aminooxy-3-phenylpropionic acid (AOPP) did not promote flowering under non-inductive normal-intensity light and suppressed flowering under inductive low-intensity light. The results indicate that the same mechanism is implicated in the flowering caused by low-intensity light in A. indica and by various stress conditions in P. nil. The observation that PAL inhibitors suppressed stress-induced blooming indicates that stress elevated PAL activity. In P. frutescens, the reduction of anthocyanin content under low-intensity light indicates that stress inhibited the function of PAL. These incongruous outcomes require elucidation.

CONCLUSION

Involvement of PnFT Genes in Stress-Induced Flowering of P. niruri and A. indica

A molecular methodology has not been previously utilised to investigate stress-induced flowering. Consequently, we investigated genes implicated in the stress-induced blooming of P. nil. The flowering of A. thaliana is stimulated by long day (LD) conditions, vernalisation, autonomous signals, and gibberellins, all of which function through a unified route mediated by FLOWERING LOCUS T (FT). This indicates that FT may also participate in flowering triggered by stressors. Two orthologs of FT, PnFT1 and PnFT2, have been found in P. niruri and A. indica; these genes are expressed in inductive short-day circumstances to facilitate flowering. Consequently, we investigated the expression of PnFT genes under situations of nutritional deficiency.
Phyllanthus niruri and
A. indica Violet was prompted to bloom through cultivation in tap water; the cotyledons and true leaves of these specimens were harvested, and the expression levels of PnFT1 and PnFT2 were analysed with RT-PCR. The expression of PnFT2 was stimulated in plants cultivated under nutrient-deficient conditions for a duration of two weeks or more. Conversely, PnFT1 was not expressed under any dietary circumstances. The results indicate that PnFT2, but not PnFT1, has a role in the stress-induced flowering of P. nil. Salicylic acid (SA) may stimulate the expression of PnFT2, or the product of PnFT2 may activate the expression of genes associated with the production, response to, or signal transduction of SA. PnFT2 participates in both photoperiodic and stress-induced flowering, whereas PnFT1 is solely associated with photoperiodic flowering. The two PnFT genes may serve distinct functions in the regulation of blooming based on the inductive stimulus. The key gene for blooming may be PnFT2, with PnFT1 expression being raised solely by SD treatment, hence redundantly augmenting the activity of PnFT2.[9,10]

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