Characterization of the role of the RadS/RadR two-component system in the radiation resistance of *Deinococcus radiodurans*

By: Shruti S. Desai, Yogendra S. Rajpuroid, Hari S. Misra and Dileep N. Deobagkar

SUPPLEMENTARY FIGURE LEGENDS

**Supplementary Fig. S1.** Generation of *radS* and *radR* deletion mutants of *D. radiodurans* R1. Genomic DNA of prospective *radS* (radS) and *radR* (radR) mutant cells was prepared and the presence of the *radS/radR* and *nptII* genes was ascertained by PCR amplification using gene-specific internal primers for the DR_B0090 and DR_B0091 ORFs (a) and *nptII*-specific primers (b). The amplification of the *radS/radR* genes was confirmed in the chromosomal DNA of *D. radiodurans* (R1). Similarly, *nptII* amplification was ascertained with both the mutant genome and the plasmid constructs (P) used for generating these mutants. A *HindIII* digest of lambda DNA was used as size marker (M).

**Supplementary Fig. S2.** Pepetide mass fingerprints of polypeptide P1 as shown in Fig. 1. The protein was cut from the polyacrylamide gel and digested with trypsin. The tryptic fragments were analysed by MALDI-TOF MS and various peptides were scanned as per their *m/z* ratio. The data were used for identification of the protein from the *Deinococcus* proteome using MASCOT software.

**Supplementary Fig. S3.** Diagrammatic representation of functional domains in the RadS and RadR proteins. The P1 protein, showing inducible synthesis at 15 min PIR, as shown in Fig. 1, was identified by MS (Supplementary Fig. S2) as a response regulator encoded by DR_B0091 (RadR) and located downstream of the histidine kinase encoded by DR_B0090 (RadS). Amino acid sequences of RadS and RadR were searched for functional domains in [www.expasy.ch/prosite](http://www.expasy.ch/prosite). RadS showed the presence of phosphorylation sites (P-site) and a dimer interface in the HisKA domain, and ATP-binding sites (ABS) and a G-X-G motif in the HATPase C domain. RadR showed the presence of a CheY-type receiver domain at the N terminus and a transcription regulation domain (Trans reg-C) for transcription factors at the C terminus.