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Thermal Stress-Induced Alterations in Cell Membrane Permeability of Two Wild Datura Species from the Menal Forest Region, Rajasthan

Ariba Khan[1], Shahdab Hussain[2]

[1]Research Scholar, Department of Botany, Sangam University, Bhilwara-311001, Rajsthan, India

[2]Associate Professor, Department of Botany, Sangam University, Bhilwara-311001, Rajsthasn, India
Email: *aribakhankayamkhani[at]gmail.com*

Abstract: ***Cell membrane permeability is a key physiological indicator of thermal stress in plants. The present investigation was conducted to evaluate the effect of hyperthermia on the membrane stability of two wild Datura species-Datura innoxia Mill. and Datura metel L. var. fastuosa (L.)-collected from the Menal Forest region of Rajasthan. Leaf samples were exposed to temperature regimes of 40°C and 45°C for 1, 2, and 4 hours. The extent of membrane injury was assessed through the quantification of soluble sugar and protein leachates expressed as a percentage of untreated controls. Results revealed that both species exhibited increased solute leakage with higher temperature and exposure duration, signifying progressive damage to membrane integrity. Datura innoxia showed moderate leakage (sugar: 21.5–82.7%; protein: 19.4–76.2%), whereas Datura metel var. fastuosa displayed comparatively higher leakage (sugar: 23.8–86.4%; protein: 21.5–82.1%). These findings indicate that Datura innoxia possesses relatively greater thermo-tolerance than D. metel var. fastuosa (L.). The study highlights differential heat resilience among congeners, which may contribute to understanding stress physiology in wild Solanaceae taxa of semi-arid environments.***

Keywords: Cell membrane permeability, *Datura innoxia* Mill., *Datura metel* L. var. *fastuosa* (L.), soluble sugar, protein leachate, thermal stress, Menal Forest.

1. Introduction

Cell membranes play a pivotal role in maintaining cellular homeostasis by regulating solute transport, compartmentalization, and protection of organelles. They are composed of lipid bilayers interspersed with proteins, whose structural integrity determines selective permeability and stress tolerance. Under hyperthermia, membrane lipids undergo phase transitions, proteins become denatured, and overall permeability increases, leading to loss of cellular solutes such as sugars and proteins (Wang et al., 2025). Recent findings demonstrate that membrane permeability serves as a sensitive indicator of thermal stress in plants. Studies on potato genotypes have shown that leaf membrane stability under 50°C strongly correlates with heat tolerance and yield performance (Plants, 2024). Similarly, research on molecular regulation under high temperatures revealed that increased permeability, altered osmotic balance, and reduced photosynthesis are common plant responses when temperature exceeds tolerance limits (Wang *et al.,* 2025). Membrane lipid remodeling and heat shock protein expression play critical roles in maintaining membrane stability during thermal stress (Maryam *et al.,* 2025). The degree of unsaturation in fatty acids, lipid-protein interactions, and antioxidant mechanisms all contribute to thermal adaptation (Harmonizing Plant Resilience, 2025). Moreover, changes in lipid signaling and osmolyte accumulation act as secondary defense mechanisms to preserve cellular integrity under extreme heat (Saleem *et al*., 2024). Despite these advances, studies on wild taxa—particularly in semi-arid ecosystems like Rajasthan—are limited. Wild Solanaceae species often display unique physiological adaptations, offering insights into stress tolerance and crop resilience. Therefore, the present investigation was designed to assess the effects of hyperthermia on cell membrane permeability in two wild Datura species ***Datura innoxia* Mill.** and ***Datura metel* L. var. *fastuosa* (L.)** collected from the Menal Forest of Rajasthan, focusing on soluble sugar and protein leakage as indicators of membrane injury.

2. Materials and Methods

2.1 Collection of Plant Material

Fresh, healthy leaves of ***Datura innoxia* Mill.** and ***Datura metel* L. var. *fastuosa* (L.)** were collected from the natural flora of Menal Forest, Rajasthan (24°59′N, 74°56′E). Selection criteria focused on fully expanded, mature leaves free from visible disease or mechanical damage to ensure uniform physiological status. Plant identification was confirmed using regional floras (Jain & Rao, 1977; Singh, 1999), and voucher specimens were prepared and deposited in the herbarium of the Department of Botany, Sangam University, Bhilwara, for future reference. Collected leaves were transported immediately to the laboratory in labeled polyethylene bags to prevent desiccation and maintain cellular integrity.

2.2 Experimental Design

Leaf discs of uniform size (diameter: 1 cm) were excised from mature leaves and subjected to controlled heat treatments in a temperature-regulated incubator. Two temperature regimes, 40°C and 45°C, were applied for three durations: 1, 2, and 4 hours. Untreated leaf discs maintained at ambient temperature (25 ± 2°C) served as controls. Each treatment was performed in triplicate to ensure reproducibility. The experimental design was a completely randomized design (CRD), which minimized potential variability due to leaf position or plant-to-plant differences. Post-treatment, leaf discs were immediately processed for biochemical analyses to prevent further metabolic changes.

2.3 Estimation of Soluble Sugars and Proteins

Soluble sugars were estimated using the anthrone reagent method (Hedge & Hofreiter, 1962), which involves extraction in ethanol followed by colorimetric determination at 620 nm. Soluble proteins were quantified according to Lowry *et al.* (1951) using bovine serum albumin as the standard. Both parameters were expressed as a percentage relative to control samples, providing a quantitative measure of cellular membrane integrity and leakage under thermal stress. Increased sugar or protein leakage indicates higher membrane permeability, reflecting stress-induced cellular damage.

2.4 Data Analysis

Data from three replicates were averaged, and mean values were tabulated. Comparative analysis was performed to evaluate the effects of temperature and exposure duration on soluble sugar and protein leakage. Statistical significance was assessed using one-way ANOVA, and differences between treatments were considered significant at $p < 0.05$. Graphical representations were prepared to illustrate trends in leakage with increasing temperature and exposure time.

3. Results

3.1 Soluble Sugar Leakage

Both ***Datura innoxia*** and ***Datura metel* var. *fastuosa* (L.)** exhibited progressive increases in soluble sugar leakage with rising temperature and duration, indicating temperature-induced membrane damage.

At 40°C**,** ***D. innoxia*** showed 21.5% leakage at 1 hour, increasing to 34.2% at 2 hours and 46.9% at 4 hours**.** ***D. metel* var. *fastuosa* (L.)** displayed slightly higher leakage, with 23.8%, 39.6%, and 53.1% at the respective time points. When exposed to 45°C, the leakage sharply increased, reaching 58.3%, 71.5%, and 82.7% for ***D. innoxia*** and 61.7%, 76.2%, and 86.4% for ***D. metel* var. *fastuosa* (L.)** at 1, 2, and 4 hours, respectively. These results clearly indicate that ***D. metel* var. *fastuosa* (L.)** is more thermally sensitive than ***D. innoxia***, as evidenced by higher sugar leakage at all exposure durations. The progressive increase in leakage with time suggests cumulative membrane damage under prolonged heat stress. Such differential responses highlight species-specific variations in heat tolerance, which could be linked to differences in membrane lipid composition, antioxidant capacity, or osmoprotectant levels.

Table 1: Effect of hyperthermia on soluble sugar leakage (% of control)

Species	40°C (1h)	40°C (2h)	40°C (4h)	45°C (1h)	45°C (2h)	45°C (4h)
Datura innoxia Mill.	21.5	34.2	46.9	58.3	68.5	82.7
Datura metel L. var. *fastuosa* (L.)	23.8	39.6	53.1	61.7	73.2	86.4

3.2 Soluble Protein Leakage

A similar trend was observed for soluble protein leakage. At 40°C, ***D. innoxia*** exhibited leakage ranging from 19.4% to 38.1%, while ***D. metel* var. *fastuosa* (L.)** ranged from 21.5% to 45.7%. At 45°C, the leakage values increased to 52.6–76.2% for D. innoxia and 56.9–82.1% for ***D. metel* var. *fastuosa* (L.)**. The higher leakage in ***D. metel* var. *fastuosa*** suggests relatively weaker membrane integrity under prolonged thermal stress.

Table 2: Effect of hyperthermia on soluble protein leakage (% of control)

Species	40°C (1h)	40°C (2h)	40°C (4h)	45°C (1h)	45°C (2h)	45°C (4h)
Datura innoxia Mill.	19.4	28.7	38.1	52.6	61.3	76.2
Datura metel L. var. *fastuosa* (L.)	21.5	33.2	45.7	56.9	66.8	82.1

4. Discussion

The results reveal that membrane permeability increases significantly under elevated temperatures, confirming that hyperthermia leads to cellular injury. High temperatures disturb lipid-protein interactions, increase fluidity, and compromise membrane semi-permeability, resulting in enhanced leakage of solutes (Maryam *et al*., 2025). ***Datura innoxia*** showed moderate increases in sugar and protein leakage, suggesting better thermostability, possibly due to higher saturated lipid content and enhanced synthesis of protective heat shock proteins (Wang *et al*., 2025). On the other hand, ***D. metel* var. *fastuosa* (L.)** exhibited greater leakage, indicating lower thermal resilience and a higher rate of membrane destabilization. Studies in other Solanaceae members have shown that heat-induced lipid oxidation and ROS generation can disrupt plasma membranes, while osmoprotectants such as soluble sugars act as stabilizers against dehydration and heat injury (Harmonizing Plant Resilience, 2025). Furthermore, lipid remodeling and antioxidant enzyme activation are essential for maintaining membrane integrity under stress conditions (Saleem *et al.,* 2024). The comparative analysis indicates that ***D. innoxia*** employs stronger defense mechanisms, possibly through improved ROS scavenging and membrane lipid reorganization, contributing to its relative heat tolerance. These findings align with contemporary studies highlighting lipid-based thermotolerance and protein stabilization mechanisms as key determinants of plant resilience under global warming scenarios (Maryam *et al.,* 2025).

5. Conclusion

Both ***Datura innoxia*** **Mill.** and ***D. metel*** **var.** ***fastuosa*** **(L.)** exhibit increased cell membrane permeability under hyperthermic conditions. However, ***D. innoxia*** demonstrates relatively higher membrane stability and lower solute leakage, suggesting greater thermo-tolerance. The study enhances understanding of interspecific differences in heat resilience among wild Solanaceae species and provides a physiological basis for selecting thermotolerant taxa for ecological and agronomic improvement programs.

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