Check for updates

Physiologia Plantarum

SPECIAL ISSUE Methodologies to Assess Crop Stress Resilience

SPPS

EDITORIAL

Editorial: Special Issue "Methodologies to Assess Crop Stress Resilience"

Christos Bazakos[1] | Michal Lieberman-Lazarovich[2] | Hélène S. Robert[3]

[1]Institute of Plant Breeding and Genetic Resources, ELGO DIMITRA, Thessaloniki, Greece | [2]Department of Vegetable and Field Crops Research, Institute of Plant Sciences, Agricultural Research Organization—Volcani Institute, Rishon LeZion, Israel | [3]Plant Biotech for Stress Resilience, Genomics and Proteomics of Plant Systems, Central European Institute of Technology (CEITEC), Masaryk University, Brno, Czech Republic

Correspondence: Christos Bazakos (cbazakos@elgo.gr) | Michal Lieberman-Lazarovich (michall@volcani.agri.gov.il) | Hélène S. Robert (helene.robert.boisivon@ceitec.muni.cz)

Received: 27 April 2026 | **Accepted:** 28 April 2026

Handling Editor: D. Gendre

Crop production is challenged by constant and fluctuating abiotic stresses, which are becoming more prevalent with the ongoing climatic changes. Abiotic stress factors include, for example, temperature extremes (cold or heat) and variations in water availability (drought or flooding) and/or quality (extreme salinity, or the existence of heavy metals). Such stresses have a profound impact on plant reproduction, a key step in the production of fruits and seeds, vital for food and feed in a world with a growing population (RECROP COST 2025). As a way to mitigate these challenges, plant researchers have been investigating how plants respond to and adapt to abiotic stresses. RECROP (Reproductive Enhancement in Crop Resilience to Extreme Climate, https://www.recrop-cost.com), an EU-supported COST Action, brings together experts from various plant science disciplines, such as physiology, agronomy, genetics, bioinformatics, molecular biology, mathematics, and ecology. One of RECROP's main goals is to establish a common language among researchers across the diverse research areas of crop science, particularly with regard to the methodologies used for applying stress and evaluating stress responses and resilience. We hope that this will facilitate better communication and a higher level of comparability between experimental systems, in pursuing future solutions for the challenge of food security.

This special issue of *Physiologia Plantarum*, initiated by RECROP, "Methodologies to Assess Crop Stress Resilience", addresses this goal by publishing a collection of 17 review articles by RECROP members and the wider plant community. Altogether, these articles cover main topics and suggest valid, up-to-date methodologies to monitor stress response and resilience in crop plants at various levels.

1 | Imaging and Phenotyping Crop Plants' Abiotic Stress Response

Plants are exposed to a wide range of temperatures throughout their life cycle, including daily and seasonal fluctuations. They need to adapt to these variations depending on their stage of development, as such temperature variations directly impact vegetative and reproductive performance. Each plant species has an optimal growth temperature. Around this optimal temperature, there are temperatures that affect plant development without inducing a stress response. This is known as thermomorphogenesis. Above and below this temperature range, at critical temperatures having a profound impact on development, a stress response is activated that leads to a variety of physiological and molecular events, which, in severe cases, can result in cell, organ, or plant death. The critical temperature range that triggers a stress response varies depending on the species, genotype, development stage (vegetative or reproductive), the tissue being analyzed, and the experimental settings. The ability to withstand extreme temperature (thermotolerance) depends on the optimal temperature range for the species under study. Such thermotolerance is categorized as either basal or acquired. Basal thermotolerance refers to the plant's ability to survive a sudden and severe temperature change without priming. Acquired thermotolerance, on the other hand, suggests an acclimation

© 2026 Scandinavian Plant Physiology Society.

or priming mechanism acquired during an initial exposure to sub-lethal temperatures. When designing experiments to study freezing and heat tolerance, it is important to consider the severity of the injuries and the response, bearing in mind the importance of intensity (temperature range) and duration of the applied stress and the stage of development (Arora 2025; Miller et al. 2026).

Freeze–thaw assays are used to determine freezing tolerance. Freezing can cause reversible and irreversible damage affecting membrane structure integrity and resulting in symplast leakage. Electrolyte leakage assays are performed on tissue exposed to sub-zero temperatures to measure freezing tolerance (Arora 2025). Other assays rely on visual indicators such as regrowth and browning, or on chlorophyll fluorescence measurements. Gradual cooling is a natural event during the freezing process in autumn or spring. Although short cold shock experiments are often performed, the effects of prolonged freezing on injury severity and post-thaw recovery are often underestimated. The length of the recovery phase is also an important parameter for monitoring injury severity. In addition to defining the mechanisms of cold stress and freezing tolerance, Arora (2025) reviews laboratory methodologies that mimic a natural response for testing freezing tolerance.

Similarly, Miller et al. (2026) propose methodologies for reproducible and rigorous study of the impact of heat stress on pollen quality and function. The authors compare and discuss the various heat stress regimes employed by the research community, suggesting an experimental design for testing acquired thermotolerance in pollen. In in vitro assays, efficient evaluation of pollen thermotolerance requires pollen to be harvested prior to analysis. The article lists relevant pollen extraction methodologies and compares their applicability based on the plant species studied. The harvested pollen is then used to assess the impact of stress on quality and viability (Miller et al. 2026). As the fitness of both male and female gametophytes is crucial for seed production, the number of viable ovules determines seed set and, thus, in seed crops, seed yield. Although the stress sensitivity of the male gametophyte and pollen is well described and studied (Miller et al. 2026), the impact of abiotic stress on the female gametophyte is much less investigated and is poorly understood. Muttappagol et al. (2025) describe the impact of abiotic stresses on ovule development and methodologies for phenotypic characterization. They also discuss omics-based tools for evaluating the molecular response of ovules to abiotic stresses, as well as putative targets for breeding varieties that are resilient to reproductive stress. In addition to molecular approaches, Daryanavard et al. (2026) describe various cutting-edge imaging technologies for monitoring the effects of abiotic stresses on reproductive tissues, such as anthers, pollen, ovules, and seeds. They list the biosensors used to monitor the stress response, including ROS and calcium.

Conducting stress experiments that mimic natural conditions is challenging. Transferring observations from laboratory conditions (e.g., phytotrons, growth chambers and greenhouses) to a more realistic setup is not straightforward. Çınar and Ünay (2025) list the pros and cons of strategies and approaches for testing the heat tolerance of crops in the field using plastic tunnels, adjusting sowing times, or using additional heating systems. The root system often receives little attention, but it also responds to abiotic stress by adapting its architecture. But how can it be assessed when it is hidden in the soil? Chandnani and Soolanayakanahally (2025) describe different high-throughput methodologies and the necessary equipment for growing roots for a non-destructive, quantitative assessment of morphological and architectural traits. Such experiments rely on imaging analysis, which is one of the limiting factors of this approach. While this has become feasible in laboratory conditions, the authors discuss the possibilities for root phenotyping in the field.

One of the challenges and gaps in translating knowledge into practical breeding strategies is the study of ornamental plant species. While traits such as seed yield and quality are selected for in staple crops, ornamental plants are selected for their aesthetic qualities. Chachar et al. (2025) discuss the molecular responses of ornamental plant species to drought stress and technological strategies for studying these responses.

Last, integrating the florigen of information and multi-omic datasets collected from various plant species, genotypes, and tissues requires mathematical modeling to analyze and predict stress response and identify breeding targets. Sadras et al. (2026) provide a comprehensive overview of how such mathematical modelling can assist in the study of stress biology in crops.

2 | Methodologies to Assess the Molecular Response of Crop Plants to Abiotic Stresses

Understanding how crop plants respond to abiotic stress at the molecular level requires methodologies that can capture changes in gene expression and metabolic activity in a comprehensive and reproducible manner. Ćavar Zeljković et al. (2026) describe recent methodological advances in transcriptomics and metabolomics, from high-throughput RNA sequencing and long-read transcriptomics to increasingly sensitive analytical platforms for metabolite profiling. The authors also discuss the challenges associated with integrating these datasets, particularly in relation to data standardization, environmental variability, and computational complexity, while highlighting emerging approaches such as spatial omics, AI-assisted analytics, and high-throughput phenotyping as promising tools for improving the assessment of crop stress resilience.

Proteomics provides an additional and essential layer of information, as protein abundance, activity, and post-translational modifications cannot be inferred directly from transcript levels. Gupta (2026) describes the introduction of 4D-proteomics as an important technological advance for studying plant proteomes under stress conditions. By incorporating ion mobility as a fourth dimension through trapped ion mobility spectrometry (TIMS) and parallel accumulation-serial fragmentation (PASEF), this methodology improves peptide separation, proteome coverage, and the detection of low-abundance proteins and post-translational modifications. The review highlights the potential of this approach to identify stress-responsive proteins that remain undetected with conventional methods and to provide a more detailed understanding of the molecular mechanisms underlying crop stress resilience.

13993054, 2026, 3, Downloaded from https://onlinelibrary.wiley.com/doi/10.1111/ppl.70921 by Readcube (Labtiva Inc.), Wiley Online Library on [01/06/2026]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

In addition to transcriptional and proteomic regulation, post-transcriptional processes, such as alternative splicing, also contribute substantially to plant adaptation to adverse environments. Vraggalas et al. (2025) review the methodologies used to analyze alternative splicing and its regulation in plants, covering both gene-specific and transcriptome-wide approaches. The authors compare methods such as RT-PCR, minigene assays, and transient expression systems and they further discuss methodologies for identifying interactions between splicing factors and their RNA targets. By summarizing the advantages and limitations of these techniques, the review provides an important framework for studying how alternative splicing contributes to transcriptome plasticity and stress adaptation in plants.

A particular challenge in stress biology is the assessment of reproductive resilience, as reproductive tissues are often more sensitive than vegetative organs and more difficult to phenotype accurately under stress. Bazakos et al. (2026) describe multi-level methodological approaches for assessing molecular and physiological traits associated with drought and heat stress tolerance in reproductive tissues. The review discusses experimental strategies ranging from controlled growth chamber conditions to greenhouse and field experiments, and highlights the integration of physiological, biochemical, and molecular analyses for both male and female reproductive organs. In addition, the authors emphasize the value of multi-omics and emerging single-cell approaches toward a more holistic framework to study stress resilience during crop reproduction.

The integration of multiple omics approaches is particularly important in crops that remain comparatively underexplored despite their agronomic and nutritional value. Najar et al. (2026) discuss how transcriptomics, metabolomics, proteomics, genotyping-by-sequencing, and high-throughput phenotyping can be combined to study sesame's response to abiotic stresses such as drought, salinity, waterlogging, and heat. The authors also highlight the value of combining multi-omics datasets with machine learning and genome–phenome association approaches to identify stress-responsive traits and accelerate the breeding of climate-resilient sesame cultivars. This review therefore highlights sesame as a useful example of how methodological progress can support crop improvement not only in major staple species but also in orphan crops of increasing economic importance.

3 | Enhancing Abiotic Stress Resilience by Priming

Priming is a promising strategy employed to enhance plant performance and resilience by enabling plants to better withstand environmental stresses. Priming, often referred to as acclimation or hardening in the context of abiotic stresses, facilitates crop improvement through the induction of a stress memory, either within the same generation or trans-generationally. This memory allows primed plants to be more tolerant to a subsequent stress in the current and subsequent generations. Priming strategies can be implemented at various stages of plant development and encompass a range of methods depending on crop species, agrological systems, economic feasibility, and the specific stress factors of interest. The priming agent can be chemical, biological, or physical. Primed plants can exhibit cross-stress tolerance, where an initial stimulus provides protection against a different, subsequent stressor. Since some priming agents provoke a general stress response in the plant, this method can be used for inducing multiple-stress tolerance.

Padilla et al. (2025) discuss integrated approaches to improve horticultural crops' resilience against combined abiotic stresses, an issue of utmost importance as real-life agrological conditions usually impose multiple stress factors. Padilla et al. (2025) describe various approaches such as breeding, grafting, and application of biostimulants and/or nanomaterials. The latter serve as priming agents that can activate plant defense mechanisms under stress conditions and were shown, for example, to improve nutrient uptake and water use efficiency under drought conditions. Importantly, nanomaterials are produced at low costs, enabling practical implementation at large scales. Using nanomaterials as priming agents involves the exposure of plants to specific agents that trigger a physiological and molecular "alert state" without causing actual stress damage. Similarly, in the review by Harzli et al. (2025), the authors explore innovative microbial strategies as "primers" to enhance crop resilience to abiotic stresses. Microorganisms like growth-promoting rhizobacteria (PGPR), mycorrhizal fungi, and endophytes are shown to improve plant stress tolerance. These biological priming agents modulate plant stress response pathways and induce resilience by inducing the production of stress-alleviating compounds and enhancing nutrient uptake at the whole-plant level.

Priming may also contribute to abiotic stress resilience at the organ level, primarily seeds. As seed performance and crop establishment are vital as the first yield-determining stage, supporting seed germination and seedling vigor under stressful conditions is crucial. Pasquali Medici de Biron et al. (2026) describe how pre- and post-harvest seed priming, influenced by environmental and maternal conditions, enhances seed vigor, germination, and stress resilience. Pre-harvest priming involves maternal plant treatments and responses to environmental cues that influence seed and seedling performance, while post-harvest priming involves various seed treatments and coatings aimed at improving germination and early growth under stress. Notably, mother–plant priming induces epigenetic changes such as histone modifications and DNA methylation, stabilizing stress-responsive gene expression. Temel and Gören-Sağlam (2026) discuss the role of histone modifications in plant priming, stress memory, response, and resilience. The authors describe in detail the well-established methodology of chromatin immunoprecipitation (ChIP) to analyze histone-tail modifications and elaborate on alternative emerging techniques that address limitations of traditional ChIP, especially in plant systems.

This collection of review articles highlights the importance of using multidisciplinary and standardized approaches to study plant resilience under increasingly changing climatic conditions. Together, the methodologies presented here, ranging from root imaging and phenotyping to advanced omics and priming strategies, provide a broad and timely framework for both fundamental and applied plant research. We believe that the development of a "common methodological language" will strengthen cross-disciplinary collaboration and contribute to

13993054, 2026, 3, Downloaded from https://onlinelibrary.wiley.com/doi/10.1111/ppl.70921 by Readcube (Labtiva Inc.), Wiley Online Library on [01/06/2026]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

the development of climate-resilient crops essential for future food security.

Author Contributions

All three authors equally contribute to drafting, writing, and proofreading the manuscript.

Acknowledgements

This special issue and this editorial article have benefited from networking opportunities provided by the COST Action RECROP (CA22157), supported by COST (European Cooperation in Science and Technology).

References

Arora, R. 2025. "Survival Under Prolonged Cold: Duration of Stress, an Overlooked Factor in Freezing Tolerance Tests." *Physiologia Plantarum* 177: e70633. https://doi.org/10.1111/ppl.70633.

Bazakos, C., M. Vidović, A. Radanović, A. Bacu, S. Francesca, and M. M. Rigano. 2026. "Multi-Level Approaches for Assessing Molecular and Physiological Traits of Drought and Heat Stress Tolerance in Plant Reproductive Development." *Physiologia Plantarum* 178: e70760. https://doi.org/10.1111/ppl.70760.

Ćavar Zeljković, S., F. Saeed, D. Šamec, and U. K. Chaudhry. 2026. "Methodological Advances in Transcriptomics and Metabolomics for Assessing Crop Stress Resilience." *Physiologia Plantarum* 178: e70717. https://doi.org/10.1111/ppl.70717.

Chachar, S., N. Ahmed, and X. Hu. 2025. "Future-Proofing Ornamental Plants: Cutting-Edge Strategies for Drought Resistance and Sustainability." *Physiologia Plantarum* 177: e70255. https://doi.org/10.1111/ppl.70255.

Chandnani, R., and R. Soolanayakanahally. 2025. "The Invisible Frontline: High-Tech Root Imaging for Crop Stress Adaptation." *Physiologia Plantarum* 177: e70572. https://doi.org/10.1111/ppl.70572.

Çınar, V. M., and A. Ünay. 2025. "Performance of the Methods Used to Reflect the High-Temperature Stress in Crops." *Physiologia Plantarum* 177: e70459. https://doi.org/10.1111/ppl.70459.

Daryanavard, H., T. Paraiso, M. C. Pasten, et al. 2026. "Imaging of Molecular and Developmental Responses to Abiotic Stresses in Reproductive Tissues." *Physiologia Plantarum* 178: e70759. https://doi.org/10.1111/ppl.70759.

Gupta, R. 2026. "Introduction of 4D-Proteomics: A Breakthrough in Analyzing the Plant Proteomes During Stress Conditions." *Physiologia Plantarum* 178: e70735. https://doi.org/10.1111/ppl.70735.

Harzli, I., K. Mostafa, V. A. Mutlu, I. H. Shamsi, and M. Kavas. 2025. "Exploring Novel Microbial Approaches for Enhancing Crop Resilience to Abiotic Stress: Mechanisms and Applications." *Physiologia Plantarum* 177: e70582. https://doi.org/10.1111/ppl.70582.

Miller, G., M. Frencken, H. Sapeta, et al. 2026. "Methodologies and Considerations in Evaluating Heat Stress Response and Thermotolerance of Pollen Grains." *Physiologia Plantarum* 178: e70731. https://doi.org/10.1111/ppl.70731.

Muttappagol, M., M. Sharma, D. N. Adhikari, et al. 2025. "Towards Climate-Resilient Crops: Integrative Strategies for Reproductive Stage Abiotic Stress Tolerance." *Physiologia Plantarum* 177: e70635. https://doi.org/10.1111/ppl.70635.

Najar, M. A., S. Dey, and S. Banerjee. 2026. "Unlocking the Stress Responses of Sesame: An Oilseed Crop Through Omics Approaches Against Abiotic Stress." *Physiologia Plantarum* 178: e70869. https://doi.org/10.1111/ppl.70869.

Padilla, Y., V. Fotopoulos, G. Ntatsi, et al. 2025. "Strategies for Enhancing Resilience in Horticultural Crops Against Combined Abiotic Stresses." *Physiologia Plantarum* 177: e70502. https://doi.org/10.1111/ppl.70502.

Pasquali Medici de Biron, N. D., L. Kougiteas, S. Farrona, A. P. Santos, and S. Brunel-Muguet. 2026. "From Stress to Success: Harnessing Pre- and Post-Harvest Seed Priming." *Physiologia Plantarum* 178: e70718. https://doi.org/10.1111/ppl.70718.

RECROP COST. 2025. "Integrative Approaches to Enhance Reproductive Resilience of Crops for Climate-Proof Agriculture." *Plant Stress* 15: 100704. https://doi.org/10.1016/j.stress.2024.100704.

Sadras, B., P. R. Rao, P. Latha, B. G. S. Madhu, K. M. Vijay, and C. S. Lakshmi. 2026. "The Role of Mathematical Modeling in Plant Abiotic Stress Biology: Current Trends and Future Prospects." *Physiologia Plantarum* 178: e70909. https://doi.org/10.1111/ppl.70909.

Temel, A., and N. Gören-Sağlam. 2026. "The Role of Histone Modifications in Plant Priming and Their Analysis by Chromatin Immunoprecipitation." *Physiologia Plantarum* 178: e70915. https://doi.org/10.1111/ppl.70915.

Vraggalas, S., O. Guennich, B. Shalha, et al. 2025. "Methods for Analyzing Alternative Splicing and Its Regulation in Plants: From Gene-Specific Approaches to Transcriptome-Wide Studies." *Physiologia Plantarum* 177: e70639. https://doi.org/10.1111/ppl.70639.

13993054, 2026, 3, Downloaded from https://onlinelibrary.wiley.com/doi/10.1111/ppl.70921 by Readcube (Labtiva Inc.), Wiley Online Library on [01/06/2026]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License