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Nitric Oxide-induced *AtWRKY6* Positively Regulates Plant Growth and Defense Response

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[Introduction]

WRKY transcription factors (TFs) are plant-specific genes characterized by one or two highly conserved WRKY domain(s) followed by zinc-finger motif. WRKYs regulate a number of processes from growth to defense and are therefore called, "Jack of various trades".

[Materials and Methods]

In the current study by using high throughput RNA-seq data, we have characterized a number of WRKY TFs that have shown response to nitric oxide (NO) donor *S*-Nitrosocysteine (CySNO). The orthologs of these differentially expressed genes (DEGs) were also found in other species especially in crops. Comparison of protein sequences from NO-responsive and non-responsive WRKYs showed a conserved Cysteine or Tyrosine residue in NO-responsive WRKYs which are potential targets for protein *S*-nitrosylation, the addition of NO moiety to the active Cysteine residues. Functional characterization using reverse genetics approach further confirmed the regulatory role of WRKYs in plant growth and defense.

[Results and discussion]

AtWRKY6 that showed up-regulation in CySNO-induced transcriptome (fold change 27.003) regulates Phosphate1 expression in response to low P stress; however, its role in plant defense is yet to be known. We exposed wild type (WT), *atwrky6* (KO), *AtWRKY6* (OX) and *atgsnor1-3* plants to oxidative and nitrosative stress media. Our results suggested that under control conditions, *atwrky6* showed similar shoot and root length while *AtWRKY6* showed increased shoot and root length compared to WT whereas under nitrosative stress condition *atwrky6* showed decrease while At*WRKY6* showed increased shoot and root length compared to WT. Exposure of both at*wrky6* and At*WRKY6* plants to virulent pathogen *Pseudomonas syringae* pv *tomato* (*Pst*) strain DC3000 showed decreased pathogen growth for *atwrky6* while increased pathogen growth for *AtWRKY6 OX* plants suggesting positive regulation of basal defense by *AtWRKY6*. We also inoculated plants to avirulent strain of *Pst* DC3000 to see their role in *R*-gene mediated resistance. We found that *atwrky6* showed increased hypersensitive response (HR) while almost no HR response was observed for *AtWRKY6* plants compared to WT. Together all, these results suggest a positive role of *AtWRKY6* in plant defense and growth.

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