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Evaluation of alterations in physio-biochemical attributes and antioxidant defense-system in heat-stressed okra (*Abelmoschus esculentus*) cultivars and its putative use as heat tolerance indicators

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The current investigation was carried out to assess, whether the alterations in the activities of antioxidant enzymes and the levels of some non-enzymatic antioxidants, could be used as potential indicators of heat-tolerance in 10 commercially important cultivars of okra (*Abelmoschus esculentus* L.). The tested cultivars were exposed to 3 different temperatures i.e., 25, 40 and 45°C under controlled conditions in growth chambers. Various growth, physiological and biochemical characters in leaf or/and root tissues were analyzed. According to the percent inhibition in shoot/root, fresh and dry biomass at the highest temperature (45°C) cultivars Sabaz Pari, Desi Okra and Green Wonder were ranked as tolerant (percent inhibition less than 60%), Shahzadi, Anarkali, Sarsabaz and Rama Krishna moderately tolerant (percent inhibition 60–70%), Click-5759 MF-03 and Pen Beauty are heat sensitive (percent inhibition more than 70%). Heat stress caused significant reductions in photosynthesis associated attributes but did not affect the number of stomata. High temperature-stress significantly elevated the activities of antioxidants, levels of osmolytes and decreased the MSI, and activities of GPX and CAT, while the internal levels of H₂O₂ remained unaffected in all tested okra cultivars. Although heat-induced oxidative stress was observed in all okra cultivars, but the response of heat-resistant and heat-sensitive cultivars with respect to the formation of enzymatic and non-enzymatic metabolites measured in the present study was not consistent. Of various gas exchange characteristics, antioxidant-enzymes and metabolites measured, only PN, MSI and activities of GPX and CAT activity was found to be a consistent indicators of heat-tolerance in tested okra cultivars.

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Dissecting the molecular mechanism of glutathione-ethylene cross-talk in plants during stress

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Plant defense is regulated by a network of signaling pathways where salicylate, jasmonate and ethylene (ET) function as key signaling molecules. Glutathione (GSH) is gradually gaining importance as a dynamic player in this network, playing critical roles during stress. We have earlier reported that enhanced GSH level can provide resistance against *Botrytis cinerea* infection presumably through its crosstalk with ET. In this study, we demonstrate that GSH induces ET biosynthesis by modulating the transcriptional and post-transcriptional regulations of its key enzymes, ACC synthase (ACS) and ACC oxidase (ACO). Transgenic *Arabidopsis* plants with enhanced GSH content exhibited remarkable up-regulation of ACS2, ACS6 and ACO1 at transcript as well as protein levels, while they were down-regulated in the GSH depleted pad2-1 mutant. We further observed that GSH induced ACS2 and ACS6 transcription in a WRKY33 dependent manner while ACO1 transcription remained unaffected. On the other hand, the mRNA stability for ACO1 was found to be increased by GSH which explains our above observations. In addition, we also identified the ACO1 protein to be a subject for S-glutathionylation which is consistent. However, S-glutathionylation of ACS2 and ACS6 proteins was not detected. Further, the transgenic plants exhibited resistance to necrotrophic infection and salt stress while the pad2-1 mutant was sensitive. Exogenous GSH improved stress tolerance in wild-type plants but not in the ET signaling mutant, ein2-1, indicating that GSH mediated resistance to these stresses occurs via an ET mediated pathway. Together, our investigation reveals a dual-level regulation of ET biosynthesis by GSH during stress.

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