

## ABSTRACT

A plethora of gene regulatory mechanisms with eccentric attributes in *Deinococcus radiodurans* confer it to possess a distinctive ability to survive under ionizing radiation. Among the many regulatory processes, small RNA (sRNA)-mediated regulation of gene expression is prevalent in bacteria but barely investigated in *D. radiodurans*. In the current study, we identified a novel sRNA, DrsS, through RNA-seq analysis in *D. radiodurans* cells while exposed to ionizing radiation. Initial sequence analysis for promoter identification revealed that *drsS* is potentially co-transcribed with *sodA* and *dr\_1280* from a single operon. Elimination of the *drsS* allele in *D. radiodurans* chromosome resulted in an impaired growth phenotype under  $\gamma$ -radiation. DrsS has also been found to be upregulated under oxidative and genotoxic stresses. Deletion of the *drsS* gene resulted in the depletion of intracellular concentration of both  $Mn^{2+}$  and  $Fe^{2+}$  by  $\sim 70\%$  and  $40\%$ , respectively, with a concomitant increase in carbonylation of intracellular protein. Complementation of *drsS* gene in  $\Delta drsS$  cells helped revert its intracellular  $Mn^{2+}$  and  $Fe^{2+}$  concentration and alleviated carbonylation of intracellular proteins. Cells with deleted *drsS* gene exhibited higher sensitivity to oxidative stress than wild-type cells. Extrachromosomally expressed *drsS* in  $\Delta drsS$  cells retrieved its oxidative stress resistance properties by catalase-mediated detoxification of reactive oxygen species (ROS). *In vitro* binding assays indicated that DrsS directly interacts with the coding region of the *katA* transcript, thus possibly protecting it from cellular endonucleases *in vivo*. This study identified a novel small RNA DrsS and investigated its function under oxidative stress in *D. radiodurans*.

## IMPORTANCE

*Deinococcus radiodurans* possesses an idiosyncratic quality to survive under extreme ionizing radiation and, thus, has evolved with diverse mechanisms which promote the mending of intracellular damages caused by ionizing radiation. As sRNAs play a pivotal role in modulating gene expression to adapt to altered conditions and have been delineated to participate in almost all physiological processes, understanding the regulatory mechanism of sRNAs will unearth many pathways that lead to radioresistance in *D. radiodurans*. In that direction, DrsS has been identified to be a  $\gamma$ -radiation-induced sRNA, which is also induced by oxidative and genotoxic stresses. DrsS appeared to activate catalase under oxidative stress and detoxify intracellular ROS. This sRNA has also been shown to balance intracellular Mn(II) and Fe concentrations protecting intracellular proteins from carbonylation. This novel mechanism of DrsS identified in *D. radiodurans* adds substantially to our knowledge of how this bacterium exploits sRNA for its survival under stresses.