Supporting Information

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SI Methods

mRNA Expression. The following primers for the genes encoding nitric oxide synthase (DR2597) and glyceraldehyde 3-phosphate dehydrogenase (DR1343): nosF 5′ctgctccagcatctcgacgacgcct 3′, nosR 5′ atgtccaggtgcatgtcgctgatg 3′, gapF 5′ gagtacgacgaaagcagcctgacg 3′ and gapR 5′ cgttggtgtagctgtgcacggtggtc3′. Controls were carried out without addition of reverse transcriptase and with RNase treatment before PCR to confirm that amplified bands were not due to contamination by genomic sequences. The PCR (50 μL) was carried out using the following cycling conditions: 98 °C for 5 min, 37 cycles of 95 °C for 1 min, 52 °C for 30 s, and 72 °C for 30 s. Samples were then electrophoresed on a 1% agarose gel, and stained with ethidium bromide. *nos* message levels were compared to that of *gap*, whose expression is not altered by ultraviolet (UV) radiation. PCRs were confirmed to be within the linear range of amplification by varying amplification cycles.

The forward and reverse primers in the 5′ →3′ order for other genes are listed below:

obgE fp: ggtcgaggaattacaacagctt
obgE rp: aactcgggcaacaactggaata
recA fp: gcaaactcgacgtgcaggtcgtcagca
recA rp: gtgctcggcgtcgataaacgcacag
uvrA fp: tcacgcaggttcttcagcgtgccgat
uvrA rp: ggctgaagtttctggtggacgt
uvsE fp: gctgtgctcacgcatctagcgccgca
uvsE rp: agcagattccagggcgtgcgcgccag

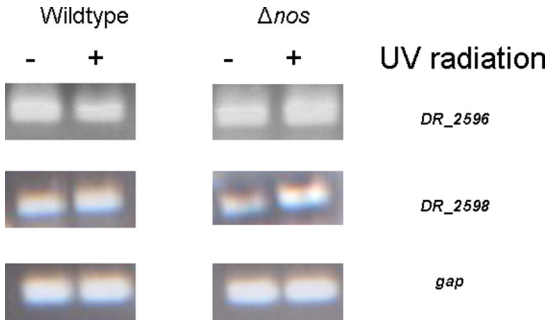

Fig. S1. The genes flanking DR_2597 (*nos*): DR_2596 and DR_2598 code for proteins of unknown function. Their expression patterns remain unchanged in the Δ*nos* compared to the wild-type strain under basal and postirradiation conditions. DR_2596 is unaffected by radiation but DR_2598 appears to be upregulated. This is not surprising as the ORFs for *nos* and *DR_2598* are expressed on the same mRNA.

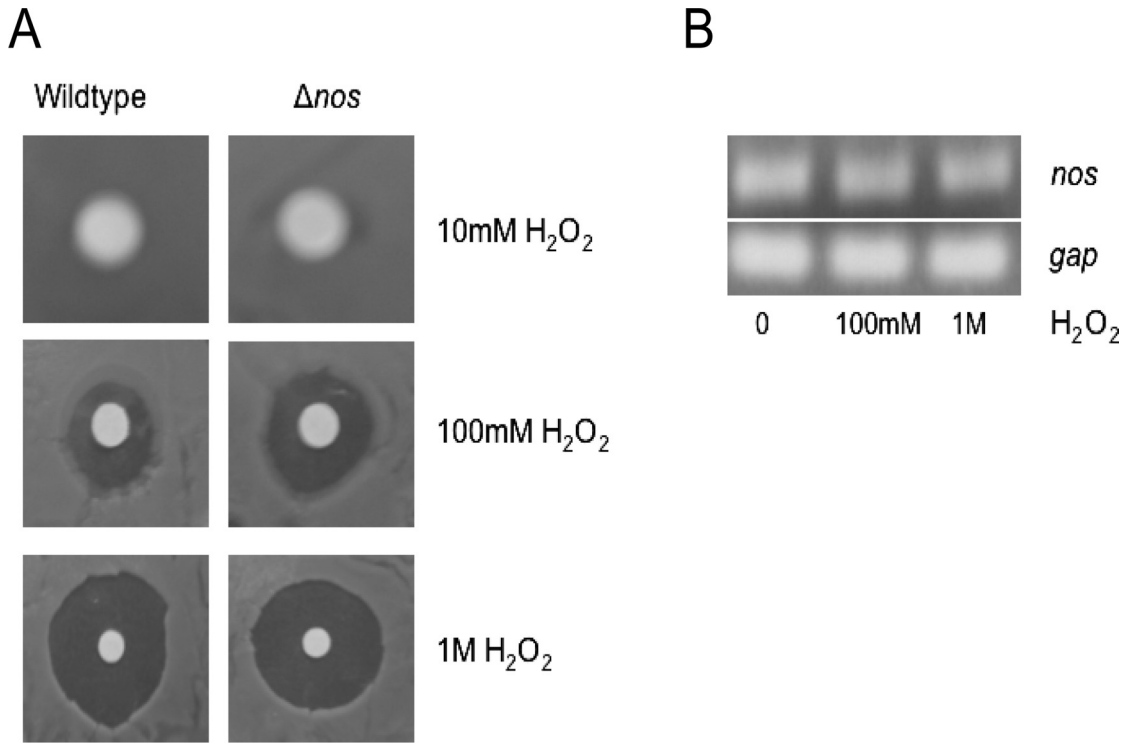

Fig. S2. (*A*) Δ*nos* does not harbor a growth defect in the presence of peroxide. Wild-type and Δ*nos* were grown to OD_{600} ~1 at 30 °C. The cells were then plated on TGY agar plates, and a filter disk, saturated in the appropriate concentration of H_2O_2, was placed on top of the lawn. The plates were allowed to grow for 2 days at 30 °C. Δ*nos* (right panel) is not affected by any concentration of H_2O_2 compared to the wild-type (left panel), ruling against *nos* playing a protective a role against peroxide in Dr. The same result is obtained if cells are grown in the presence of H_2O_2 in liquid cultures. Dr shows high tolerance to peroxide, as cell growth is unaffected by 10 mM H_2O_2. (*B*) Additionally, the *nos* mRNA levels remain unchanged after exposure to as much as 1M H_2O_2 for 1 h (more DNA was loaded at concentrations comparable to the UV irradiation experiment in Fig. 3 to clearly show the presence of the *nos* gene, which is not highly expressed under these conditions).

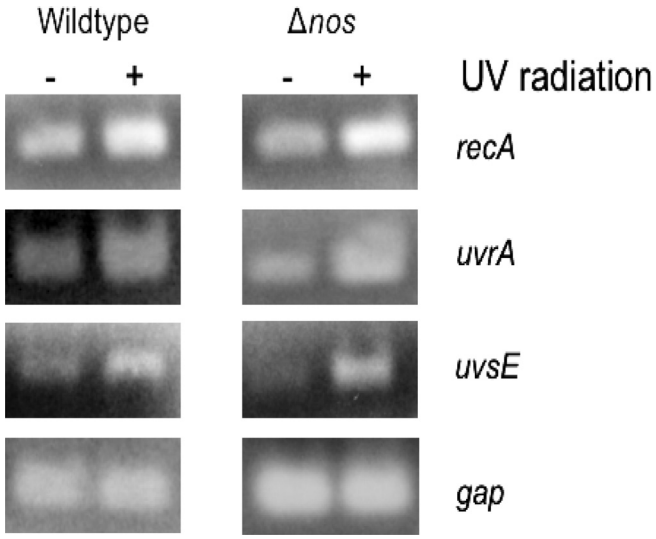

Fig. S3. NOS does not affect the induction of key UV DNA repair genes. mRNA expression levels were evaluated for *recA*, *uvrA*, and *uvsE* with the same procedure used to measure *nos* mRNA levels. In the wild-type, the mRNA levels of these genes increase after exposure to radiation. However, Δ*nos* also shows a similar increase in mRNA levels. Thus, *nos* does not appear to be required for the induction of these repair genes.

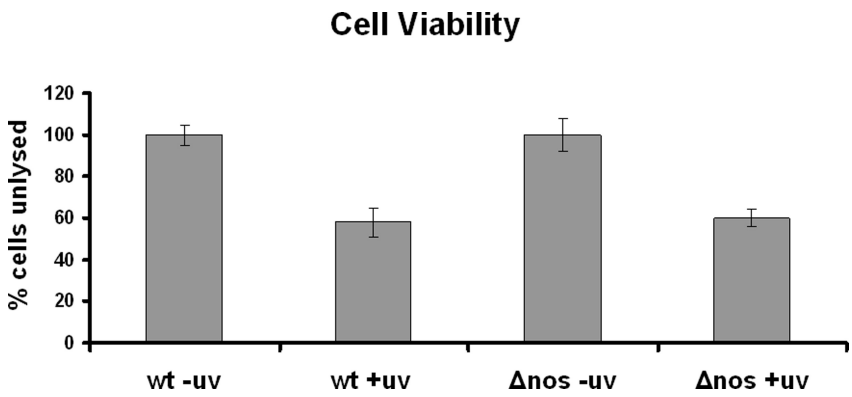

Fig. S4. The percent of input CFU viable following irradiation of wild-type and Δ*nos* is the same using the LIVE/DEAD BacLight Bacterial Viability Kit (Molecular Probes). The assay has two fluorescent probes which can distinguish cells with intact membranes from those with damaged membranes. The experiment was carried out following the manufacturer's protocol.

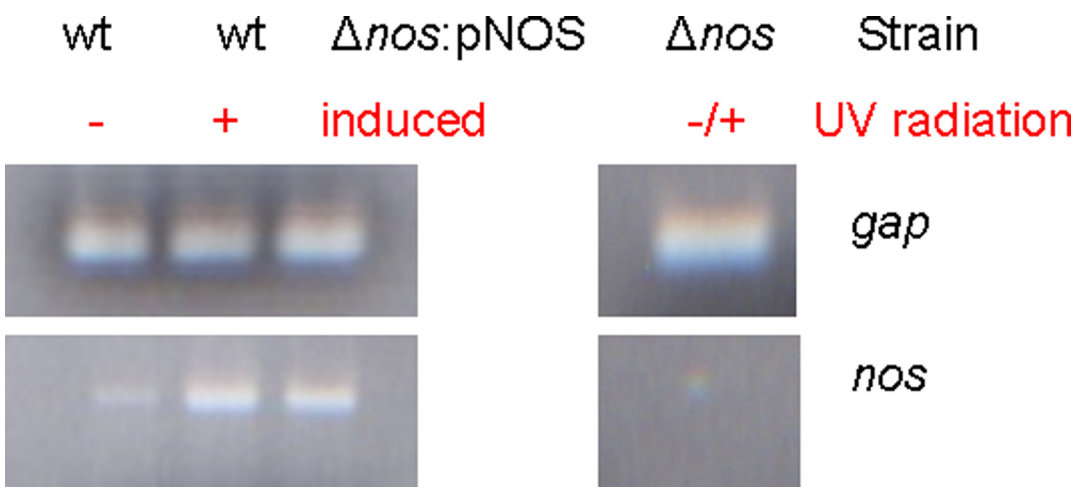

Fig. S5. *Nos* gene expression in the complemented strain. mRNA was extracted and converted to cDNA using protocols described in *Methods*. The expression levels of *nos* from the complementation (Δ*nos* :pNOS) is higher than the levels of wild-type without UV exposure; however, the levels are lower than wild-type with UV exposure. Δ*nos* does not show any *nos* mRNA.

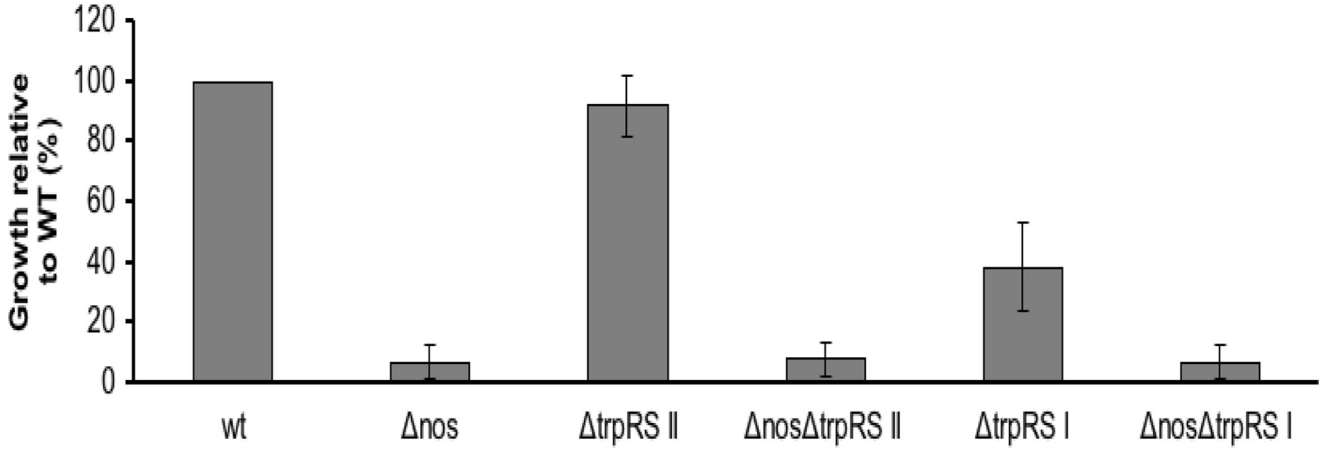

Fig. S6. Gene deletion of *trpRS* I and *trpRS* II. The Δ*nos* strains show the largest growth defects following UV irradiation. Δ*trpRS* II is unaffected by UV, while Δ*trpRS* I grows 40% to that of wild-type because TrpRS I is the primary TrpRS in Dr. Both double knockouts Δ*nos*Δ*trpRS* II and Δ*nos*Δ*trpRS* I show growth patterns similar to the single mutant Δ*nos*. Notably, growth of the Δ*trpRS* I and Δ*trpRS* II strains are not differentiated in the absence of UV exposure. Thus, TrpRS I provides an advantage only under stress conditions. The data represents an average of three independent experiments ± SD.

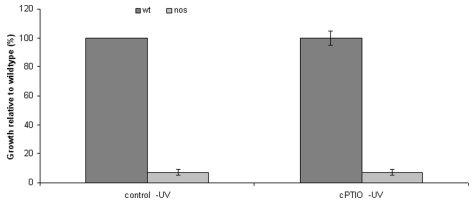

Fig. S7. Effects of secondary products on UV recovery. Both wt (*dark gray bars*) and Δ*nos* (*light gray bars*) cells were grown for 22 h with pre-incubation (prt) of 10 μM L-citrulline, 1 mM ferrous cyanide, and 100 μM spermine after 5 min of UV radiation. Control cells were grown in the absence of supplemented chemicals. The relative growth of wt cells exposed to UV radiation was given a value of 100. The same protocol as in the main paper was followed for standardizing growth conditions. There was no change in the growth of the strains in the presence of these compounds without UV irradiation.

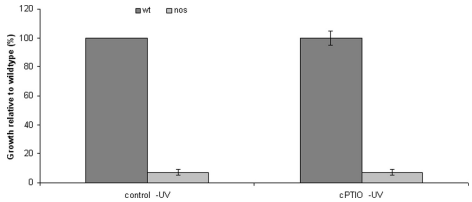

Fig. S8. 100 μM cPTIO [2-(4-carboxyphenyl)-4,4,5,5-tetramethylimidazoline-1-oxyl-3-oxide] was added to wt and Δ*nos* for 10 min and has no effect on either WT or Δ*nos* without UV irradiation. The same protocol for assaying growth with NO donor compounds and other additives was followed here.

Condition Tested	Reactive Oxygen Species (ROS) Detected
PBS + UV then NOHA	-
PBS + NOHA + UV	+
Anaerobic PBS + NOHA + UV	-
Dr cells + UV then NOHA	-
Dr cells + NOHA – UV	-
Dr cells + NOHA + UV	+

Fig. S9. Presence of reactive oxygen species (ROS) during and following UV irradiation. ROS generated during irradiation reacts with added N-hydroxyl-L-Arginine (NOHA) and lead to formation of nitrite, which can be detected by the Griess Assay. NOHA in anaerobic phosphate buffered saline (PBS) does not yield any ROS on irradiation as detected by this method, but aerobic PBS gives a strong nitrite signal. Likewise, if NOHA is added to cells and irradiated, large amounts of nitrite are detected. However, if NOHA is added, even seconds after irradiation is ceased, no nitrite production is observed. Thus, ROS are rapidly depleted after exposure ends.

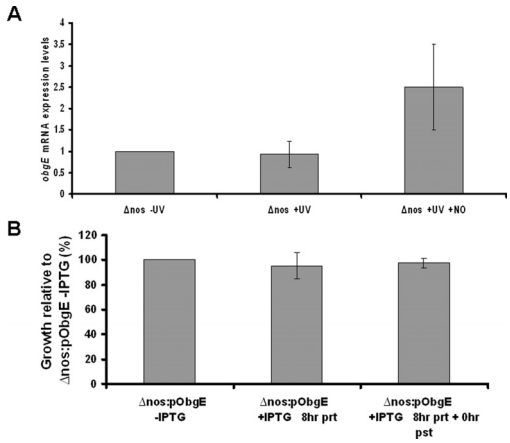

Fig. S10. (*A*) The mRNA levels of the *obgE* gene from Δ*nos* cells without UV irradiation, after irradiation, and with irradiation on the addition of exogenous NO were measured using a similar protocol to that used for detecting expression of the *nos* gene. The level of *obgE* increases in the irradiated Δ*nos* cells upon treatment with NO, although not to the same extent as that observed in the wt with UV irradiation (Fig. 5*A*). (*B*) The growth of Δ*nos*:pObgE is unaffected upon induction of the *obgE* gene under normal growth conditions (no UV irradiation).