THERMOTOLERANCE

DNA repair meets climate change

A direct link between DNA repair and heat tolerance has been revealed in *Arabidopsis thaliana*.

Annika Dorn and Holger Puchta

As global climate change threatens food security, heat tolerance in plants has become a central topic for researchers. It is known that rising temperatures affect crop growth and yield in multiple ways. However, a topic of essential importance, yet widely uncharted, is the maintenance of genome integrity after heat-stress-induced DNA damage[1].

In this issue of *Nature Plants*, Han et al.[2] unveiled a surprising and direct link between the transcriptional control of DNA repair genes and thermotolerance in plants. Already more than ten years ago, in a groundbreaking study, it was shown that the expression of DNA repair genes is upregulated by the plant-specific transcription factor SUPPRESSOR OF GAMMA RESPONSE 1 (SOG1) in the presence of DNA damage[3]. Now, the current study has demonstrated that there is crosstalk between heat stress and genotoxic stress in *Arabidopsis*.

In eukaryotes, central players for maintaining genome stability are RECQ helicases[4]. Although they are present as a multigene family in plant genomes[5], so far only RECQ4 homologues have been functionally well-characterized in vivo and demonstrated to be involved in DNA repair in somatic cells[6] as well as in crossover control in meiosis[7]. The AtRECQ2 helicase is homologous to the Werner protein, which has been shown to result in a severe genetic disease in humans when mutated. Although biochemical studies have demonstrated early on that its open reading frame codes for an active DNA helicase[8,9], surprisingly, no defect in DNA repair could be revealed by mutant analysis, until now[10]. The study of Han et al. has solved this mystery by demonstrating that RECQ2 does indeed have an important function in DNA repair in vivo; however, this function operates exclusively under heat stress. The starting point of the study from Han et al. was the analysis of a distinct thermosensitive phenotype of *Arabidopsis* mutants lacking the RING finger-containing E3 ubiquitin ligase HIGH EXPRESSION OF OSMOTICALLY RESPONSIVE GENES 1 (HOS1). These mutants depicted a lower survival rate than wild-type plants after heat treatment at 37 °C, and the increased leakage of electrolytes hinted to drastic cell damage. However, so far characterized roles of HOS1 as a ubiquitin ligase and suppressor of thermomorphogenesis via PHYTOCHROME INTERACTING FACTOR 4 (PIF4) suppression did not turn out to play a role in this heat stress endurance. Therefore, the authors performed a RNA sequencing analysis to elucidate how HOS1 might convey thermotolerance. By comparing the global expression levels of wild-type and *hos1* mutant plants grown under normal and high temperatures, they were able to not only define differences in global gene expression profiles but also to identify genes specifically regulated by HOS1 under heat stress. Surprisingly, a large number of DNA damage response genes did not show transcriptional upregulation in *hos1* mutants in response to high temperatures. And indeed, this astonishing link could be confirmed using the comet assay: DNA damage was shown to accumulate in the *hos1* mutants. These findings opened the door for a more detailed analysis of DNA repair under heat stress. The RECQ helicase RECQ2 came into focus in the study, as the *recq2* mutant depicted the highest reduction of thermotolerance of all tested mutants of DNA repair factors regulated by HOS1. In this mutant, an increased amount of DNA breaks at high temperatures was detected. Moreover, the involvement of HOS1 and RECQ2 in a common, so far unrevealed, repair pathway for heat-induced DNA damage could be demonstrated by the enhanced sensitivity of both mutants towards DNA crosslinking agents exclusively at high temperatures. Thus, for the first time a specific temperature-dependent DNA damage response could be documented in plants.

However, the central question of how heat stress signals are integrated into the transcriptional control of DNA repair genes by HOS1 remained. Therefore, the authors measured protein levels of HOS1 before and after heat exposure. Interestingly, the protein amount increased fivefold after heat exposure. This effect could be imitated at lower temperatures through the application of a proteasome inhibitor, resulting in a threefold induction. Apparently, HOS1 gets degraded by the proteasome at low temperatures, which is inhibited when the temperature rises, leading to a temperature-dependent activation of HOS1.

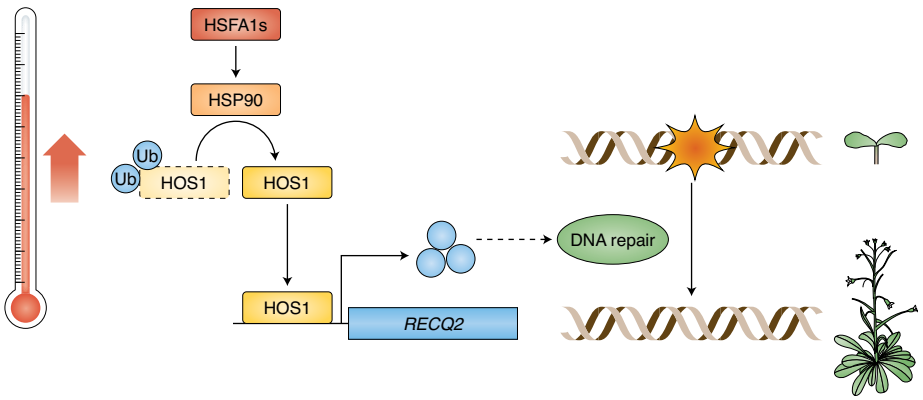

Fig. 1 | Model for the occurrence of thermotolerance by HOS1-mediated DNA damage response activation. At high temperatures, the HOS1 protein is protected from degradation by HSP90, which itself is controlled by the master regulator HSFA1. Growing amounts of HOS1 will induce the transcription of DNA repair genes, such as RECQ2. Heat-stress-induced DNA lesions can thus be repaired effectively, enhancing thermotolerance in *Arabidopsis*.

The final clue for solving the puzzle of thermal regulation came with the fact that this effect was diminished in the presence of HSP90 inhibitors and in *HSP90* RNA interference lines. A lack of the HSF1A master regulators, necessary for the heat-dependent accumulation of HSP90, resulted in a similarly reduced accumulation of HOS1 at high temperatures, accompanied by an increased number of DNA breaks. Therefore, the authors concluded that under heat stress, HSP90, regulated by its master regulator HSFA1, mediates the stabilization of HOS1, finally resulting in a transcriptional activation of DNA damage response factors (Fig. 1). That this kind of mechanism is of high importance has been shown by the demonstration that another DNA repair helicase, UV-HYPERSENSITIVE 6 (UVH6), is also part of the HOS1-dependent thermal regulation of DNA repair. In future research, it will be interesting to find out how many more DNA repair factors are regulated by this temperature-dependent repair mechanism. Naturally, a number of open questions remain. The exact mechanism of how HOS1 activates transcription and which possible co-activators might be involved is still unknown. Since the overexpression of *RECQ2* in the *hos1* mutant background did not completely complement thermotolerance, it directly implies the existence of further HOS1 targets. If and how RECQ2 and UVH6 cooperate in a common pathway, or if there are multiple sub-pathways for heat-induced DNA repair, remain to be elucidated. Moreover, in yeast two-hybrid assays and split-YFP analyses, no direct interaction between HSP90 and HOS1 could be detected; therefore, additional supporting factors, yet to be identified, have to be involved in the thermostabilization of HOS1. An intriguing open question involves the interrelation between the HOS1- and SOG1-dependent pathways of DNA damage response, and whether the SOG1 pathway itself is influenced by temperature.

Thus, the authors were not only able to uncover a yet uncharted phenomenon, they also opened a new door for further research. The new avenue might enable not only the identification of factors that are specifically required for the repair of heat-induced DNA damage, but we may also learn more about the interlink between different stress responses in plants. Most importantly, in the long run, following this path might enable us to engineer crops that survive better in a world continuously heating up due to global warming. ❐

Annika Dorn and Holger Puchta
Botanical Institute—Molecular Biology and Biochemistry, Karlsruhe Institute of Technology, Karlsruhe, Germany.
✉*e-mail: holger.puchta@kit.edu*

Published online: 16 November 2020
https://doi.org/10.1038/s41477-020-00804-x

References

1. Kantidze, O. L., Velichko, A. K., Luzhin, A. V. & Razin, S. V. *Acta Naturae* **8**, 75–78 (2016).
2. Han, S.-H., Park, Y.-J. & Park, C.-M. *Nat. Plants* https://doi.org/10.1038/s41477-020-00809-6 (2020).
3. Yoshiyama, K., Conklin, P. A., Huefner, N. D. & Britt, A. B. *Proc. Natl Acad. Sci. USA* **106**, 12843–12848 (2009).
4. Dorn, A. & Puchta, H. *Genes* **10**, 1028 (2019).
5. Hartung, F., Plchová, H. & Puchta, H. *Nucleic Acids Res.* **28**, 4275–4282 (2000).
6. Hartung, F., Suer, S. & Puchta, H. *Proc. Natl Acad. Sci. USA* **104**, 18836–18841 (2007).
7. Mieulet, D. et al. *Nat. Plants* **4**, 1010–1016 (2018).
8. Kobbe, D., Blanck, S., Demand, K., Focke, M. & Puchta, H. *Plant J.* **55**, 397–405 (2008).
9. Klaue, D. et al. *Nat. Commun.* **4**, 2024 (2013).
10. Röhrig, S. et al. *New Phytol.* **218**, 1478–1490 (2018).

Author contributions

A.D. and H.P. wrote the manuscript.

Competing interests

The authors declare no competing interests.